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Pitout JDD, Church DL, Gregson DB, Chow BL, McCracken M, Mulvey M, Laupland KB (2007). Molecular epidemiology of CTXM-producing Escherichia coli in the Calgary Health Region: emergence of CTX-M-15-producing isolates. Antimicrob. Agents Chemother. 51: 1281-1286.

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Vol. 10(10), pp. 189-202, October 2016

DOI: 10.5897/AJPS2016.1426 Article Number: FD3BBA161075

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Full Length Research Paper

# Genotype by environment interaction in sesame (Sesamum indicum L.) cultivars in Uganda

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Received 10 May, 2016; Accepted 4 August, 2016

Sesame (Sesamum indicum L.) is an important and ancient oilseed crop cultivated in hot, dry climates for its oil and protein rich seeds. On the African continent, Uganda ranks seventh in sesame production. The improvement of new genotypes with the desired yield stability and performance in different environments is an important issue in breeding programs. In order to identify high yielding and stable sesame genotypes across environments, field experiments were conducted with 16 genotypes for four seasons (2011-2013) at three locations, viz. Serere, Kaberamaido and Ngetta. The objective of the study was to use additive main effects and multiplicative interaction (AMMI) and genotype by genotype environment interaction (GGE) biplot statistical analysis to identify the stability and yield potential of sixteen sesame genotypes. The results of AMMI analysis of variance for seed yield (kg/ha) showed that all the sources of variations that included treatments, genotypes, environments, blocks, interactions, IPCA1 and IPCA 2 were highly significant (P<0.001). The combined analysis of variance indicated that season, season x location, genotype and location x genotype had highly significant (P<0.001) variation. The GGE biplot suggested the existence of only one sesame mega-environment with genotype G9 (Local 158-1) best adapted in that mega-environment followed by G1 (Ajimo A1-6//7029)-1-1. The mega-environment had environments K2011B, K2012A, K2012B, N2012B and K2013B. The vertex genotypes which indicated that they were the most responsive in their respective environments were G2 (Ajimo A1-6//7029)-1-9, G3 (Local 158//6022)-1-2-1, G8 (EM15-3-2), G9 (Local 158-1), G12 (Renner 1-3-1-16) and G14 (Renner 1-3-1-17-1). Genotypes G2 and G12 performed poorly in poor environments. Genotypes were categorized into stable and high yielding, stable but poor yielding, unstable but good yielding and unstable and poor yielding. Environment K2013B was the most discriminating environment. According to the ideal-genotype biplot, genotype G9 (Local 158-1) was the best performing genotype and Kaberamaido was the nearest to ideal environment. It was officially released as Sesim 3 variety for commercial production because of its yield, stability, tolerance to pests and high oil content.

**Key words:** Adaptation, additive main effects and multiplicative interaction (AMMI), genotype environment interaction (GGE) biplot, principal component analysis, stability.

#### INTRODUCTION

Sesame (Sesamum indicumL.) belongs to the Pedaliaceae (order Lamiales) with a small family of 15 genera and 70 species characterised by annual and perennial growth forms. Sesame is an important and

ancient oilseed crop cultivated in hot, dry climates for its oil and protein-rich seeds (Bedigian and Harlan, 1986). Currently, sesame is grown throughout the tropical and subtropical regions of the world with Sudan, China, India,

and Myanmar being the top producers in 2014, together covering 46% of the world production (FAOSTAT, 2015). On the African continent, Uganda (124,300 t) ranks seventh in sesame production (FAOSTAT, 2015). Sesame is commonly known as simsim in Uganda and East Africa generally.

It is often referred to by the epithet "the queen of oil seeds" because of its nutritive value, quality and quantity of its oil which is rich in vitamin E and has a significant amount of linoleic acid that can control blood cholestrol levels (Vijayarajan et al., 2007).

It is an important annual oilseed crop in the tropics and warm sub-tropics where it is mainly grown in small plots as source of edible oil and one of the ingredients in food products. The seed is also rich in protein, vitamins including minerals and lignans such as sesamolin and sesamin (Moazzami and Kamal-Eldin, 2006). Sesame oil has medicinal and pharmaceutical value and is being used in many health care products (Coulman et al., 2005). The seed contains 50 to 60% oil and 25% protein with antioxidants lignans such as sesamolin, sesamol, sesamin which impart to it a high degree of resistance against oxidative rancidity and gives it a long shelf life (Ashri, 1989). It has been used as an active ingredient in antiseptics, bactericides, viricides, disinfectants, moth repellants and anti-tubercular agents (Bedigian et al., 1985). It is a source of calcium, tryptophan, methionine and many minerals (Johnson et al., 1979).

Plant variety trials are routinely conducted to compare multiple genotypes in multiple environments (years and locations) for multiple traits, resulting in genotype by environment by trait three-way data (Yan and Tinker, 2006). Variety trials provide essential information for selecting and recommending crop cultivars. However, variety trial data are rarely utilized to their full capacity. Furthermore, analysis of genotype by environment data is often limited to genotype evaluation based on genotype effect (G) while genotype-by-environment main interactions (GE) are treated as noise or a confounding factor. A larger GEI variation usually hinders the accuracy of yield estimation and reduces the correlation between genotypic and phenotypic values.

According to Yan and Kang (2003), it is known that mean yield across environments are sufficient indicator of genotype performance only in the absence of genotype by environment interaction. Most of the time, GEI complicates breeding, testing and selection of superior genotypes. It is important for breeders developing new varieties to identify specific genotypes adapted or stable to different environment(s), thereby achieving quick genetic gain through screening of genotypes for high adaptation and stability under varying environmental conditions prior to their release as cultivars.

In variety selection experimentation, many genotypes are normally tested over a wide range of environments. Plant breeders perform multi-environmental trials (MET) to evaluate new improved genotypes across test environments before a specific genotype is released for production (Rahmatollah et al., 2013). In such experiments, genotype x environment (GE) interaction is commonly evaluated (Yan et al., 2007). GE interaction effects are of special interest for identifying the most stable genotypes, mega-environments and adaptation targets in most plant breeding programs (Sabaghnia et al., 2013).

Some parameters could be used to study the stability such as regression slope (bi), equivalency (Wi2), coeefficient of determination (Ri2), Si2 ), analysis of variance (ANOVA), principal component analysis (PCA), additive main effects and multiplicative interaction (AMMI) and genotype genotype x envionment (GGE) biplot. Nevertheless, visualization of the test would be much helpful in concluding the results (Susanto et al., 2015). GGE biplot analysis is one appropriate tool to evaluate representation of an environment, genotype stability, and the effect of G x E to the peformance of a genotype (Rahmatollah et al., 2013). GGE biplot analysis provides an easy and comprehensive solution to genotype by environment data analysis, which has been a challenge to plant breeders, geneticists, and agronomists. GGE biplot analysis is a statistical method which used multivariate approach in the analysis. It is better than univariate approach in dissecting GxE components into specific interaction between genotype and environmental components (Flores et al., 1998). GGE biplot is able to show the best genotype with the highest yield in a quadran containing identical locations environments), genotype average performance and stability, ideal genotype and ideal location to increase yield, and specific location (Farshadfar and Sadeghi, 2014). In the AMMI biplot, each genotype is represented by a linear line defined by the genotype's mean yield and its interaction principal components axis (IPCA) score on the y-axis and mean yield on the x-axis.

Multivariate techniques like the additive main effects and multiplicative interaction (AMMI) procedure with prediction assessment can be a powerful tool in analyzing multilocation trials (Gauch and Zobel, 1988). The AMMI model combines regular analysis of variance (ANOVA) for additive effects with Principal Component Analysis (PCA) for multiplicative structure within the interaction. A modification of the conventional AMMI analysis proposed by Yan et al. (2000) called GGE biplot (genotype and Genotype-Environment Interaction) has been used to study the GE interaction. The GGE analysis groups together the GE interaction multiplication effect

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and the genotype effect, which is an additive effect in the AMMI analysis, and analyses these effects by principal components.

Understanding the structure and nature of genotype x environment interaction (GEI) is important in plant breeding programs because a significant GEI can seriously impair efforts in selecting superior genotypes relative to new crop introductions and cultivar development programs. Information on the structure and nature of GEI is particularly useful to breeders because it can help determine if there is need to develop cultivars for all environments of interest or if the need is to develop specific cultivars for specific target environments. In Uganda, work on sesame research for G x E has basically been on the use of ANOVA. This is the first time detailed statistical analysis using multivariate techniques like AMMI and GGE has been applied.

The objective of this study was to use multivariate statistical methods of the GGE biplot and AMMI methodologies to identify the stability and adaptability of sixteen sesame genotypes.

#### **MATERIALS AND METHODS**

#### Experimental sites and planting materials

Sixteen genotypes including the local selected breeding lines and introduced accessions (Table 1) selected for better yield potential and other yield characters among genotypes at National Semi-Arid Resources Research Institute (NaSARRI), Serere were evaluated in three locations of Serere (latitude 1°31'N; longitude 33°27'E; and altitude 1,139 mean above sea level (masl), Kaberamaido (latitude 1°44'N; longitude 33°09'E; altitude 1,080 masl) and Ngetta (latitude 2°17'N; longitude 32°56'E; altitude 1,189 masl) for four seasons. These locations are situated in Serere, Kaberamaido and Lira districts representing different farming systems and different tribal setting. The evaluation experiments were done in 2011B, 2012A, 2012B and 2013B seasons. The first rainy season planting starts about mid-March to April and harvested in July, while the second season's planting starts about July to September and harvested about October to December depending on the location and onset of rains. The farmers in Kaberamaido district used to grow their sesame during the first rainy season which starts about mid-March. Serere farmers plant sesame in both seasons. Meanwhile, in Lira district where Ngetta is located, sesame was mainly grown during the second season although in the southern part of the district, sesame is planted during the first season. With changes in weather pattern and increasing market, farmers across locations are now growing sesame twice in a year.

#### **Procedure**

A randomized complete block design with 3 replications was used across locations and seasons. Each entry was planted in eight 4-m rows, with spacing of 30 cm between rows and continuous sowing within a row. Thinning within the rows was done to approximately 10 cm between plants. Standard recommended practices like insecticides and fungicides were applied where necessary in order to control insect pests and fungal diseases respectively in order to produce good results. The six middle rows were harvested and used as yield per plot and converted to yield in kilograms per hectare.

#### Statistical analysis

The the data for grain yield were pooled to perform the analysis of variance across locations. Since the pooled analysis of variance considers only the main effects, the additive main effect and multiplicative interaction model (AMMI) was computed. Beginning with the ordinary ANOVA procedure for two way analysis of variance, the AMMI analysis first separates additive variance from the multiplicative variance (interaction), and then applied PCA to the interaction, that is, to the residual portion of the ANOVA model to extract a new set of coordinate axes which accounts more effectively for the interaction patterns (Gauch and Zobel, 1988). AMMI analysis was also used to determine stability of the genotypes across locations using the PCA (principal component axis) scores.

To graphically visualize the relationship between environments (locations by seasons) and entries in order to determine the 'which won where' portion, and to identify mega environment, a GGE biplot (Yan, 2001) analyis was also undertaken using GGE biplot in the Meta analysis of Genstat 14<sup>th</sup> edition (Payne et al., 2010).

Analysis of variance (ANOVA) for yield was carried out for individual locations, seasons and for combined analysis across locations. Statistical procedures applied were Principal Component Analysis (PCA), additive main and multiplicative interaction (AMMI), genotype and genotype by environment biplot analysis (GGE). For a simple analysis of variance of a randomized complete block design, the model:  $Y_{ijk} = \mu + G_i + E_j + GE_{ij} + B_{ij} + \epsilon_{ijk}$  was applied where  $\mu$  is the mean,  $G_i$  is the effect of the  $i^{th}$  genotype,  $E_j$  is the effect of the  $j^{th}$  environment,  $GE_{ij}$  is the interaction of the  $i^{th}$  replication in the  $j^{th}$  environment, and  $\epsilon_{ijk}$  is the random error.

Genotype-focused scaling was used in visualizing for genotypic comparison, with environment-focused scaling for environmental comparison. The symmetric scaling was preferred in visualizing the "which-won-where" pattern of multi-environment yield trials (MEYTs) data (Yan, 2002). The biplots were constructed using GenStat 14<sup>th</sup> Edition (Payne et al., 2010).

AMMI and GGE procedures for estimating G x E interaction were used. The combined analysis was performed and the means served as basis for the AMMI analysis, considering the following model:  $Y_{ij}$ 

=  $\mu$  +  $G_i$  +  $E_j$  +  $\sum_{i=1}^{\sum} \lambda_k \gamma_{ik} \alpha_{jk}$  +  $\rho_{ij}$  +  $\epsilon_{ij}$ , Where:  $Y_{ij}$  is the mean response of genotype i in environment j;  $\mu$  is the overall mean;  $G_i$  is the genotype effect;  $E_j$  is the environment effect;  $GE_{ij}$  is the

A GGE biplot was constructed by plotting the first principal components against their respective scores for the second principal component (PC2) that results from singular value dimension (SVD) of environment-centred or environment-standardized.

#### **RESULTS**

#### Analysis of variance

Analysis of variance (ANOVA) across locations and seasons is presented in Table 2. There was highly significant difference (P<0.001) among the four seasons suggesting high variability among seasons. Seasons

**Table 1.** Genotypes, origin and their major identifiable characteristics.

Code name	Genotype	Origin	Yield	Resistance to Fusarium wilt disease	Resistance to gall midge	Hairiness on capsules	Branching habit	Plant height	Number of capsules per plant	Days to maturity
G1	(Ajimo A1- 6//7029)-1-1	A homozygous cross between a selected Ugandan land race (Ajimo A1-6) and a Thai accession (7029)	High	Resistant	Highly Resistant	Hairy	High	Tall	Many	>85
G2	(Ajimo A1- 6//7029)-1-9	A homozygous cross between a selected Ugandan land race (Ajimo A1-6) and a Thai accession (7029)	Low	Susceptible	Resistant	Hairy	less	Short	Few	Less than 75
G3	(Local 158//6022)- 1-2-1	A homozygous cross between an Egyptian accession (Local 158) and a Thai accession (6022)	High	Resistant	Highly resistant	Hairy	High	Tall	Many	>85
G4	(Sesim2//5181)-2- 2-1	A homozygous cross between a released commercial variety (Sesim 2) and a Thai accession (5181)	High	Resistant	Highly resistant	Hairy	Medium	Tall	Many	>85
G5	AD-1-1-1	Pureline Ugandan land race	Medium	Resistant	Medium resistant	Smooth	High	Tall	Many	>85
G6	Adong 4-4	Pureline Ugandan land race	Medium	Resistant	Medium resistant	Smooth	High	Tall	Many	>85
G7	EM15-1-5	Pureline Ugandan land race	Low	Resistant	Medium resistant	Light hairy	High	Tall	Medium	>85
G8	EM15-3-2	Pureline Ugandan land race	Medium	Resistant	Medium resistant	smooth	High	Tall	Many	>85
G9	Local 158-1	Egyptian pureline	High	Medium resistant	Highly resistant	Hairy	Medium	Tall	Many	>80
G10	Local 158-5	Egyptian pureline	High	Medium resistant	Highly resistant	Hairy	Medium	Tall	Many	>80
G11	Renner 1-3-1-14	USA pureline	Low	Resistant	Medium resistant	smooth	Medium	Medium	Few	>80
G12	Renner 1-3-1-16	USA pureline	Low	Resistant	Medium resistant	smooth	Medium	Short	Few	>80
G13	Renner 1-3-1-17	USA pureline	Medium	Resistant	Medium resistant	smooth	Medium	Medium	Medium	>80
G14	Renner 1-3-1-17-1	USA pureline	High	Resistant	Medium resistant	smooth	Medium	Medium	Medium	>80
G15	Sesim 1	Ugandan commercial variety	High	Resistant	Low resistant	smooth	High	Tall	Many	>85
G16	Sesim 2	Ugandan commercial variety	High	Resistant	Medium resistant	smooth	High	Tall	Many	>85

contributed 10% of the total sum of squares for variation. Location showed no significant difference suggesting that locations were similar in conditions. Highly significant difference (P<0.001) was recorded for season x location (S x

L). This showed that there was high interaction between the seasons and locations. Season by location contributed highest in the total sum of squares of variation with 45%. Highly significant difference (P<0.001) was recorded for genotypes.

This showed that there were differences in yield performance among the genotypes. Some genotypes were high yielding while others were poor yielding. Genotypes contributed 4% of the total sum of squares due to variation. Season x

Table 2. Combined analysis of variance of yield data of 16 sesame genotypes tested in 3 locations for 4 seasons.

Source of variation	D.F	S.S	M.S	F-value	p-value	%variation
Season(S)	3	4107465	1369155***	19.12	<0.001	10
Location (L)	2	12899	6449ns	0.09	0.914	0
Season x Location	6	18088342	3014724***	42.09	< 0.001	45
Residual	24	1718873	71620	3.21		
Genotype (G)	15	1934120	128941***	5.77	< 0.001	4
Season x Genotype (S x G)	45	1339039	29756ns	1.33	0.083	3.3
Location x Genotype (L x G)	30	1717895	57263***	2.56	< 0.001	4.3
Season x Location x Genotype (S x L x G)	90	3076274	34181**	1.53	0.003	7.7
Residual	360	8042700	22341			
Total	575	40037608				

ns = Non-significant; Significant: \*\*P<0.01, \*\*\*P< 0.001%.

Table 3. Mean yield performance (kg/ha) and ranking of each genotype for each location over four seasons.

Code no.	Genotype	NaSARRI	Rank	Kaberamaido	Rank	Ngetta	Rank	Overall mean	Overall rank
G1	(Ajimo A1-6//7029)-1-1	416	6	587	2	566	2	523	2
G2	(Ajimo A1-6//7029)-1-9	309	16	234	16	338	16	294	16
G3	(Local 158//6022)-1-2-1	401	9	563	3	501	7	488	4
G4	(Sesim2//5181)-2-2-1	447	2	531	5	536	4	505	3
G5	AD-1-1-1	393	10	490	8	501	7	461	8
G6	Adong 4-4	425	4	508	7	475	10	469	7
G7	EM15-1-5	345	13	481	9	464	12	430	12
G8	EM15-3-2	375	11	470	10	403	15	416	13
G9	Local 158-1	461	1	700	1	532	5	564	1
G10	Local 158-5	420	5	462	12	444	13	442	11
G11	Renner 1-3-1-14	413	7	293	14	428	14	378	14
G12	Renner 1-3-1-16	338	15	287	15	510	6	378	14
G13	Renner 1-3-1-17	408	8	470	10	541	3	473	6
G14	Renner 1-3-1-17-1	359	12	454	13	571	1	461	8
G15	Sesim 1	431	3	525	6	472	11	476	5
G16	Sesim 2	343	14	541	4	481	9	455	10
	Cv%	19.7		24.8		24			
	Prob.	0.218ns		0.001***		0.38 <sup>ns</sup>			

genotype (S x G) did not show any significant difference suggesting that high yielding genotypes performed better in all seasons and low yielding genotypes performed poorly in all seasons.

#### Combined performance analysis across locations

The overall mean yield performance across locations over seasons is presented in Table 3. Genotype G9 (Local 158-1) had the best performance at Serere and Kaberamaido and also the best yield of 564 kgha<sup>-1</sup> across locations followed by G1 (Ajimo A1-6//7029)-1-1 with 523

kgha<sup>-1</sup>. G2 (Ajimo A1-6//7029)-1-9 was the poorest yielding genotype in each location and across locations. The other poor yielding genotypes were G11 (Renner 1-3-1-14) and G12 (Renner 1-3-1-16). There were changes in the ranking within the locations and across locations. Highly significance among genotypes was only recorded for Kaberamaido.

#### AMMI and principal component analysis (PCA)

AMMI analysis of variance for seed yield of 16 sesame genotypes is presented in Table 4. The results showed

Table 4. AMMI analysis of variance for seed yield (kg/ha) of 16 sesame genotypes

Source of variation	df	SS	MS	F	F prob	% of variation
Replication	24	1718873	71620	3.21	0.00000***	
Treatments	191	30276034	158513	7.10	0.00000***	
Genotypes	15	1934120	128941	5.77	0.00000***	6.5
Environments	11	22208706	2018973	28.19	0.00000***	73.4
Interactions	165	6133208	37171	1.66	0.00004***	20.3
IPCA 1	25	2067259	82690	3.70	0.00000***	33.7
IPCA 2	23	1149880	49995	2.24	0.00108***	18.7
Residuals	117	2916069	24924	1.12	0.22414ns	
Error	360	8042700	22341			
Total	575	40037608	69631			

Plot of Gen & Env IPCA 2 scores versus means

Level of significance: ns=non-significant, \*\*\* (P< 0.01).

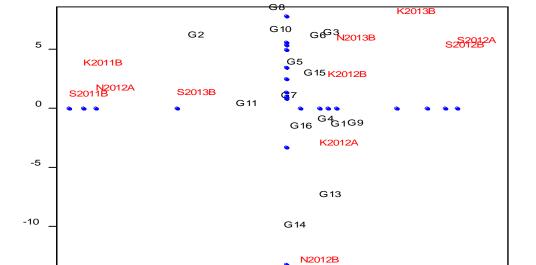


Figure 1. AMMI biplot showing the main and interaction (PC1) effects of both genotypes and location by seasons on sesame grain yield.

Genotype & Environment means

500

G12

400

that all the sources of variations were highly significant (P<0.001). Environment contributed the highest total variation of sums of squares with 73.4% followed by interactions with 20.3% and genotypes with 6.5%.

200

300

The AMMI biplot showing the main and interaction (PC1) effects of both genotypes and environments on sesame grain yield is presented in Figure 1. The grand mean for the genotype and environment is indicated by vertical dotted lines which was about 500 kg/ha. High yielding genotypes were located on the positive side of

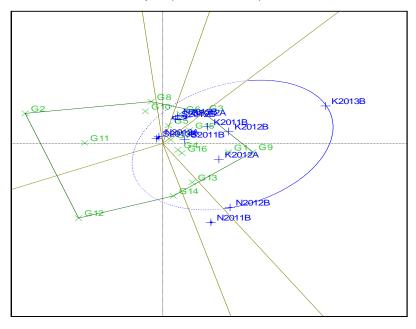
the graph. These were: G1 (Ajimo A1-6//7029)-1-1, G3 (Local 158//6022)-1-2-1, G4 (Sesim 2//5181)-2-2-1), G5 (AD-1-1-1), G6 (Adong 4-4), G7 (EM15-1-5), G9 (Local 158-1), G13 (Renner 1-3-1-17), G14 (Renner 1-3-1-17-1), G15 (Sesim 1) and G16 (Sesim 2). Stable and high yielding genotypes were G4 (Sesim 2//5181)-2-2-1), G6 (Adong 4-4), and G13 (Renner 1-3-1-17) as they are near the origin (Laurentin and Montila, 1999). Stable but poor yielding genotypes were G8 (EM15-3-2) and G10 (Local 158-5) as they are near the origin but on the left side of

N2011B

700

600

#### Scatter plot (Total - 61.47%)



#### PC1 - 47.21%



**Figure 2.** Polygon view of GGE biplot analysis showing the mega-environments and their respective high yielding genotypes for 'which-won-where' pattern.

the vertical line of the genotype and environment means. Poor yielding and unstable genotypes were G2 (Ajimo A1-6//7029)-1-9), G11 (Renner 1-3-14) and G12 (Renner 1-3-1-16) as they are far away from the origin and also positioned on the left side of the vertical line for genotype environment means. The best performing environments (location x season) were N2011B (Ngetta 2011B), S2012A (Serere 2012A), S2012B (Serere 2012B), K2012A (Kaberamaido 2012A), N2013B (Ngetta 2013B), K2012B (Kaberamaido 2012B) and N2012B (Ngetta 2012B). The poor performing environments were S2013B (Serere 2013B), N2012A (Ngetta 2012A), S2011B (Serere 2011B) and K2011B (Kaberamaido 2011B) with less than 500 kg/ha as the genotype and environment grand mean. Genotypes and location per season located near to the origin are considered to be stable.

#### **GGE** analyses

#### Which-won-where

The results of a pattern of "which-won-where" are

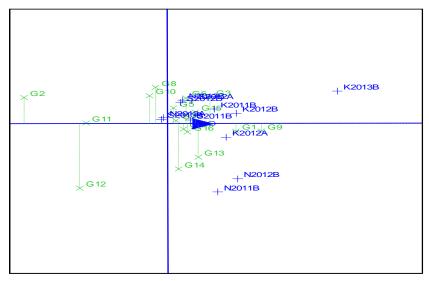
presented in Figure 2. Based on the three locations and seasons used in this study, only one mega-environment was identified as all environments were located in one sector. Locations by seasons within mega-environment 1 were Kaberamaido 2011B, 2012A, 2012B, 2013B and Ngetta 2011B and 2013B. For this mega-environment, G9 (Local 158-1) was the most positively responsive as it was at the vertex and therefore was the highest yielder at the vertex.

The polygon view of the GGE biplot showed that all test environments were divided into 6 sectors.

#### Means performance and stability

The results of the average environment coordination (AEC) views of the GGE-biplot based on environment-focus scaling for the means performance and stability of genotypes are presented in Figure 3. GGE biplot analysis produced good visual assessment of GGE with PCA1 (47.21%) and PCA2 (14.25%) explaining 61.47% of the total GE sum of squares. The environmental vector biplot identified N2011B and N2012B as highly

#### Ranking biplot (Total - 61.47%)

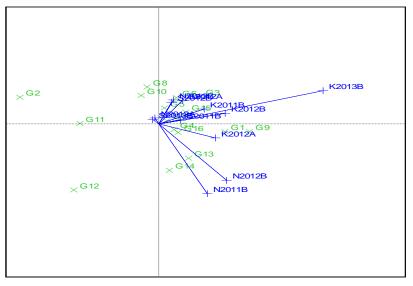


# PC1 - 47.21%

× Genotype scores
+ Environment scores
AEC

**Figure 3.** The average environment coordination (AEC) views of the GGE-biplot based on environment focused scaling for the means performance and stability of genotypes.

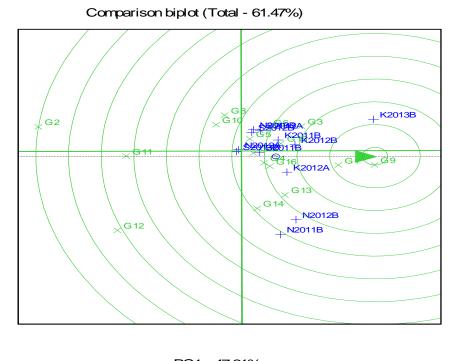
#### Scatter plot (Total - 61.47%)

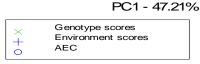


PC1 - 47.21%

Genotype scoresEnvironment scoresVectors

**Figure 4.** The environment vector biplot showing environmental differences in discriminating the 16 genotypes for yield at the twelve test environments





**Figure 5.** GGE-biplot for comparison of the genotypes with the ideal genotype.

discriminating for the genotypes tested, as evidenced by the long environment vectors. The yield and stability were considered simultaneously and the average environment coordinate (AEC) biplot was generated. The highest yielding and stable genotype was G9 followed by G1. G11 was the most stable but one of the poorest yielders. G12 was the most unstable as it had the longest vector. Unlike PC1, genotypic PC2 scores near zero exhibited large scores stable genotypes whereas PC2 discriminated the unstable ones. S2011B, K2011B, K2012A and K2012B were stable environments while N2011B and N2012B were the most unstable environments. K2013B was the most productive environment but most discriminative for genotypes.

#### Discriminant analysis

Discriminating vector showing ability of an environment is presented in Figure 4. Kaberamaido during 2013B (K2013B) was the most discriminating and responsive as it had the longest vector from the biplot origin. They are stable environments but not informative as they were not discriminatory. Environments were positively correlated since the cosine angles were less than 90°. K2011B, K2012A, K2012B, S2011B, N2012A, N2013B and all the

seasons at Serere had short vectors. Genotypes G2, G5, G10, G11, G12 and G14 were not located in any environment. They are poor genotypes that did poorly in most environments.

#### Ideal genotype

The GGE-biplot for comparisons of the genotypes with the ideal genotype is presented in Figure 5. The genotype G9 followed by genotypes G1 which were closest to the inner most concentric center. Genotypes G3, G4, G5, G6, G7, G13, G14, G15 and G16 were above the grand mean indicated by the vertical line in the centre are considered desirable genotypes because they are within the circles that surround the circle of the virtual ideal genotype. Genotypes G2, G12 and G11 were furthest from the ideal genotype.

#### Ideal environment

Comparison of environments to an ideal environment is presented in Figure 6. In this experiment, the ideal environment was also represented by the central concentric circle with an arrow passing through it and it

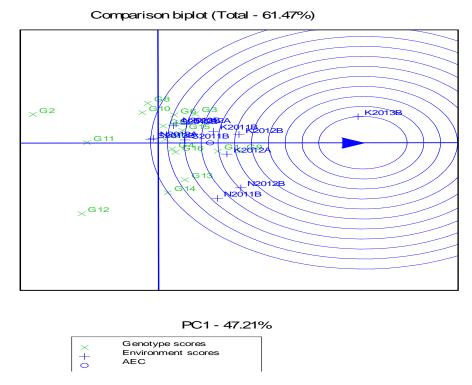


Figure 6. GGE-biplot for comparison of environments with the ideal environment.

showed that K2013B (Kaberamaido 2013B) was the closest to ideal environment. K2011B, K2012A, K2012B and N2012B were also desirable environments as they were located in the middle parts of the concentric rings towards the ideal environment. Ngetta 2012A, S2012A, N2013B and S2013B were the worst seasons (environments) as they were the furthest from ideal environment which is in the innermost concentric rings.

#### DISCUSSION

#### **Analysis of variance**

Analysis of variance (ANOVA) across locations and seasons recorded highly significant difference (P<0.001) among the four seasons suggesting that there were changes in the conditions of the weather in the different locations in different seasons. Climatic conditions and different soil constituents cause annual variations (Mortazavian et al., 2014). Seasons contributed 10% of the total sum of squares for variation. Location showed no significant difference suggesting that locations were similar in conditions. Rahmatollah et al. (2013) in their study found similar results. Highly significant difference (P<0.001) was recorded for season x location (S x L). This showed that there was high interaction between the seasons and locations. Season by location contributed highest in the total sum of squares of variation with 45%.

Rahmatollah et al. (2013) recorded similar highly significant year x location effect. Highly significant difference (P<0.001) was recorded for genotypes. This showed that there were differences in yield performance among the genotypes. Some genotypes were high yielding while others were poor yielding. Genotypes contributed 4% of the total sum of squares due to variation. Rahmatollah et al. (2013) also recorded similar significant effect for genotypes and attributed that possibly due to changes in genotype characteristics, varying from one genotype to another. Velu and Shunmugavalli (2005) also reported similar highly significant difference among genotypes. genotype (S x G) did not show any significant difference suggesting that high yielding genotypes performed better in all seasons and low yielding genotypes performed poorly in all seasons. Rahmatollah et al. (2013) also recorded no significant effect in genotype by year although they did not use seasons. Location by genotype interaction (L x G) was highly significant (P<0.001) suggesting that genotypes interacted with locations and performed differently thus changing in ranking. Rahmatollah et al. (2013) also recorded significant location by genotype effect .The season x location x genotype (S x L x G) interaction showed highly significant difference (P<0.001) suggesting that there was high interaction among genotypes during the seasons in those different locations. John et al. (2001) also observed the mean squares for genotypes, seasons (environments)

and G x E interactions to be highly significant

Partitioning of variance components for environment into predictable (locations) and unpredictable (seasons or year) is very important (Rahmatollah et al., 2013). When GE interaction is due to variation in predictable factors, a plant breeder has the choice of either developing specific genotypes for selected environments or broadly adapted genotypes that can perform well under variable conditions (Dehghani et al., 2006). When GE interaction results from unpredictable sources, a plant breeder needs to develop stable genotypes that perform reasonably well under a range of environmental conditions.

#### Combined performance analysis across locations

The overall mean yield performance across locations over seasons showed that genotype G9 (Local 158-1) had the best yield of 564 kgha followed by G1 (Ajimo A1-6//7029)-1-1 with 523 kgha<sup>-1</sup>. G2 (Ajimo A1-6//7029)-1-9 was the poorest yielding genotype across all locations. G2 is an early maturing genotype and susceptible to sesame Fusarium wilt which tends to reduce its yield in some seasons. Normally early or very early varieties do not perform very well. The local selected lines were adaptable to the local conditions as they have withstood selection pressure within the environment over the years. This is also reported by Ashri (1989). Local 158-1 and its other series, and Sesim2//5181 are hairy genotypes as shown in Table 1 and they have been recorded to be tolerant to sesame gall midge that cause flower and capsule damage to sesame thus reducing yield (Ogwal et al., 2003). Sesim 1 and Sesim 2 had already been officially released in Uganda for commercial production in 2001 and 2003 respectively. Local 158-1 was officially released for commercial production as Sesim 3 in 2013 because of its high yield, stability, seed color and hairyness of the stem and capsules that gives it resistance to gall midge insect (Asphondylia sesame).

#### AMMI and principal component analysis (PCA)

AMMI analysis of variance for seed yield of 16 sesame genotypes showed that all the sources of variations were highly significant (P<0.001). Environment contributed the highest total variation of sums of squares with 73.4% followed by interactions with 20.3% and genotypes with 6.4%. Laurentin and Montilla (1999) recorded 94% of the AMMI model sum of squares attributed totally to G x E interaction.

The AMMI biplot showing the main and interaction (PC1) effects of both genotypes and environments on sesame grain yield presented in Figure 1 had high yielding genotypes located on the positive side of the graph. Stable and high yielding genotypes were G4

(Sesim 2//5181)-2-2-1), G6 (Adong 4-4), and G13 (Renner 1-3-1-17) as they are near the origin. Stable but poor yielding genotypes were G8 (EM15-3-2) and G10 (Local 158-5) as they are near the origin but on the left side of the vertical line of the genotype and environment means. Poor yielding and unstable genotypes were G2 (Ajimo A1-6//7029)-1-9), G11 (Renner 1-3-14) and G12 (Renner 1-3-1-16) as they are far away from the origin and also positioned on the left side of the vertical line for genotype and environment means which indicates areas of low yield (Laurentin and Montila, 1999). Genotypes and location per season located near to the origin are considered to be stable (Laurentin and Montila, 1999). Also, genotypes and environments that fall in the same sector interact positively while those falling into opposite sectors interact negatively (Velu and Shunmugavalli, 2005).

#### **GGE** analyses

#### Which-won-where

The results of a pattern of "which-won-where" as presented in Figure 2, based on the three locations and seasons used in this study showed that only one megaenvironment was identified as all environments were located in one sector. Mega environments are test environments with different winning genotypes located at the vertex of the polygon and located in different sectors (Gauch et al., 2008). Locations by seasons within megaenvironment 1 were Kaberamaido 2011B, 2012A, 2012B, 2013B and Ngetta 2011B and 2013B. For this megaenvironment, G9 (Local 158-1) was the most positively responsive as it was at the vertex and therefore was the highest yielder at the vertex. Although other genotypes were located in other sectors, they did not fall in any environment and therefore those sectors were not considered as mega-environments. Yan et al. (2007) suggested that environment markers that fall into a single sector indicate rank-two approximation with a single cultivar having the highest yield in all environments. If environment markers fall into different sectors, then different cultivars won in different sectors. Since a megaenvironment is defined as a group of locations that consistently share the best set of genotypes (Yan and Rajcan, 2002), data from multiple years are essential to decide whether or not the target region can be divided into different mega-environments. Each sector has its most favourable genotypes and corner genotypes not in the environments are the poorest yielding (Karimizadeh, 2013; Pourdad and Moghaddam, 2013). They are located far away from all test locations, reflecting the fact that they yielded poorly at each location.

The polygon view of the GGE biplot showed that all test environments were divided into 6 sectors. Visualization of which-won-where pattern of MEYTs data is important for studying the possible existence of different megaenvironment in a region (Yan and Rajcan, 2002). Corner genotypes which are at the vertex located at the extreme point of the polygon in a sector are the most responsive ones (Rahmatollah et al., 2013). They are the best or the poorest as they are farthest from the origin of the biplot (Yang and Kang, 2003). The polygon view of a GGE biplot explicitly displays the which-won-where pattern (Yan et al., 2001) because each sector would show the vertex with the the indicative genotype and the positions of all other genotypes showing their responsiveness to the environment under study. Genotypes within the polygon are less responsive to location than the vertex genotypes (Rahmatollah et al., 2013).

#### Mean performance and stability

The results of the average environment coordination (AEC) views of the GGE-biplot based on environmentfocus scaling for the means performance and stability of genotypes presented in Figure 3 produced good visual assessment of GGE with PCA1 (47.21%) and PCA2 (14.25%) explaining 68.47% of the total GE sum of squares. Farshadfar et al. (2012) partitioned GE interaction through GGE biplot analysis and showed that PC1 and PC2 accounted for 39.1 and 37.79% of GGE sum of squares, respectively, explaining a total of 76.8% variation. The yield and stability were considered simultaneously and the average environment coordinate (AEC) biplot was generated. The average environment, represented by a small circle, was defined by the PC1 and PC2 scores of the environments (Yan and Kang. 2003). The ordinate of the AEC is the line that passes through the origin and is perpendicular to the abscissa. The genotypes G9 (Local 158-1) was the top yielding genotype, as presented on the front of an average environment towards the pointing arrow of the AEC abscissa. In addition, the biplot indicated that G9 had the highest mean yield and one of the most stable genotypes as it was positioned close to the AEC abscissa (Yan, 2002). The second highest yielding and most stable genotype was G1. G12 was the most unstable as it had the longest vector to the AEC. Farshadfar and Sadeghi (2014) explained that genotypic PC1 scores>0 classified the high yielding genotypes while PC1< 0 identified low yielding genotypes. The genotypes with PC1 scores close to zero expressed general adaptation whereas the larger scores depicted more specific adaptation to environments with PC1 scores of the same sign (Ebdon and Gauch, 2002). Unlike PC1, genotypic PC2 scores near zero exhibited stable genotypes whereas large PC2 scores discriminated the unstable ones. S2011B, K2011B, K2012A and K2012B were stable environments while N2011B and N2012B were the most unstable environments. K2013B was the most productive but responsive suggesting that it is high yielding in particular environment and not stable across environments. The smaller the absolute length of projection of a genotype, the more stable it is (Yan, 2002). A longer projection to the AEC ordinate, regardless of the direction, represent a greater tendency of the GEI of a genotype, which means it is more variable and less stable across environments or vice versa. However, considering both mean yield and stability concepts, plant breeders explore genotypes that indicate yield stability as well as high yield across environments (Kang, 2002).

#### Discriminant analysis

Discriminating vector showing ability of an environment presented in Figure 4 showed that location vectors are lines that connect the biplot origin and the marker of test locations. A long environment vector represents good discriminating ability for a given environment (Yan et al., 2000). The lines of the GGE biplot in which the environments are connected with the biplot origin are called vectors. The cosine of the angle between the vectors of two environments is approximate to the correlation coefficient between them (Kroonenberg, 1995; Yan, 2002). The angles between vectors are related to the correlation coefficient (Kroonenberg, 1995). The angles between the environments were less than 90° indicating that most environments were positively correlated to each other. K2013B had the longest vector from the origin and therefore it is highly discriminating for the genotypes tested. Another interesting observation from the vector view of the biplot is that the length of the environment vectors is approximate to the standard deviation within each environment, which is a measure of their discriminating ability (Yan et al., 2003). Long vectors are least stable and those with short vectors and near the horizontal axis are most stable.

#### Ideal genotype

The GGE-biplot for comparisons of the genotypes with the ideal genotype presented in Figure 5 showed that the center of the concentric circles is where an ideal genotype (high mean yield and the most stable one) should be located (Yan, 2002). In other words, projection of the ideal genotype on the ATC horizontal axis is equal to the longest vector of all genotypes and its projection on the ATC vertical axis is obviously zero (it is absolutely stable(Yan et al., 2003)). The smaller the distance from genotype to the virtual ideal genotype, the better yielding the genotype. Therefore genotype G9 followed by genotypes G1 which were closest to the concentric center which indicates ideal genotype were the most yielding and stable genotypes. Genotypes G3, G4, G5, G6, G7, G13, G14, G15 and G16 are considered desirable genotypes because they are within the circles

that surround the circle of the virtual ideal genotype. Genotypes G2, G12 and G11 were the genotypes furthest from the ideal genotype and so considered as the poorest yielding genotypes. An ideal genotype is defined as one that is the highest yielding across all test environments and is absolutely stable in all test environments (Yan et al., 2003). Although such an ideal cultivar may not exist in reality, it can be used as a reference for cultivar evaluation (Yan, 2002; Mitrovic et al., 2012).

#### Ideal environment

Comparison of the test environments used in this study to an ideal environment presented in Figure 6 indicated that the ideal environment was also represented by the central circle with an arrow passing through it. It showed that K2013B (Kaberamaido 2013B) was the closest to ideal environment and therefore the most desirable of all the seasons and most effective season for selecting superior genotypes. K2011B, K2012A, K2012B and N2012B were also desirable environments as they were located in the middle parts of the concentric rings towards the ideal environment. Ngetta 2012A, S2012A, N2013B and S2013B were the worst seasons (environments) as they were the furthest from ideal environment. Yan and Rajcan (2002) defined an ideal test environment as having small PC2 scores (more representative of the overall environment) and large PC 1 scores (power to discriminate).

#### Conclusion

Different genotypes performed differently in the seasons and locations. There were interactions between locations and seasons in the performance of genotypes. The GGE model aided in determination of the relative performance of genotypes at different environments and identification of genotypes suitable for groups of environments. The genotypes at the vertex showed the which-won-where. That is, it showed the identification of mega-environments as well as the winning genotypes in those mega-environments.

One season was identified as mega-environment for sesame in the trial sites with G9 (Local 158-1) being the highest yielder suggesting that the locations were similar in their conditions.

#### Conflict of Interests

The authors have not declared any conflict of interests.

#### **ACKNOWLEDGEMENT**

The authors are grateful to the sesame technician, field

staff at NaSARRI and Trial Verification Officers of Kaberamaido and Ngetta centres who maintained the trials. The study was funded by the Austrian Development Agency (ADA) through the International Crops Research Institute for the Semi-Arid Tropics (ICRISAT) for the trials in 2011B, 2012A, 2012B and RUFORUM during 2013B. Dr. George William Otim-Nape is acknowledged for helping access funding support of this project.

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Vol. 10(10), pp. 203-210, October 2016

DOI: 10.5897/AJPS2016.1454 Article Number: 767035561077

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Full Length Research Paper

# Antifungal and cytotoxic activity of purified biocomponents as carvone, menthone, menthofuran and pulegone from *Mentha* spp.

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Received 10 August, 2016; Accepted 20 September, 2016

Medicinal plants are attractive sources in the search of bioactive compounds in the treatment of infectious diseases. Considering that the infectious agent often develops resistance to existing treatments rapidly, the searches for such compounds are a never-ending process. One such infection, caused by *Candida* spp. is candidiasis, which is a public health problem. Additionally, many strains have resistance to traditional therapies. Therefore, we tested the antifungal activity of four compounds present in *Mentha* spp. with promising antifungal precedents. We measured inhibition of growth by microdilution, disruption of biofilm viewed by electron microscopy, inhibition of germ tube formation by optical microscopy and toxicity on HaCaT cells. Tests showed that the compounds tested had antifungal activity with a minimum inhibitory concentration of 0.5 mg/mL, at least, 50% of biofilm inhibition in the 0.5 mg/mL concentration, an inhibition of polymorphism to 86% and the changes in the cell envelope of yeast (SEM) and cell viability above 50% among the *Candida* strains tested. Therefore, the compounds exhibit promising antifungal properties and provide a reasonable therapeutic window to be used in association with other traditional antimitotic.

Key words: Carvone, menthone, menthofuran, pulegone, Candida spp., biofilm, cytotoxicity.

#### INTRODUCTION

The emergence and propagation of microorganisms resistant to traditional chemotherapies has become an important public health problem (Sprenger and Fukuda, 2016). *Candida* species also has develop strategies to evade antifungal treatment (Acker et al., 2014). Recently,

researchers have elucidated that *Candida* species have a series of mechanisms involving virulence factors that contribute to their infection, proliferation, adaptation, and long-term survival in the human body. Two factors important in virulence are (1) polymorphism, a

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morphological transition where the yeast cell forms filaments that aids in infection of the host (Jacobsen et al., 2012); and (2) biofilm formation, which reduces the susceptibility of microorganisms to various immunological and chemotherapeutic modes of action, contributing to persistent infection (Chandra and Mukherje, 2015).

In order, to combat the increase of microorganisms resistant, it becomes necessary to continually develop new classes of antifungal compounds. Plant extracts provide a promising source of molecules, due to the long history of mutual tolerance and co-regulation between plants and fungi (Saharkhiz et al., 2012). We chose to focus our attention on the essential of Mentha spp. because it has been shown to contain a number of bioactive compounds, including those that may have antimicrobial activity (Schelz et al., 2006; Mkaddem et al., 2009; Zore et al., 2011).

We evaluated carvone, menthone, menthofuran and pulegone present in *Mentha* spp. against yeasts of the genus *Candida* and how they affect growth and survival, germ tube formation and biofilm formation and maintenance. In addition, we evaluated the toxicity of these compounds on HaCaT cells.

#### **MATERIALS AND METHODS**

Planktonic anti-Candida assay, minimum fungicidal concentration (MFC) and inhibition of germ tube formation were performed with three independent experiments; biofilm and cytotoxicity assay were performed with two independent experiments in triplicate. All tests were performed include positive control (without compounds) and negative control (without cell suspension).

#### Reagent preparation

Carvone, menthone, menthofuran and pulegone (acquired from Sigma- Aldrich; the purity was adjusted to 100%) were diluted in aqueous Tween 80 (0.05%). The commercial antifungal fluconazole was diluted in dimethyl sulfoxide (DMSO).

#### **Candida strains**

Candida species standard: Candida rugosa (IZ 12), Candida lusitaniae (IZ 06), Candida glabrata (IZ 07), Candida utilis (CBS 5609), Candida krusei (CBS 573), Candida albicans (ATCC 90028), Candida albicans (ATCC MYA-2876), Candida guilliermondii (CBS 566), Candida tropicalis (CBS 74), C. albicans (CBS 562), Candida parapsilosis (CBS 604) and Candida dubliniensis (CBS 7987). (CBS- Centraal bureau voor Schimmel cultures; IZ: Zimotécnico Institute-ESALQ-USP, Brazil; ATCC: American Type Culture Collection- The Global Bioresource Center. Clinical isolates obtained from oral cavity and oral prosthesis were provided by Prof. Dr. Marcelo Fabiano Gomes Boriollo: Candida albicans (314), Candida albicans (330), Candida albicans (335), Candida albicans (368), Candida albicans (378) and Candida albicans (462).

#### Planktonic anti-Candida assay

The assay planktonic anti-Candida (Table 1) was made according

to the CLSI, 2008- M27-A3 reference method. In summary, yeast was inoculated in Sabouraud Dextrose Agar (SDA) culture medium and incubated aerobically at 37° C for 24 h. After that, was diluted to  $2.5\times10^3$  CFU/mL in RPMI culture medium and measured at  $A_{530}$ . In microtiter plates, were made dilution series with compounds in RPMI (concentration range from 8 to 0.125 mg/mL) and added the cell. Fluconazole was tested (concentration range 64 to 0.5  $\mu$ g/mL). The plates were incubated for 48 h at 37°C in aerobic conditions, followed by visual reading to evaluate cell growth. The concentrations that no showed cell growth was considered minimum inhibitory concentration (MIC).

#### Minimum fungicidal concentration (MFC)

The fungal viability after incubation with test compounds (concentration range 16 to 0.125 mg/mL) was performed by plating an aliquot of the MIC plate well contents to SDA plates and incubating at 37°C for 48 h. MFC was determined as the lowest concentration of a given compound that did not allow the growth of any fungal colony on SDA after the incubation period. The read was made visually (Gullo et al., 2012).

#### Inhibition of germ tube formation

The compounds were tested at sub-MIC concentrations (Table 2). *Candida albicans* MYA-2876 cells were grown on SDA for 24 h at 37°C under aerobic conditions and adjusted to 2.5×10<sup>7</sup> CFU/mL in PBS (1x) and Neubauer chamber under optical microscope (magnification 400x). The cell suspension was added a 1:1 mixture of RPMI culture medium (with diluted compounds) and fetal bovine serum. The cells count that forming germ tubes were conducted using an optical microscope at 400 x magnifications at intervals of 2, 4 and 6 h of aerobic incubation at 37°C and compared to positive control. Values were calculated as percentage (Consolaro et al., 2005).

#### Effect of compounds on biofilm

#### Inoculum adjustment

*C. albicans* MYA-2876 were incubated in yeast extract peptone dextrose (YPD) liquid medium for 24 h in orbital shaker (30 rpm) at 30°C. An aliquot of 7 mL of the inoculum was centrifuged at 10.8 *g* for 4 minutes. This process was performed twice with a phosphate buffered saline (PBS) (1x) wash step. The pellet was resuspended in 7 mL of RPMI. Cell concentration was determined using a Neubauer chamber under optical microscope (magnification 400x); the suspension was diluted to 1.0x10<sup>6</sup> CFU/mL in RPMI (Pierce et al., 2008).

#### **Biofilm formation**

The cell suspension (100  $\mu$ L) was added to a sterile microtiter plate with U-shaped wells. The cell suspension were incubated for 2 h under agitation (100 rpm) at 37°C. After incubation, the plate was washed 3 times with saline (0.9%). Solutions of the compounds (concentration range 16 to 0.125 mg/mL) were added to the wells. The cells were incubated for 24 h at 37°C under aerobic conditions. After incubation period, were compared to the positive control (Da Silva et al., 2010).

#### Mature biofilm

The cell suspension (100 µL) was added to a sterile microtiter plate

Table 1. MIC and MFC of the compounds tested against Candida spp.

	Carv	one/	Menthone		Menth	Menthofuran		gone	Fluconazole
Strains	MIC	MFC	MIC	MFC	MIC	MFC	MIC	MFC	MIC
	(mg/mL)	(mg/mL)	(mg/mL)	(mg/mL)	(mg/mL)	(mg/mL)	(mg/mL)	(mg/mL)	(µg/mL)
Cr 12	4	8	8	8	8	8	1	2	-
CI 06	2	4	4	4	2	2	1	2	0,5
Cg 07	4	8	8	-	8	8	2	2	32
Ck 573	2	2	8	8	4	4	1	1	16
Ca 90028	2	4	8	8	4	4	2	4	-
Ca 2876	2	4	8	8	4	4	2	2	-
Cg 566	1	1	4	8	4	4	1	1	1
Ct 94	2	4	-	-	8	8	2	2	-
Ca 562	4	8	8	8	8	8	2	4	-
Cp 604	1	2	8	-	4	8	1	2	4
Cd 7987	2	4	8	8	4	8	1	2	1
Ca 314	1	2	8	8	4	8	1	1	-
Ca 330	1	2	8	8	8	8	1	1	-
Ca 335	1	2	8	8	4	8	1	1	-
Ca 338	0,5	1	8	8	4	8	1	1	-
Ca 368	0,5	2	4	8	4	8	1	1	-
Ca 378	0,5	2	8	8	4	8	1	1	-
Ca 462	1	4	8	8	4	8	1	1	

<sup>-,</sup> Without activity; Cr 12, C. rugosa IZ; Cl 06, C. lusitaniae IZ; Cg 07, C. glabrata IZ; Ck 573, C. krusei CBS; Ca 90028, C. albicans ATCC; Ca 2876, C. albicans ATCC; Cg 566: C. guilliermondii CBS; Ct 94, C. tropicalis CBS; Ca 562, C. albicans CBS; Cp 604: C. parapsilosis CBS; Cd 7987: C. dubliniensis CBS. C. albicans clinical isolates: Ca 314; Ca 330; Ca 335; Ca 338; Ca 368; Ca 378; Ca 462.

Table 2. Inhibition of germ tube formation expressed in percentage.

Incubation paried (baura)	Inhibition percentage						
Incubation period (hours)	Carvone (1 mg/mL)	Menthone (4 mg/mL)	Menthofuran (2 mg/mL)	Pulegone (1 mg/mL)			
2	72	65	65	79			
4	79	64	69	85			
6	79	66	70	86			

with U-shaped wells. The cells were incubated aerobically at 37°C for 24 h. Solutions of the compounds (concentration range 16 to 0.125 mg/mL) were added to the wells. The suspension cell with compounds was incubated for 24 h at 37°C under aerobic conditions. After incubation period, were compared to the positive control (Pierce et al., 2008).

#### Biofilm quantification

After incubation period, the plates were washed 3x with saline (0.9%) for remove planktonic cells. The quantification of the fungal cell viability was calculate using a colorimetric XTT [2,3-bis(2-metoxi-4-nitro-5-sulfo-fenil) -2 H -tetrazolium-5-carboxanilida] reduction assay, in which were add in the plates 80  $\mu$ l solution XTT and measured in reader microplate spectrophotometer at A<sub>490</sub>.

#### Scanning electron microscopy (SEM) of the biofilm

The biofilm formation and mature biofilm samples (in 0.5 mg/mL

concentration) were grown in culture slides (BD Falcon). After the incubation period, the supernatant was discarded and biofilm fixed with glutaraldehyde (2.0%) for 30 min followed by drying ambient temperature. Specimens were dehydrated with increasing concentrations of ethanol (50, 70, 90 and 100%) for 10 min each. After that, specimens were dried, metallized with gold and observed by SEM (JEOL JSM 5600LV) to view the biofilm structure in the compounds presence. The images were selected randomly.

#### Cytotoxicity of the compounds

HaCaT cells (epithelial cells from normal human keratinocytes immortalized but not transformed) were grown in RPMI/FBS and adjusted to  $6.5\times10^4$  CFU/mL in Neubauer chamber under optical microscope (magnification 400x). Posteriorly, 100  $\mu L$  of the cells was added to microtiter plate and incubated for 24 h at 37°C and 5% CO2. Subsequently, the compounds (concentration range 16 to 0.125 mg/mL) were added in the cells and incubated for 24 h under the same conditions. The cells were fixed with trichloroacetic acid (TCA) 10% and incubated for 1 h at 4°C. The plates were washed

with distilled water (3x), dried at ambient temperature and stained with sulforhodamine B (SRB, 0.4) dissolved in 1% aqueous acetic acid, washed 4 times with distilled water, and then treated with SRB in 10  $\mu$ M TRIZMA pH 8.0. The cell suspension were measured at  $A_{530}$  in microplate reader to evaluate cell viability after treatment with the compounds (Endo et al., 2010) and reported as IC $_{50}\%$ , the concentration of the compound that induces 50% lysis or cell death (Liu et al., 2016).

#### Statistical analysis

The biofilm and cytotoxicity tests were analyzed by the Dunnett statistical test (ANOVA) using the software Bioestat 5.0; a p<0.05 was considered statistically significant.

#### **RESULTS**

#### Planktonic anti-Candida assay

The compounds tested showed inhibitory activity against nearly all *Candida* spp. for the concentration range tested, with the exception of menthone for *C. tropicalis* CBS 94 (Table 1). Carvone and pulegone showed inhibitory activity at lower concentrations compared to menthone and menthofuran. A commercially available antifungal agent tested was Fluconazole, to compare the results of inhibition obtained by isolated compounds of essencial oils obtained from *Mentha* spp. Fluconazole showed inhibitory activity only in some strains tested.

#### Minimum fungicidal concentration (MFC)

Carvone and pulegone expressed MFC for the strains tested in lower concentrations compared to the other compounds. The concentration of 2 mg/mL of carvone and 1 mg/ml of pulegone showed most fungicidal activity inhibiting most species tested (Table 1).

#### Inhibition of germ tube formation

Pulegone showed the greatest efficiency inhibiting until 86% germ tube formation compared to other compounds and positive control as shown in Table 2.

#### Action of compounds on biofilm

The compounds tested prevented at least 50% adhesion, inhibited progress of biofilm formation, and damaged of mature biofilms at low concentrations: 0.250 mg/mL for carvone and pulegone; 1 mg/ml for menthone and 0.5 mg/mL for menthofuran compared to the positive control (Figure 1).

SEM assay was made to view the action of the compounds on biofilm. The images obtained showed changes in cells as pore formation, roughness and

presence of leakage of cellular contents after treatment with the compounds. The results of biofilm formation are shown in Figure 2 and mature biofilm in Figure 3.

#### Cytotoxicity of the purified compounds

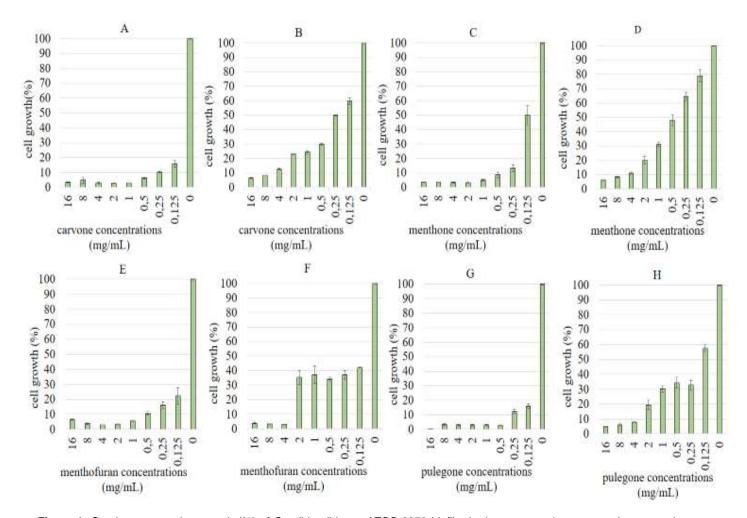
Cytotoxicity evaluation of the compounds in HaCaT cells using menthone and menthofuran showed cell viability above 50% at concentrations 4 to 0.125 mg/mL; carvone and pulegone showed cell viability above 50% at concentrations of 8 to 0.125 mg/mL compared to positive control (Figure 4).

#### DISCUSSION

In planktonic anti-Candida assays, all compounds showed antifungal activity, however, carvone and pulegone inhibited the growth of most strains in less concentrations compared to other compounds. These results indicate a greater antifungal potential of carvone and pulegone against these species. The results obtained with the commercial antifungal Fluconazole did not exhibited antifungal activity against some species including clinical isolates. These results corroborate with Ramesh et al. (2010) demonstrating that clinical isolates strains of Candida spp. were resistant to Fluconazole. In this sense, the results obtained was satisfactory because the compounds showed antifungal activity in species that Fluconazole did not showed, suggesting that such compounds may be a possible source for new drugs, especially as adjuvant. Minimum fungicidal concentration was performed to confirm cell death in MIC through observation of no colony growth in solid medium and classify fungistatic or fungicidal activity by compounds (Gullo et al., 2012). Carvone and pulegone expressed MFC for the strains tested in lower concentrations demonstrating most fungicidal character compared to the other compounds.

We also observe that the compounds tested acted in virulence factors of *Candida* spp. such as hyphae and biofilm. The germ tube formation is the beginning of the growth of hyphae (Ellepola and Saramayake, 2001). We observed that compounds reduced germ tube formation. These results are important because this form is related to the invasiveness of the fungus to the host tissue (Jacobsen et al., 2012; Mayer et al., 2013). In biofilm tests, the compounds reduced biofilm formation and deconstructed the mature biofilm at low concentrations tested. Considering that biofilms are associated with approximately 80 of infections caused by *Candida* spp. (Tsang et al., 2012), such compounds can be promise as therapeutic agents against this virulence factor.

The antifungal action of the compounds can be attributed to their chemical class. Other members of the terpenoids are known to destabilize cell membranes and increase cellular permeability which permit the disruption



**Figure 1.** Graphs representing growth (%) of Candida albicans ATCC-2876 biofilm in the compounds presence (concentration range 16.0 mg/ml to 0.125 mg/ml). All results showed statistical significance (Dunnett test, ANOVA). 0 mg/ml indicates positive control. Biofilm formation in the presence of (A) carvone; (c) menthone; (E) pulegone. Mature biofilm exposed to (B) carvone; (D) menthone; (f) menthofuran; (H) pulegone.

and death of the microorganisms (Zore et al., 2011). The SEM assay was performed to evaluated the action of the compounds in *C. albicans* MYA-2876 biofilms. The images obtained showed changes in cells as the presence of pores and ridges in the cell envelope and extravasation of cellular contents confirming this hypothesis.

With the purpose to evaluate the therapeutic applicability of the compounds, we performed cytotoxicity assays on HaCaT epithelial cells. The compounds showed results with cell viability above 50% in the most part concentrations tested although high concentrations of some neared the 50% viability limit. The low cytotoxicity of compounds isolated from *Mentha* spp. has also been demonstrated by Amaral et al. (2015) who evaluated cytotoxicity of rotundifolone, a major constituent of the essential oil of *Mentha x villosa* against tumor cell lines HCT-116 and SF-295. The data obtained in the initial cytotoxicity assays indicate a potential

therapeutic window for these compounds isolated to be used as antifungal agents.

#### Conclusion

The compounds tested modify the pathogenic *Candida* spp. fungus in many ways, including inhibition of growth, formation and maintenance of biofilms, as well as germ tube formation. Although each compound and strain has their unique profile of action or resistance, the low cytotoxicity and therapeutic effect of these compounds make them worthwhile candidates for complementing antifungal clinical protocols.

#### **Conflicts of Interests**

The authors have not declared any conflict of interests.

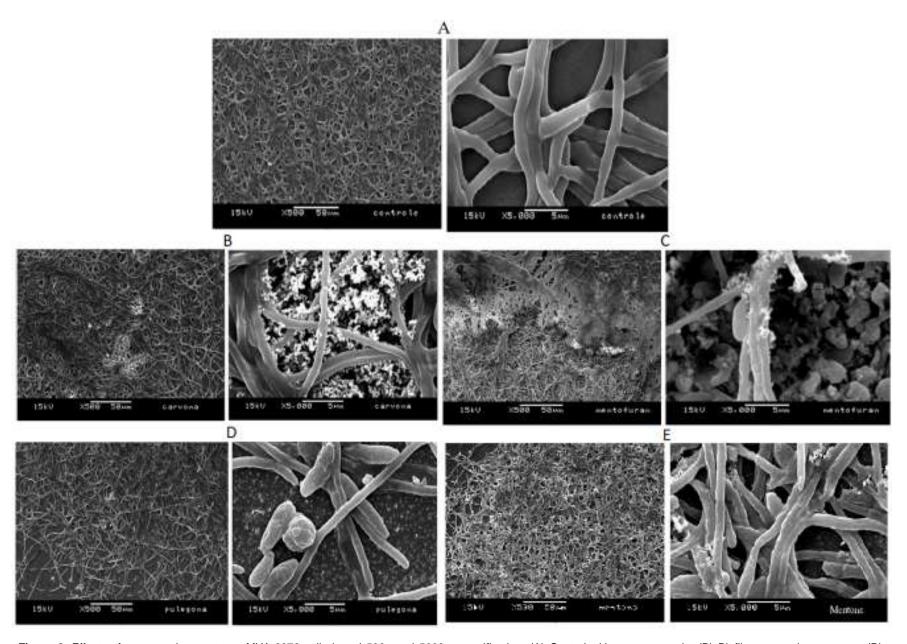
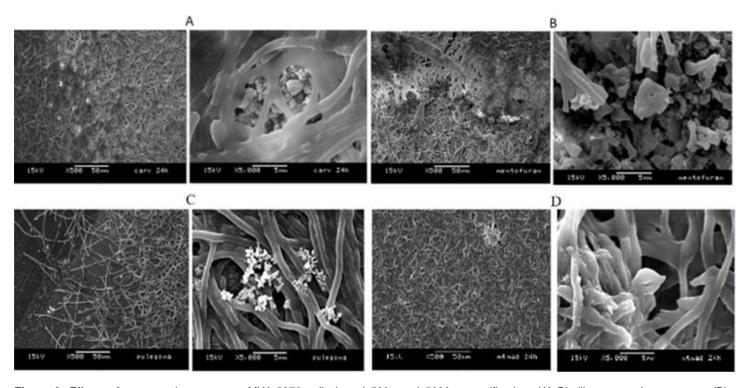


Figure 2. Effects of compounds on mature MYA-2876 cell viewed 500x and 5000x magnification. (A) Control without compounds. (B) Biofilm exposed to carvone; (D) pulegone; (E) menthone.



**Figure 3.** Effects of compounds on mature MYA-2876 cell viewed 500x and 5000x magnification. (A) Biodilm exposed to carvone; (B) pulegone; (D) menthone.

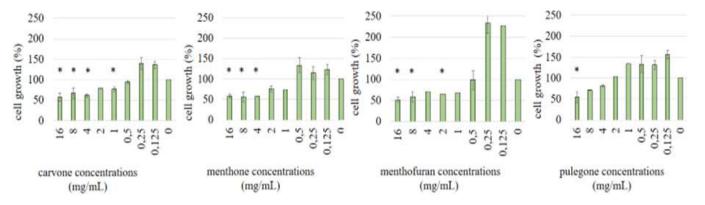


Figure 4. Graphs representing growth (%) of HaCat cells exposed to the compounds (concentration range 16 to 0.125 mg/ml). (A) HaCat cells exposed to carvone; (c) HaCat cells exposed to menthone; (C) HaCat cells exposed to menthone; (C) HaCat cells exposed to menthone; (D) HaCat cells exposed to pulegone. \*Statistically significant (p< 0.05). Dunnett, ANOVA.

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Vol. 10(10), pp. 211-233, October 2016

DOI: 10.5897/AJPS2016.1439 Article Number: 87758C961079

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### Full Length Research Paper

# Useful plant species diversity in homegardens and its contribution to household food security in Hawassa city, Ethiopia

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Received 28 June, 2016; Accepted 31 August, 2016

The study was conducted on homegardens of Hawassa city, Southern Ethiopia with the aim of documenting useful plant species; identifying the internal and external household factors related to useful plant species diversity in and around home gardens and examining its contribution to household food security and income generation. A random sample of 120 homegardens from eight sub-cities of Hawassa city was used to collect useful plant species data. Techniques used were focus group discussion, semi-structured interviews, home garden tour, market survey, free listing, priority ranking, and preference ranking. A total of 258 useful plant species were documented, of which 47.29% were ornamental plants, 29.75% food plants, and 15.89% medicinal plants. Fabaceae was the dominant family represented by 9 genera and 20 species, followed by Euphorbiaceae and Asteraceae with 17 and 16 species each respectively. Homegarden size of the study area ranged from 220 to 1235 m<sup>2</sup> with a mean size of 571 m<sup>2</sup>. The age of homegarden is ranged from 15 years old to 55 years old with a mean aged of 28. The number of species in the homegarden ranges from 10 to 45 with the mean of 23. The study indicates that home gardens are contributing to food security, income generation and livelihoods in Hawassa city through production of ornamental, food plants, fodder, medicinal, timber and construction. The study recommended that the management of useful plant species in homegardens will be scaled up and further expanded and assisted by agricultural extensions in urban areas in Ethiopia.

Key words: Urban home garden, plant species diversity, household livelihood, food security.

#### INTRODUCTION

Homegardens are production system of diverse crop plants, which is easily accessible and adjacent to household (Sunwar et al., 2006). It is the site of highest species diversity where several landraces, cultivars and rare/endangered species have been maintained and

conserved (Watson and Eyzaguirre, 2002). The compositions of crops grown in home gardens can be grouped based on function as ornamental, fruits, food crops, vegetables, medicinal, spices and fodder, building materials and fuel woods (Kumar and Nair, 2004).

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Homegarden systems provide an additional food supply and cash income for the people (Das and Das, 2005).

Worldwide, homegardens are a community's most adaptable and accessible land resources and important components in reducing vulnerability and ensuring food security (Buchmann, 2009). The features of homegardens are year round production of food, decreased risks of production failure due to high diversity of species, increased resource productivity over time, expansion of the amount and quality of labour applied in the farm, provision of output flexibility and alternative production (Senanayake et al., 2009).

Homegardens in Ethiopia may broadly be categorized into two types (Zemede, 2001; Tesfaye, 2005). The first category of home gardens is small-sized gardens in which vegetables, spices, oil seeds and fruits are cultivated to supplement cereals and pulses raised in adjoining fields. This type of gardens is characteristic of cereal crop based farming areas of the country and is also found in urban centers. The other type of homegardens, which is characterized by a diverse mixture of crop plants with enset (Ensete ventricosum) making the basic framework, is that found in the south and southwestern part of the country. Advocates of gardening cite evidence that home gardening can be a sustainable strategy for improving food security and incomes when gardens are well adapted to local agronomic and resource conditions, cultural traditions and preferences (Midmore et al., 1991; IIRR, 1991). Plant diversity is often used as a measure of health of biological system (Naeem, 2002). It is threatened by the agricultural expansion, deforestation, and development activities including rapid urban expansion (Ricketts and Imhoff, 2003). Urbanization is one of the recent important issues in the enormous reduction of plant diversity. Currently the world urban population (3.2 billion) exceeds the number living in rural areas. People create rapid demands for food, settlements, jobs, waste management, and all basic needs for living (Rizvi, 2007). Dense settlements, traffic congestion, air and soil pollution, and waste dumps, reduce the space for plants, especially natural domestic plants (Mckinney, 2002).

Although urbanization is a global phenomenon, its magnitude differs widely among regions (Reid, 1998). In Ethiopia, cities are currently growing rapidly. Hence addressing the global problem of reversing plant diversity in urban areas requires multiple innovative ways. Urban and suburban home gardens play a major role in providing food, breeding sites, shelter for animals and plants also modifying microclimate (Smith et al., 2006).

In the present study most of the useful plant species diversity are almost lost by human impact and hence, there is glaring loss of biodiversity, disruption of indigenous knowledge, practices and culture are becoming evident due to limited integration of traditional practices and modern science in the study area, and the value of traditional home gardening in the conservation

and management of useful plant species by indigenous people of Hawassa city is minimal and there is a problem of food insecurity in and around Hawassa city (Reta, 2013). Thus, the purpose of this study was to document, identify the internal and external household factors related to useful plant species diversity in and around home gardens of Hawassa city and examining its contribution to the household food security.

#### MATERIALS AND METHODS

#### Study area

The study was conducted in homegardens of Hawassa city (07°05' latitude North and 38°29' longitude east) with an altitude of 1680 m above sea level and covers total area of 157.2 km<sup>2</sup> and has a mean annual rainfall and temperature of 953.4 mm and 20.3°C, respectively (SNNPRS, 2005). Hawassa is the capital city of Southern Nations, Nationalities and Peoples Regional state and Sidama zone, located 273 km from Addis Ababa, capital of Ethiopia. It is surrounded by Lake Hawassa in the west, Hawassa zuria woreda in the south and east part and Oromiya Region in the north. Based on figures from CSA (2007), Hawassa city has an estimated population of 304,479; i is home to about more than 50 ethnic groups. Each ethnic group has their own composition of tribes with distinctive language, custom, traditional beliefs and cultural diversity. It is sub divided in to 8 sub city, namely Tabore, Hayekdar, Menaharia, Misrak, Bahale adarash, Addis Ketema, Mehale Ketema and Awela Tula in which the present study was carried out and 32 kebeles (Figure 1). The land form is plain with reddish volcano soil which is ideal for construction.

#### **Data collection**

The study of homegarden was carried out in the Hawassa city in 2014. Field work was conducted during the period from February 2014 to September 2015. Each site was visited three times including the reconnaissance survey. Techniques used were homegarden tour, complete plant inventory, focus group discussion, semi-structured interviews, free listing, market survey, priority ranking, and preference ranking. The interview and discussions was conducted in Amharic language and translated into English language during data analysis. Ethno botanical techniques were employed to collect data on knowledge and management of home garden plants used by people in Hawassa city as described in Martin (1995) and Cotton (1996). A total of 120 home gardens were randomly selected from seven sub cities (17 homegardens from each sub city). Forty five homegardens (6 from each sub city) were preferentially selected for detailed study, which represented 37.5% of the garden visited. The distance between each home garden was 300 m apart. During the different visits to the households semistructured interviews with both household heads were conducted on different aspects: Categories of use of plants in the garden: preferred useful plant species by home gardeners, planting, consumption, income they get, benefits and source habitats of spicy plants; history of the garden, observed change in home garden composition; perception and valuing of diversity; local resource use pattern, challenges and constraints and categorization and local religious practices. Information obtained was recorded and coded for latter analysis.

#### Data analysis

Descriptive statics such frequency and percentage was used for

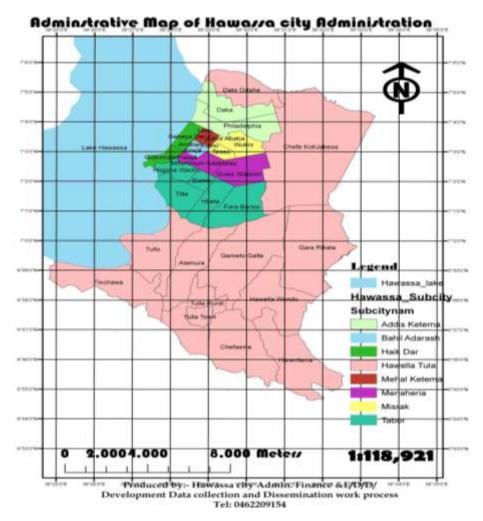


Figure 1. Map of the study area showing the study site (HCA, 2011).

analysis and summarizing the data. The diversity indices, Shannon-Weaver index (SWI), Evenness index and Simpson's index were employed to determine the species richness, evenness and dominance of the species in the homegardens. Free listing was used by asking participants to list the name of all useful plants found in their homegardens and the uses of each plant. Jaccard's similarity coefficient (JSC) was estimated for comparing homegardens number of species in eight purposively selected study areas in different regions of the country. The formula used was JSC = c/(c+a+b), where, a = number of species found in the study area but not in other study site, b = number of species absent in the study area, and c = number of species common to the study area (Jaccard, 1912).

#### **RESULTS**

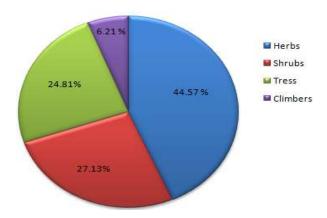
#### Useful plant species diversity

In the surveyed homegardens a total of 258 useful plant species were observed and identified, including 14(5.43%) vegetable plant species, 23 (8.92%) fruit plant species, 15(5.81%) spices plant species, 12(4.65%) root and tubers plant species, 8(3.1%) cereals, pulses and oil

seeds plant species, 3(1.16%) stimulant plant species,12(4.65%)fragrant plant species,122(47.29%) ornamental plant species, 39(15.12%) firewood plant species, 4(1.55%) animal feed plant species and 41 (15.89%) medicinal plant species. The average plant species per household was 21 ranging from 10 to 45 throughout the homegardens.

A total of 258 plant species belonging to 186 genera and 76 families were inventoried from home gardens of Hawassa city. Fabaceae was the highest number of species followed by Euphorbiaceae and Asteraceae. The genera represented by the highest number of species were Euphorbia (8 species) followed by Astera 7 species. Out of the 258 useful plant species 244 species were Angiosperms, 10 species were Gymnosperms and 4 species were Pteridophytes.

Multipurpose trees showed the highest and the most frequent occurrence (Appendix I and II). Useful species such as Cordia africana, Moringa stenopetala, Melia azerdarch, Croton macrostachys, Calpurea aurea were showed the highest frequency. The most cultivated useful



**Figure 2.** Percentage of useful plant species by their habits in the study homegardens.

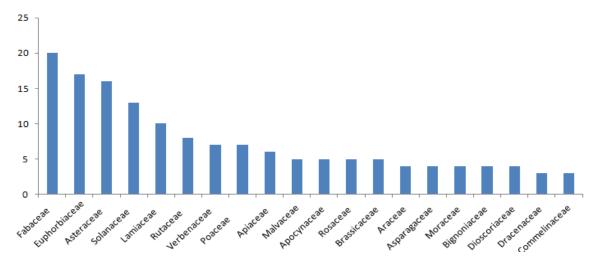


Figure 3. Top 20 useful plant species family diversity in the homegardens of Hawassa city.

food plant species in the homegarden were *Enset* ventricosum, Carica papaya, Persea americana, Coffee arabica, Musa paradisca, Mangifera indica and Zea mays.

Most species (83% of identified) were cultivated, 5% were both cultivated and wild, 12% were wild plants. Most home garden species were perennials (83%), annuals (15%) and biennials (2%). Among 258 species, 64 species (24.8%) were trees, 70 (27.13%) shrubs, 115 (44.57%) herbs, 16 (6.2%) climbers (Figure 2). Herbs were the most numerous species in the homegardens studied followed by shrubs (27.13%), trees 64(24.8%) and climbers 16(6.2%). Among 258 useful plant species recorded, 83(32.17%) was native to Ethiopia, 15(5.81%) was unidentified, 160 (62.02%) was introduced from other parts of the world.

Family wise distribution shows that Fabaceae is the most dominant family with 20 species; Euphorbiaceae is the second dominant family with 17 species and

Asteraceae is the third dominant family with 16 species followed by Solanaceae (13 species), Lamiaceae (10 species), Rutaceae (8 species), Verbenaceae and Poaceae (7 species each), Apiaceae (6 species); Malvaceae, Apocynaceae and Rosaceae (5 species each). The twenty most dominant families represent 150(27.9%) of the total number of species recorded. Top 20 useful plant species families in the home garden of Hawassa city are given in Figure 3.

#### Food plants

The food plant constitutes 72(27.91%) species of the total useful plant flora of Hawassa city home garden. Among food plants fruits comprises 23(31.94%), spices 15(20.83%), vegetables 14(19. 4%), root and tuber crops 12(16.7%), cereals, pulses and oils 8(11.11%). A high number of food plants belonged to the Dioscoriaceae,

Lamiaceae and Rutaceae (8 species each) followed by Apiaceae and Fabaceae (5 species each), Araceae, Euphorbiaceae and Verbenaceae (4 species each), Asteraceae, and Myrtaceae (3 species each). The most widely distributed food crops are Zea mays with a frequency of occurrence (118), E. ventricosum (117), M. paradisiaca (115), M. indica (99), C. papaya (97), P. americana (89), Sccharum officinarum (85) and Brassica rapa (84) respectively (Appendix II). The majority food crops cultivated are used for household consumption. Fruit species commonly found in the study homegardens are Papaya (Carica papaya), Banana (M. paradisiaca), Avocado (P. americana), Guava (Pisdium guajava), Mango (M. indica), and Roman (Punica granatum).

#### **Ornamental plants**

The ornamental plant use category consisted of 122 species from which 18(14.75%) are native to Ethiopia, 104(85.25%) is exotic. The ornamental plant species are distributed among 73 families with Euphorbiaceae, Lamiaceae (12 species each) and Asteraceae (11 species each) presented the largest number of species corresponding to 30.7% of the total ornamental plants found in the homegardens. Most of the plants surveyed in the homegardens of Hawassa city are exotic and widely disseminated throughout Hawassa city. Ornamental plants are found in more than 87% of home gardens. The most frequently distributed ornamental plants are Melia azedarch, Jacaranda mimosifolia, Cupressus Iusitanica, Callistemon citrinus, Hibiscus rosa-sinensis, Senna spectabilis. Duranta repens. Duranta erecta. Nerium olander, Terminalia Bougainnvillea glabra, mentalis, Araucaria heterophylla, Thevetia peruviana, cupercus lustantica and Ficus benjamina.

The homegardens consisted of 122 (47.29%) of ornamental plants. Among these 208 were perennials plant species. Mean number of ornamental plant species in the homegardens was 15 with the range of 10 to 35 for all surveyed households. Euphorbiaceae contained the highest number of ornamental species (12), Asteraceae is the second number of ornamental species with 11 species, Verbenaceae and Malvaceae contained 5 species each, while Asparagaceae, Lamiaceae, Apocynaceae and Bignoniaceae contained 4 species each.

#### **Medicinal plants**

A total of 41 plant species with medicinal value were recorded and this accounted for 15.89% of the total plant species documented. Species of family Asteraceae and Solanaceae were the most used for remedies representing nearly 24.39% of all medicinal plants. The majority of medicinal plants are herbs 16 (39.02%)

followed by trees 15(36.58%), shrubs 9(21.95%), climbers 1(2.44%). The most frequently utilized plant parts were leaf 22(53.66%), stem 8(19.51%) followed by root 6 (14.63%). Ninety seven percent of medicinal plants documented in the study area are indigenous. Top ten medicinal plants species occurred in more than 50% of the homegardens, namely Achranthes aspera, C. papaya, Artemisia absinthium, Artemisia afra, Ocimum lamiifolium, Withania somnifera, Vernonia amygdalina, Ruta chalepensis, Croton macrostachyus and Cucumis ficifolius (Appendix III).

#### **Spices**

A total of 16 spices plant species were documented. It is distributed among 8 genera and 9 families. Spices plants consisting of 6.202% of the total useul plant species documented. A high number of spices belonged to Lamiaceae (5 species). Alliaceae (2 species). Solanaceae (2 species), verbenaceae (2), and Rutaceae and Brassicaceae (1 species each). The most commonly used spices were Allium sativum (Onion), Allium cepa, R. chalpensis, Zingiber officinale, Rosmarinus officinalis, Ocimum basilicum, Becium filamentasum and Brassica nigra.

#### Fragrant, stimulants and fodder plant species

A total of 12 species of fragrants, 3 stimulants and 4 fodder species were documented. The three use categories togeother consisted of 7.36% of the total useful plant species documented. The five most commonly used fragrant plant species in the majority of homegardeners were Olea europea, Cympogen citrates, Lippia adoensis, A.abysinthium, and A. abyssinica.

#### **Timber (furniture) plants**

Timber plant species constitue 29 plant species which accounted 11.4% of all plant species documented. Timber species which occured in more than 50% of the homegarden namely Melia azedarch, Grevillea robusta, Cupresuss lustanica, Cordia africana, Casuarina equisetifolia, Acacia melanoxylon. Among 29 plant species recoreded 17 were indigenous plants which were highly treated in the forest namely Prunus africana, Hagenia abyssnica, Juniperus procera, Podocarpus falcatus, O. europea, Celtis africana, and Aningeria adolfi friedericii (Appendix IV and V).

The highest Shannon-Wiener Diversity Index (H') of useful plant species was recoreded for Tabor sub city (H' = 5.87) followed by Haik dar subcity (H' = 3.80) and the lowest diversity index was recorded at Menhara sub city (H' = 2.77) (Table 1).

Study sites	Species richnes	Shannnon's index(H')
Haik dar	45	3.80
Tabor	48	3.87
Misrak	35	3.55
Addis ketema	30	3.40
Bahladrash	27	3.29
Mehal ketema	28	3.33
Menhara	16	2.77

Table 1. Shannon-Wiener Diversity Index (H') for seven study sites.

**Table 2.** Jaccard's similarity coefficient for comparing homegardens number of plant species composition in the homegardens of Hawasa City with other areas of Ethiopia.

Study site	Sabata town	Holeta town	Arba minch zuria	Sidama zone	Basketo and Kefa	Gedeo zone	Wolayta zone	Selected areas of Amhara
Total number of species	135	112	133	198	224	165	159	85
Common species	120	106	70	120	50	130	58	30
JSC	0.465	0.421	0.272	0.44	0.12	0.443	0.22	0.096
Percentage similarity	46.5	42.1	27.2	44	12	44.3	22	9.6
Source	Habtamu and Zemede (2011)	Mekonen etal. (2014)	Belachew etal. (2006)	Tesfaye (2005)	Feleke (2011)	Solomon (2011)	Talemos et al. (2013)	Fentahun (2008)

The highest vaues of Jaccard's Coefficient of Similarity index (JCS) indicate a higher similarity in homegarden species diversity. The JCS result indicates that homegardes of Hawassa city was the highest similarity with homegarden composition of Sabata town (JCS = 0.46), Gedeo zone (JCS = 0.43) and Sidama zone (JCS = 0.44). Home gardens of selected areas of Amhara (JCS = 0.096), Basketo and Kefa (JCS = 0.12) and Wolyta (0.22) showed the weakest similarity coefficient (Table 2).

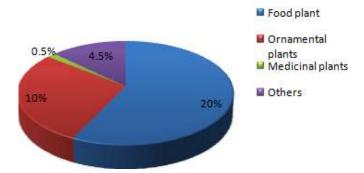
# Contribution of urban homegarden to household food security

In Hawassa city, the role of homegarden for cash income generation and house hold consumption was highly increased particularly in Haikidar sub-city, while it is decreased at the center of the city. The ornamental function of home gardens increased particularly in the center of the city, where 50% of the gardeners mentioned decoration as the main function of their gardens in the study survey. About 40% of the respondents report that home garden is a source of their income. Ten percent of them reply that homegarden is a supplementary source of their income and 50% use homegarden as a place of enjoyment. Poverty and unemployment is high in Hawassa city, most youth rely on cultivation of ornamental plants to generate income by selling ornamental plants to support their families at road side of

the city. About 75% of the homegardeners explained that they conserve useful plant species for foods, 10% for income generation, 25% for pleasure, 25% for medicinal use, 15% for construction and other livelihood needs. The study showed that the majority of homegardeners are under food insecurity especially the poor urban dewellers. Food security assessment survey indicates that 25% of the homegardeners were found food secured throughout the year, 15% of the gardeners are food secured only for six months. The poor homegardeners attained food security through production in their own garden but the reach homegardeners purchase from local market.

The homegardens contributions to household's annual income was 35% of the total income, among which 20% from food plants, 10% from ornamental plants, 0.5% from medicinal plants and 4.5% from others (Figure 4). Araucaria heterophylla is the most expensive ornamental plant species sold in the market. One plant of A. heterophylla is sold at 500 to 1500 ETB (\$24 to 72). Terminalia mentalis is the second expensive ornamental plant sold. One plant species of T. mentalis is sold as up to 250 to 500 ETB (\$12 to 24). Medicinal pants are no direct income to households. Poor urban women are preparing E. ventricosum corm kocho for food security (Figure 5).

Only a few homegardeners has sufficient food for a year. The homegardens in the Hawassa city only contributes 10% fresh vegetables. Livestock and poultry farming in the homegardens also another source of



**Figure 4.** Percentage of contribution of categories to the total income earned from food, ornamental, medicinal and other use categories.



**Figure 5.** Women pulverizing *Ensete ventricosum* corm in Kocho preparation for food security in Hawassa city home garden near Haikdar.

income generation contributes 15%, cow milk (10%), poultry (15%), pig (0.5%) chickens (5%), ducks (0.5%). Sugar cane (S. officinarum), Kocho (E. ventricosum), Muzi (M. paradisiaca) accounted for about 35% of the homegardens income contribution. Income from homegarden increases an average household income from 1177 to 4580 Birr.

Preference ranking of top ten useful food plant species by home gardeners for household income generation shows that *M. paradisiaca* is the most preferable food crops in the first rank with a score of 120 with maximum yearly income generation of 15000 Ethiopian Birr (ETB), *S. officinarum* is the second with a score of 117 with yearly income generation of 10000 ETB and *E. ventricosum* and *Zea mays* are the third and forth places with income generation of 6000 and 5500 ETB respectively (Table 3).

The categories of use identified are ornamental,

medicine, food, firewood, timber, construction, fodder, spices, fragrant and others. A total of 71 species are reported as having one use type, 103 species having two use types and 86 species with three use types. The most numerous species were ornamental 122 species followed by food crops, medicinal plants, fuel wood and constructions (Figure 6).

#### Gender role in the homegarden management

The management of homegardens includes tree planting, watering, weeding and fencing. The homegardeners maintain their homegarden soil fertility by using animal manure and leaf litter. Both men and women are involved in the management of homegardens. Mostly the old aged people are spent most of their time in the management of homegardens in the Hawassa city.

C/NI	Household use rai	Yearly income generation(ETB)						
S/N	Scientific name	Total score	Rank	minimum	maximum	Rank		
1	Lactuca sativa	99	9 <sup>th</sup>	500	2500	5 <sup>th</sup>		
2	Brassica rapa	84	4 <sup>th</sup>	300	2000	6 <sup>th</sup>		
3	Musa x paradisiaca	120	5 <sup>th</sup>	5000	15000	1 <sup>st</sup>		
4	Saccharum officinarum	117	$3^{rd}$	1000	10000	2 <sup>nd</sup>		
5	Ensete ventricosum	114	2 <sup>nd</sup>	1500	6000	3 <sup>rd</sup>		
6	Persea americana	80	6 <sup>th</sup>	250	900	7 <sup>th</sup>		
7	Carica papaya	79	10 <sup>th</sup>	200	850	8 <sup>th</sup>		
8	Mangifera indica	69	8 <sup>th</sup>	370	600	9 <sup>th</sup>		
9	Zea mays	110	1 <sup>st</sup>	2500	5500	4 <sup>th</sup>		
10	Dioscorea sagittifolia	55	7 <sup>th</sup>	150	450	10 <sup>th</sup>		

**Table 3.** Top ten ranking food crops of HG as determined by preference ranking with income generation.

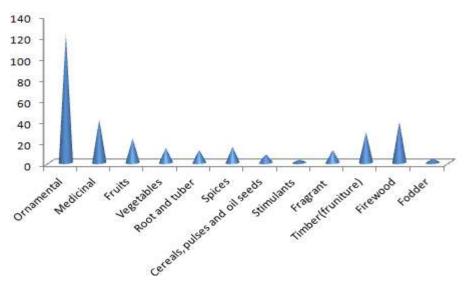


Figure 6. Categories of uses identified in the homegardens of Hawassa city.

Females managed 47% of useful plant species diversity by planting, watering, weeding and selling while males managed 53% by cultivation of food crops, ornamental, medicinal plants, fencing, digging, designing, searching seeds and other useful plants (Figure 7).

Most youth participated in the cultivation of ornamental plants near road side of the city for income generation (Figure 8).

### Constraints of homegardens in Hawassa city

According to the semi structured interview report the main constraints of homegardens of the study area were knowledge gap in plant breeding (66.7%), lack of planting materials and seeds (63.3%), lack of agricultural support system (57.5%), and lack of awareness (55%) respectively (Table 4).

The main source of planting materials in the study homegardeners are market (45%), cultivating in their homegardens (20.83%) and from relatives (16.67%). Agricultural office, local and international NGOs are the least source of planting materials (Table 5).

### **DISCUSSION**

A total of 258 species (64 trees, 70 shrubs, 115 herbs and 16 climbers) belonging to 186 genera under 76 family were documented. In Hawassa city, more than 50 ethnic groups are living which have different language, culture, custom, beliefs and religion. Cultural diversity in Hawassa city helps to conserve useful plant species biodiversity in homegardens. Different ethnicity, culture and religion make a unique plant species diversity in the homegardens of Hawassa city (Reta, 2013). Sthapit et al.



Figure 7. The role of Men and Women in the management of Home garden in Hawassa.



Figure 8. One of the youth selling ornamental plants at the road side in Hawassa city.

**Table 4.** Challenges of homegarden with percentage distribution with frequency (n = 120).

Challenges	Frequency	Percentage	Rank
Knowledge gap in plant breeding	80	66.66	1
Lack of planting materials &seeds	76	63.3	2
Lack of agricultural support system	69	57.5	3
Lack of awareness	66	55	4
Water lodging during winter season	45	37.5	5
Lack of water availability	35	29.17	6
Destruction by animals	21	17.5	7
Disease infestation	20	6.66	9
Lack of access to land (Size of home garden)	18	15	8

Source	No. of respondents	Percentage
Market	54	45.0
Relatives	20	16.67
Neighbors	10	8.33
Cultivate in their homegarden	25	20.83
Agricultural office	6	5.0
NGOs	5	4.17

**Table 5.** Source of plant materials in the homegardens of Hawassa city.

(2004) showed that the composition of unique plants in homegardens varies with ethnicity, food culture, religion and spirituality. The total numbers of species recorded in the homegardens of Hawassa city are greater than number of species reported from other parts of Ethiopia. For example, Feleke (2011) reported 224 plant species from homegardens of Basketo and Kafa, Sothern Ethiopia; Mathewos et al. (2013) reported 214 plant species from homegardens of Dwaro zone, southern Ethiopia: Tesfave (2005) reported 198 plant species from homegardens of Sidama, Southen Ethiopia; Solomon (2011) reported 165 plant species from Gedeo zone. The present study reported 72(27.9%) of food plant species from the total record of 258 useful plants species from homegardens of Hawassa city. Zemede (1997) reported about 126 (75% of the total record) plant species used as food from Ethiopian homegardens. Moreover, Belachew et al. (2003) and Habtamu (2008) reported 48 and 37 edible plant species from homegardens of Arbaminch and Sebeta areas respectively. Solomon (2011) identified about 68 plant species used as food from homegarden areas in Kochere Wereda. Feleke (2011) and Mathewos et al (2013) reported 102 and 77 food plants from homegardens of Basketo and Dwaro zone, respectively.

Urban homegardens are sources of food crops, vegetables, cereals, pulses, fruits, spice, milk and livestock etc. Therefore, it is important contributors to household food security of poor urban dewllers and the rich ones also. Urban homegarding is one of the best methods for food production which feed high population, as food security is a major concern in many parts of the world and in many of urban and rural areas of Ethiopia. Urban home garden is a future promising agricultural activity that reduces urban food insecurity. In Ethiopia, cities are not practicing urban agriculture even though there are enough free spaces in and around city gardeners. Mohammed (2002) reported that Ethipia is the country where existence and significant contribution of urban agriculture was not only disregarded and unrecognized by researchers but also underestimated and given very little attention by urban development studies.Limited homegarden size available for gardeners make them to grow different homegarden species. In the present sudy the majority of home gardens even though they have large home garden size, they contain low number of species diversity due to lack of knowledge gaps on cultivation, management and conservation of useful plant species.

The total number of species in a single homegarden was found to be a maximum of 45 with more than 85% households having the species numbers up to 10 to 45. The highest number of species was highest in the Haikdar sub-city and Tabor sub-city respectively (45 and 35 species) because there is sufficient irrigation water and large garden size in Hayikdar sub city and large home garden size in Tabor sub-city. The home gardens was the richest as more than 60% of the home gardens had more than 30% species per homegarden and Bahladarash, Mehalketema, Menaheria sub cities are the poorest species diversity where more than 65% homegardens had less than 20 species in home garden. In Ethiopia, there are very few studies on useful plant species diversity and its contribution to food security in urban homegardens. Many studies on home gardening and plant diversity have concentrated on rural areas (Das and Das, 2005). There are very few systematic studies on domestic garden diversity in urban or sub urban areas (Smith et al., 2005). Urban homegardening ensures households food security by providing vegetables, fruits, medicinal plants, fuel wood, ornamental plants, fodders, construction materials, root and tubers. About 25% of the respondents reported that annually they earned a high income of 10,000 and 25,000 ETB from selling various products of useful plant species.

The most useful plant species cultivated in the home gardens of Hawassa city were ornamental plants (47.29%) and food plants (27.91%). Ornamental plant species are the most diversified, abundant and species rich use category. Similar study was report from homegardens of Tlhakgameng in which 57% were ornamental plants and 27% were food plants (Molebatsi, 2011). Cilliers (2010) also reported 28% of food plants in Ganyesa home gardens. This shows that most poor peoples in urban areas are largely dependent on cultivation of ornamental plants for aesthetic value, selling to sustain their livelihood and food plants for consumption purposes.

According to Nair (1993), the high number of ornamental plants is associated with the aesthetic role of home gardens in cities, since they are not used for

subsistence in urban areas except among low income populations (Ninez, 1984). The number of ornamental plants has increased in areas near, as well as in urban areas in response to the process of modernization and the large supply of these plants in cities (Moura and Andrade, 2007).

The major contributor to diversity of urban environments is horticultural floras which are mostly characterized by ornamental plants and vegetables (Gaston et al., 2005, Marco et al., 2008). The most cultivated crops in the homegarden were *E. ventricosum*, *C. papaya*, *P. americana*, *C. arabica*, *M. paradisca*, *P. guajava* and *M. indica*. *Zea mays* was the most widely used cereal crops in the homegardens of Hawassa city as it occurred in 85% of sampled households.

Within home garden the number of species per homegarden ranged from 10 to 45 and the mean was 21. Similar research reports on the number of species in home garden of different areas by different researchers for instance, Kabir and Webb (2009) reported 419 species of plants with an average of 34 species per household across 402 homegardens from Bangladesh. Mendez et al (2001) reported a total of 324 species with nine different uses from Nicaragua with an average of 70 species perhomegarden. Tynsong and Tiulari (2010) reported 197 plant species with an average of 89 plant species per homegarden average size of 750 m<sup>2</sup>. Tesfaye et al. (2010) reported 78 cultivated crops within 44 homegardens from Sidama southern Ethiopia with 16 as an average number of species per farms. Mekonnen et al. (2014) reported 112 plant species in the homegardens of Holeta town with the mean of 22 species perhomegarden.

Olajide-Taiwe et al. (2010) reported 36 plant species in homegarden from Ibadan, Oyo state. The total number of species and average number of species per homegarden in the present study was less compared to the previous report. Fabaceae had the highest number of species recorded in the homegardens study.

The dominance of Fabaceae was reported from other homegarden studies in Ethiopia (Tefera, 2010; Mekonnen et al., 2014). This may indicate that homegardeners mostly cultivated Fabace for food security purposes.

The present study agrees with many previous researches finding on significance of homegarden to household food security. For examples, Olajide-Taiwode et al. (2010) reported 36 plant species from Ibadan, Oyo state showed that homegarding increased family supply. Maroyi (2009) reported 69 plant species from Nhema, Zimbabwe indicated homegarden as important for poor households to overcome adversity and meet basic needs. Tynsong and Tiwari (2010) finding from Meghalaya, India showed that homegarden contributed 7% of the total household income.

Tesfaye (2005) found that richness is positively related with household income, evenness of species is low in homegarden owned by rich household compared to that

of poorer households. Kumari (2009) has argued that the higher the household expenses, the higher the food plant density and the lower the total plant diversity. The same author has observed that rich households in urban areas tend to plant more ornamental plants with higher economic values in their home gardens (Kumari, 2009).

### Conclusion

The homegardens of the study area is home for many useful plant species diversity. These useful plant species are a great value for household income generation, food security, medicinal, ornamental, and other non food livelihood needs of poor urban dewellers. The present sudy indicates that high useful plant species diversity documented in the homegarens of Hawassa city was associated with diversity of ethinicity with different language, culture, custum and beliefs. In addition to this, Hawassa city is the fastest growing city in Ethiopia. This also have ement contribution to high useful plant species diversity in the area. The poor urban dewellers are highly interested in homegarden activities to sustain their livelihoods. The number of ornamental plant species diversity in the study area is higher. This shows that urban homegardeners gave more pirority for ornamental plant cultivation for aeshetic value. The rich people have not shown much interest in the cultivation of food crops eventhough they have large homegarden size. The rich people gave more pirority for conservation and management of ornamental plant species while the poor urban dewellers gave more pirority for conservation and management of food crops to sustain their livelihoods. Proper management of homegardens has a great potential for biodiversity conservation, improving food security and provides contribution for ecosystem services in the study area. The present study indicates that there is a knowledge gap in the cultivation, conservation and management of useful plant species in homegardeners. Therefore, incorporating indigenous knowledge with scientific management and conservation of useful plant species, creating awarness among urban dewellers', will promote urban agriculure in Ethiopia ingeneral and Hawassa city in particular.

### **Conflict of Interests**

The authors have not declared any conflict of interests.

### **ACKNOWLEDGEMENTS**

The author is highly grateful to the urban homegarden owners for allowing the study as well as sharing their knowledge and time without which the study was impossible. He extends his thanks to his friend

Hayimanot Tao for helping in data collection and taking picture of the study sample. The reviewers are highly acknowldge for their critical comments to improve the paper.

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Appendix I. List of ornamental plants, their local name, family, status, duration and frequency of occurrence (n=120).

No.	Scientific name	Family	Local name	Habit	Fr.	Status	Duration
1	Acacia melanoxylon R.Br.	Fabaceae	Omedlla(A)mh	Tree	67	С	Р
2	Acalypha wilkesiana Mull. Arg.	Euphorbiaceae	Abeba	Shrub	23	С	Р
3	Achillea millefolium L.	Asteraceae	Abeba	Herb	12	С	Р
4	Agave americana L.	Agavaceae		Shrub	14	С	Р
5	Agave americana var marginata	Agavaceae		Shrub	25	С	Р
6	Alcea rosea L.	Malvaceae	Abeba	Herb	45	С	Р
7	Aloe vera L.	Aloaceae	Argisaa	Herb	8	С	Р
8	Aloe gilbertii Reynolds	Aloaceae	Argisaa	Herb	7	С	Р
9	Alocasia macrorrhizos (L.)G.Don	Araceae	Elephant ears	Herb	29	С	р
10	Araucaria heterophylla (Salisb.)Franco	Araucariaceae	Yeferejitid	Tree	10	С	Р
11	Arundinaria alpina K. Schum.	Poaceae	Kerkeha	shrub	5	С	Р
12	Asparagus setaceus (Kunth) Jessop	Asparagaceae	Seriti	CI	13	С	Р
13	Asparagus africanus	Asparagaceae	Seriti	CI	15	С	Р
14	Azadirachta indica A.Juss.	Meliaceae	Neem	Tree	6	С	Р
15	Bougainvillea glabra Choisy	Nyctaginaceae	Bugambe	Shrub	24	С	Р
16	Bougainvillea spectablis Willd.	Nyctaginaceae	Bugambe	Shrub	20	С	Р
17	Brugmansia x candida Pers.(Pro.sp)	Solanaceae	Angel's trumpets	Shrub	18	С	Р
18	Calathea zebrina (Sims)Lindl	Marantaceae		Herb	36	С	Р
19	Callistemon citrinus(Curtis) Seekls	Myrtaceae	Bottle brush	Tree	47	С	Р
20	Canna indica L.	Cannaceae	Siet-akuri	Herb	35	С	Р
21	Canna x generalis L. H. Bailey	Cannaceae	Enset abeba	Herb	10	С	Р
22	Chrysanthemum coronarium L.	Asteraceae	Abeba	Herb	43	С	Α
23	Casuarina equisetifolia L.	Casuarinaceae	Shewshewe	Tree	76	С	Р
24	Casuarina cunninghamiana	Casuarinaceae	Shewshewe	Tree		С	Р
25	Catharanthus roseus (L.)G.Don	Apocynaceae	Abeba	Herb	37	С	A/P
26	Ceiba pentandra (L.) Gaertn	Bombacaceae		Tree	8	С	Р
27	Centella asiatica (L.) Urb.	Apiaceae		Herb		С	Р
28	Clerodendrum myricoides (Hochst.) Vatke.	Lamiaceae		Herb	6	С	Р
29	Codiaeum variegatum (L.)A.Juss.	Euphorbiaceae	Masincho	Shrub	22	С	Р
30	Codiaeum spp.	Euphorbiaceae	Masincho	Shrub	21	С	Р
31	Combretum collinum Fresen	Combretaceae		Tree	5	W/C	Р
32	Cordyline terminalis	Agavaceae		Herb 26		С	Р
33	Cosmos bipnnatus Cav	Asteraceae	Abeba	Herb	7	С	Р
34	Crassula schimper Fisch. & Mey.	Crasulaceae	Abeba	Herb	13	С	Р
35	Croton gratissimus Burch.	Euphorbiaceae	Masincho ferenje	Shrub	21	С	Р
36	Cupressus Iusitanica Mill.	Cupressaceae	Homme	Tree	74	С	Р

# Appendix I. Contd.

37	Cyperus bulbosus Vahl	Cyperaceae	Kunti	Herb	11	С	Р
38	Cyperus rotundus L.	Cyperaceae		Herb		С	Р
39	Dahlia pinnata Cav.	Asteraceae	Abeba	Herb	10	С	Р
40	Datura metel L.	Solanaceae		Herb	5	С	A/P
41	Delonix regia (Boj.ex Hook.)Ref.	Fabaceae	Yedirezaf	Tree	38	С	Р
42	Dianthus caryophyllus L.	Caryophyllaceae	Abeba	Н	9	С	Р
43	Dracaena afromontana	Dracaenaceae	Abeba	Tree	5	С	Р
44	Dracaena steudneri Engl.	Dracaenaceae	Lanticho	Tree	28	С	Р
45	Dracena sanderia	Dracaenaceae	Happy plant	Herb	47	С	Р
46	Duranta erecta L.	Verbenaceae	Sky flower	Shrub	49	С	Р
47	Duranta erecta aureo-variegata	Verbenaceae		Shrub	20	С	Р
48	Duranta repens L.	Verbenaceae		Shrub	48	С	Р
49	Duranta repens Linn.var.variegata	Verbenaceae		Shrub	44	С	Р
50	Epipiremnum aureum (L.)Engi.	Araceae		CI	3	С	Р
51	Euphorbia antiquorum L.	Euphorbiaceae		Herb	6	С	Р
52	Euphorbia cotinifolia L.	Euphorbiaceae	Duumo daraaro	Shrub	15	С	Р
53	Euphorbia baioensis S.Carter	Euphorbiaceae		Herb		С	Р
54	Euphorbia griffithii Hook.F.	Euphorbiaceae		Shrub	9	С	Р
55	Euphorbia myrsinites L.	Euphorbiaceae		Herb	13	С	Р
56	Euphorbia pulcherrima (R.Grah.)Wild.	Euphorbiaceae	daraaro	Shrub	12	С	Р
57	Euphorbia milii (Bojerex Hook.)Ursch & Leandri	Euphorbiaceae	Ye'aklil eshoh	Shrub	8	С	Р
58	Ficus benjamina L.	Moraceae	Ornamental fig	Shrub	5	С	Р
59	Ficus elastica Roxb.	Moraceae	Yegoma zaf	Tree	6	С	Р
60	Gazania rigens var. rigens (L)Gaertn. var. uniflora (L.f.) Roessler	Asteraceae	Abeba	Herb	7	С	Р
61	Grevillea robusta R.Br.	Proteaceae	Temenjazaf	Tree	39	С	Р
62	Hibiscus acetosella Welw. ex Hiern	Malvaceae	Abeba	Shrub	11	С	Р
63	Hibiscus rosa-sinensis L.	Malvaceae	Abeba	Shrub	18	С	Р
64	Hibiscus sp.	Malvaceae	Abeba	Shrub	23	С	Р
65	Hippeastrum puniceum (Lam.) Kuntze	Amaryllidaceae		Herb	10	С	Р
66	Hypercum revolutum Vahl	Hypericceae	Garaanbicho	Shrub	2	С	Р
67	Indigofera spicata Forssk.Var.spicata	Fabaceae	Abeba	Herb	6	С	Р
68	Ipomoea purpurea (L.)Roth	Convolvulaceae	Abeba	CI	9	С	Р
69	Iresine herbstii Hook.ex Lindi.	Amaranthaceae	Abeba	Herb	39	С	Р
70	Jacaranda mimosifolia D.Don.	Bignoniaceae	Jacaranda	Tree	78	С	Р
71	Juniperus procera HochstexEngl.	Cupresaceae	Honcho	Tree	12	С	Р
72	Kalanchoe lanceolata (Forssk.)Perr.	Crassulaceae	Hanculuulle	Herb	5	С	Р
73	Lantana camara L.	Verbenaceae	Yewofkolo	Shrub	25	C/W	Р

# Appendix I. Contd.

74	Matteuccia struthiopteris (L.)Todaro	Dryopteridaceae	Ferns	Herb	45	С	Р
75	Melia azedarch L.	Meliaceae	Neem	Tree	99	С	Р
76	Mirabilis jalapa L.	Nyctaginaceae	Abeba	Herb	39	С	Р
77	Nephrolepis cordifolia (L.)Presl	Polypodiaceae	Farnii	Herb	65	С	Р
78	Nerium oleander L.	Apocynaceae		Shrub	81	С	Р
79	Oenothera biennis L.	Onagraceae		Herb	5	С	В
80	Olea europaea L.ssp. Cuspidata (Wall.ex G.Don) Cif.	Oleaceae	Ejersu	Tree	37	С	Р
81	Passiflora caerulea L.	Passifloraceae		CI	7	С	Р
82	Phalaris arundinaceae L.	Poaceae		Herb	12	С	Р
83	Phoenix reclinata Jacq.	Arecaceae	Saticho	Tree	40	С	Р
84	Pavonia urens Cav.	Malvaceae	Abeba	Herb	6	С	Р
85	Pelargonium x hortorum L.H.Bail.	Geraniaceae	Abeba	Herb	12	С	Р
36	Pelargonium zonale (L.)L'He'r. ex Aiton	Geraniaceae	Abeba	Н	13	С	Р
87	Pinus patula L.	Pinaceae	Patula	Tree	18	С	Р
38	Pinus radiata L	Pinaceae		Tree	13	С	Р
39	Plumbago auriculata Lam.	Plumbagnaceae	Abeba	Shrub	5	С	Р
90	Plumeria alba L.	Apocynaceae	Plumera	Shrub	14	С	Р
91	Plumeria rubra L.	Apocynaceae	lumera	Shrub	15	С	Р
92	Pyrostegia venusta (Ker Gawl.)Miers	Bignoniaceae	Flame vine	CI	11	С	Р
93	Rosa richardii Hart.	Rosaceae	Tsgereda	Shrub	49	С	Р
94	Salvia leucantha Cav.	Lamiaceae	Abeba	Herb	10	С	Р
95	Salvia splendens Sellow exRoem.& Schult.	Lamiaceae	Abeba	Herb	7	С	Р
96	Scadoxus multiflorus (Martyn)Raf.	Amarvllidaceae	Arfaasa	Herb	2	С	Р
97	Sanseveria trifasciata var. laurentii (DeWild.)	Asparagaceae		Herb	37	С	Р
98	Sansevieria trifasciata Prain.	Asparagaceae	Mother low's tongue	Herb	38	С	Р
99	Schefflera arboricola (Hayata) Merr.	Araliaceae	Umberella tree	Shrub	10	С	Р
100	Schinus molle L.	Anacardiaceae	Kundeberbere	Tree	12	С	Р
101	Senna siamea(Lam.)H.S.Irwin &Barneby	Caesalpiniaceae	Siamese cassia	Shrub	13	С	Р
102	Senna spectabilis (Dc.)Irwin & Barneby	Caesalpiniaceae		Shrub	18	С	Р
103	Sisyrinchium californicum KerGawler)Dryander	Iridaceae	Yellow eyed grass	Herb	16	С	Р
104	Solenostemon scutellarioides (L.)Codd	Lamiaceae	Painted nettle Coleus	Herb	49	С	Р
105	Spathodea campanulata P.Beauv. ssp. nilotica.	Bignoniaceae		Tree	8	С	Р
106	Tagetes erecta L.	Asteraceae		Herb	13	С	Р
107	Tagetes minuta L.	Asteraceae		Herb	11	С	Р
108	Tagetes patula L.	Asteraceae		Herb	9	С	Р
109	Tecoma capensis (Thunb.) Spach	Bignoniaceae		Shrub	6	С	Р
110	Tecoma stans (L.) Juss ex kunth	Bignoniaceae		Shrub	8	С	Р

# Appendix I. Contd.

111	Terminalia mentalis(T.Mantaly)	Combretaceae	Tree	39	С	Р
112	Thuja orientalis L.	Cupresaceae	Tree	11	С	Р
113	Thevetia peruviana Luckey Nut(Eng.)	Apocynaceae	Shrub	16	С	Р
114	Tradescantia pallida (Rose)D.R.Hunt.	Commelinaceae	Herb	13	С	Р
115	Tradescantia zebrina Bosse	Commelinaceae	Herb cclent	19	С	A/P
116	Tradescantia spathacea Sw.	Commelinaceae	Herb	13	С	Р
117	Tropaeolum majus L.	Tropaeolaceae	Herb	8	С	A/P
118	Vinca minor L.	Apocynaceae Abeba	Herb	17	С	Р
119	Vitis vinifera L.	Vitaceae	Liana	5	С	Р
120	Washigtonia robusta (Lindl.) H.Wendl.	Arecaceae Saticho	Tree	21	С	Р
121	Zamioculcas zamiifolia(Lodd.et al)Engl.	Araceae	Shrub	10	С	Р
122	Zephyranthes candida (Lindi.)Her.	Amaryllidaceae	Herb	7	С	Р

Appendix II. List of food plant species documented in Hawassa city homegardens.

	Vegetables							_
No.	Scientific name	Family	Local name	Habit	Fr.	Status	Parts Used	Duration
1	Allium sativum L.	Alliaceae	Nechishinkurt	Н	8	С	Bulb	Р
2	Allium cepa L.	Alliaceae	Keyishnkurt	Н	15	С	Bulb	Р
3	Amaranthus hybridus L.	Amaranthaceae		Н	11	W/C	Leaves	Α
4	Brassica carinata A.Br.	Brassicaceae	Gomen	Н	49	С	Leaves	Α
5	Brassica integrifolia L.	Brassicaceae	Yeguragegomen	Н	51	С	Leaves	Α
6	Brassica oleracea L.	Brassicaceae	Tiklegomen	Н	45	С	Leaves	Р
7	Brassica oleracea L.var.capitata	Brassicaceae	Tiklegomen	Н	32	С	Leaves	Р
8	Beta vulgaris L.	Brassicaceae	Kosta	Н	39	С	Leaves	Α
9	Lycopersicon esculentum Mill.	Solanaceae	Timaatim	Н	67	С	Fruits	Α
10	Lactuca sativa L.	Asteraceae	Selata	Н	78	С	Leaves	Α
11	Saccharum officinarum L.	Poaceae	Shonkora	Н	81	С	Stem	Р
12	Solanum nigrum L.	Solanaceae	Tunayee	Н	49	W/C	Leaves	Α
13	Solanum melongena L.	Solanaceae	Sarajan/eggplant	Н	23	С	Fruits	Р
14	Moringa stenopetala	Moringinaceae	Shifera/Halako	Т	56	С	Leaves	Р
15	Solanum tuberosum L.	Solanaceae	Dinichi	Н	21	С	Leaves	Α
Root	t Crops							
1	Beta vulgaris L.	Chenopodiacea	Keysir	Н	78	С	Root	В
2	Colocasia esculeta (L.)Schott	Araceae	Godare	Н	90	С	Root	Α
3	Dacus carota L.	Apiaceae	Karot	Н	25	С	Root	В

# Appendix II. Contd.

4	Dioscorea alata L.	Dioscoreaceae	Boyna	CI	59	С		Root	Α
5	Dioscoria bulbifera L.	Dioscoreaceae	Kotehare	CI	22	С		Root	Α
6	Dioscorea sagittifolia Pax.	Dioscoreaceae	Keyi bohina	CI	43	С		Root	Α
7	Dioscorea praehenslis Benth	Dioscoreaceae	Nechi bohina	CI	42	С		Root	Α
8	Enseteventricosum (Welw.)Cheesman	Musaceae	Wesse	Sh	117	С		Root	Р
9	Ipomoea batatas (L.)Lam.	Convolvulaceae	Maxaxisha	Н	54	С	R	Root	Α
10	Manihot esculenta Crantz	Euphorbiaceae	Cassava	Sh	29	С	R	Root	Α
11	Solanum tuberosum L.	Solanaceae	Dinich	Н	21	С	S	Stem	Α
12	Xanthosoma sagittifolium (L.)Schott	Araceae	Tikur godare	Н	73	С	R	Root	Α
Frui	t crops								
1	Ananas comosus (L.) Merr.	Bromeliaceae	Ananas	Herb	2		С	Fruit	Р
2	Annona squamosa L.	Annonaceae	Gishta	Tree	70		С	Fruit	Р
3	Carica papaya L.	Caricaceae	Papaya	Tree	97		С	Fruit	Р
4	Casimiroa edulis Laliave	Rutaceae	Kasmir	Tree	89		С	Fruit	Р
5	Citrus aurantium L.	Rutaceae	Komtate	Sh	69		С	Fruit	Р
6	Citrus medica L.	Rutaceae	Tirengo	Sh	25		С	Fruit	Р
7	Citrus aurantifolia (Christm.)Swingte	Rutaceae	Lomi	Sh	19		С	Fruit	Р
8	Citrus sinensis (L.)Osb.	Rutaceae	Birtukan	Sh	16		С	Fruit	Р
9	Cucurbito pepo L.	Cucurbitaceae	Duba	CI	59		С	Fruit	Α
10	Balanites aegyptica	Balanitaceae	Badana	Tree	21		W	Fruit	Р
11	Dovyalis caffra (Hook.f.&Harv.	Flacaurtiaceae	Koshim	Т	49		W	Fruit	Р
12	Ficus sur	Moraceae	Shola	Т	18		W	Fruit	Р
13	Mangifera indica L.	Anacardiaceae	Mango	Т	98		С	Fruit	Р
14	Malus sylvestris Mill.	Rosaceae	Apple	Т	24		С	Fruit	Р
15	Morus alba L.	Moraceae	Gora	Т	19		С	Fruit	Р
16	Musa x paradisiacal L.	Musaceae	Muzi	Sh	99		С	Fruit	Р
17	Passiflora edulis Sims.	Passifloraceae	Hopi	CI	11		С	Fruit	Р
18	Persea americana Mill.	Lauraceae	Avocado	Т	98		С	Fruit	Р
19	Prunus x domestica L.	Rutaceae	Prim	Т	2		С	Fruit	Р
20	Prunus persica (L.)Batsch.	Rosaceae	Kok	Т	9		С	Fruit	Р
21	Psidium guajava L.	Myrtaceae	Zeyitun	Т	97		С	Fruit	Р
22	Punica granatum L.	Punicaceae	Roman	Sh	30		С	Fruit	Р
23	Syzygium guineense (Willd.)Dc	Myrtaceae	Dokima	Т	2		W	Fruit	Р
	eal, pulses and oil crops	,	- <del>-</del> -	-	_		-		
1	Cajanus cajan (L.)Mill.	Fabaceae	Yewof ater	Shrub	20		С	Seeds	Р
2	Canavalia africana L.	Fabaceae	Adenguare	Herb	54		C	Seeds	Α
3	Carthamus tinctorius L.	Asteraceae	Suf	Shrub	43		C	Seeds	Α

# Appendix II. Contd.

4	Jatropha curcas L.	Euphorbiaceae	Jatroba	Shrub	14	С	Seeds	Р
5	Phaseolus vulgaris L.	Fabaceae	Boloqqie	Climber	40	С	Seeds	Α
6	Phaseolus lunatus L.	Fabaceae	Adengware	Climber	45	С	Seeds	Α
7	Ricinus communis L.	Euphorbiaceae	Gulo	Shrub	52	C/W	Seeds	Р
8	Zea mays L	Poaceae	Badala	Herb	118		Seeds	Α
List	of spices							
1	Allium sativum L.	Alliaceae	Nechishnkurt	Herb	Bulb	16		Р
2	Allium cepa L.	Alliaceae	Keyi shunkurt	Herb	Bulb	23		Р
3	Becium filamentasum (Forssk.)Cliab.	Lamiaceae		Herb	Fruit	5		В
4	Brassica nigra (L.)Koch	Brassicaceae	Sinafich	Herb	Seed	5		Α
5	Capsicum annuum L.	Solanaceae	Miximixa	Herb	Fruit	14		Α
6	Capsicum frutescens L.	Solanaceae	Berberie	Herb	Fruit	12		Α
7	Coriandrum sativum L.	Apiaceae	Dinbilali	Herb	Fruit	52		Α
8	Lippia adoensis var. kosert Sebsebe	Verbenaceae	Kosert	Shrub	Leaves	10		Р
9	Lippia adoensis	Verbenaceae	Kasse	Shrub	Leaves	15		Р
10	Menta spicata L.	Lamiaceae	Nana	Herb	Leaves	14		Р
11	Ocimum basilicum L.	Lamiaceae	Besobila	Herb	Seeds, leaves	37		Α
12	Ocimum basilicum var. basilicum L.	Lamiaceae	Besobila	Herb	Seeds, leaves	20		Α
13	Rhamnus prinoides L'Herit	Rhamnaceae	Gesho	Shrub	Leaves, stem	6		Р
14	Rosmarinus officinalis L.	Lamiaceae	Sigametbesha	Shrub	Leaves	68		Р
15	Ruta chalpensis L.	Rutaceae	Sunkurta	Herb	Leaves and seed	79		Р
16	Zingiber officinale L.	Zingiberaceae	Zingibel	Herb	Stem	2		Р
List	of stimulant species							
1	Catha edulis (vahl.)Forssk.ex.Endl.	Celastraceae	Chat	Shrub	Leaves	12		Р
2	Coffee arabica L.	Rubiaceae	Buna	Shrub	Fruits	97		Р
3	Nicotiana tobacum L.	Solanaceae	Timbaho	Herb	Leaves	13		Α
List	of fragrant plant species							
1	Artemisia absinthium L.	Asteraceae	Ariti	Herb	Leaves	5	С	Р
2	Artemisia abyssinica L.	Asteraceae	Chuqun	Herb	Leaves	12	С	Р
3	Cympogen citrates (DC.) Stapf.	poaceae	Hexicho	Herb	Leaves	29	С	Р
4	Faeniculum vulgare	Apiaceae	Insilal	Herb	Leaves	11	С	В
5	Lippia adoensis var adoensis Hochst.exWalp	Verbenaceae	Kessie	Shrub	Leaves	33	С	Р
6	Lippia adoensis var koseret Sebsebe	Verbenaceae	Kosert	Shrub	Leaves	45	С	Р
7	Myrtus communis L.	Myrtaceae	Ades	Shrub	Leaves	4	С	Р
8	Ocimum lamiifolium Hochst.ex Benth.	Lamiaceae	Demakase	Shrub	Leaves	89	С	Р
9	Otostogia integrifolia Benth.	Lamiaceae	Tinjuit	Shrub	Leaves/stem	6	С	Α
10	Olea europea	Oleaceae	Weira	Tree	Leaves/stem	23	С	Р

# Appendix II. Contd.

11	Ruta chalepensis L.	Rutaceae	Tena adam	Herb	Leaves	115	С	Р
12	Rosmarinus officinalis L.	Lamiaceae	Siga metibesha	Shrub	Leaves	68	С	Р
List	of fodder species							
1	Cynodon dactylon (L.)Pers	Poaceae	Sardo	Herb	Leaves	37	W	Р
2	Pennisetum puprureum Schumach	Poaceae	Elphant grass	Herb	Leaves	21	С	Р
3	Sesbania sesban L. Merr	Fabaceae	Sesbania	Shrub	Seeds	26	С	Р
4	Vetiveria zizanioides (Linn.)Nash	Gramineae	Vetiver grass	Herb	Leaves	13	С	Р

# **Appendix III.** List of medicinal plants documented in the Hawassa city homegardens.

No.	Scientific name	Family	Local name	Habit	Parts used	Disease treated	Fr.	Duration
1	Achranthes aspera L.	Amaranthaceae	Telnji	Н	Root	Pneumonia	78	Р
2	Allium sativum L.	Alliaceae	Nechshinkurt	Н	Bulb	Malaria	14	Р
3	Aloe vera (L.) Burm.f.	Aloaceae	Ret	Н	Stem	Malaria, wound	18	Р
4	Azadiachta indica	Meliaceae	Neem	Т	Leaves	Malaria	12	Р
5	Artemisia abyssinica L.	Asteraceae	Ariiti	Н	Leaves	Evil eye, stomach ache	46	Р
6	Artemisia absinthium L.	Asteraceae	Chkun	Н	Leaves	Hemorrhoid	35	Р
7	Artemisia afra	Asteraceae		Н	Leaves	Evileye	40	Р
8	Carica papaya	Caricaceae	Papaya	Т	Leaves	Malaria	97	Р
9	Carissa edulis	Apocynaceae	Agam	Sh	Stem	Eveil eye	5	Р
10	Cassia occidentalis (L.)Link.	Fabaceae	Hamashaqa	Н	Leaves	Body swelling	15	Р
11	Coffee arabica L.	Rubiaceae	Bunna	Sh	Seeds	Gastric illness	98	Р
12	Commelina benghalensis L.	Commelinaceae		Н	Stem	Wound	42	Α
13	Croton macrostachyus	Euphorbiaceae	Bisana	Т	Leaves	Cancer	64	Р
14	Cucumis ficifolius A.Rich	Cucurbitaceae	Yemed emboy	CI	Leaves,fruits	Cold,heart disease	29	Р
15	Datura stramonium L.	Solanaceae	Asangira	Н	Leaves, seeds	Wound,	21	Α
16	Dodonaea angustifolia	Sapindaceae	Ittancha	Т	Stem	Tooth ace	5	Р
17	Eucalyptus globulus	Myrtaceae	Nechi barzaf	Т	Leaves	Common cold	3	Р
18	Euphorbia tirucalli	Euphoriaceae	Qincib	Sh	Stem fluid	Hemorrhoid	15	Р
19	Foeniculum vulgare	Apiaceae	Insilal	Н	Leaves	Stomach pain, urine problem	6	В
20	Hagenia abyssinica	Rosaceae	Kosso	Т	Flowers	Tape worm	9	Р
21	Juniperus procera	Cupressaceae	Yeabesha tid	Т	Seeds	Flue	7	Р
22	Kalachoe petitiana A.Rich	Crassulaceae	Hanculullee	Н	Leaves	Swelling	11	Α
23	Melia azedaracha	Meliaceae	Niimi	Т	Shoot tip	Malaria, toothache	98	Р
24	Millettia ferruginea (Hochst.)Bak	Fabaceae	Hengedicho	Tree	Stem bark	Ecto- parasite	16	Р
25	Moringa stenopetala L.	Moringaceae	Shifera	Tree	Leaves	Malaria, hypertension	23	Р

# Appendix III. Contd.

26	Nicotiana tabacum L.	Solanaceae	Araddo	Herb	Leaves	Common cold	14	А
27	Olea europaea ssp.cuspidata	Oleaceae	Ejersu	Tree	Stem	Tooth ache	19	Р
28	Ocimum lamiifolium	Lamiaceae	Damakasse	Shrub	Leaves	Sun stroke	71	Α
29	Phytolacca dodecandra L' Herit	Phytolaccaceae	Endod	Shrub	Root, leaves	Blahariza	5	Р
30	Podocarpus falcatus (Thunb.)Mirb	Podocarpaceae	Zigiba	Tree	Stem bark	Jaundice	12	Р
31	Prunus africana (Hook.F.)Kalkm.	Rosaceae	Garbicho	Tree	Bark	Cancer	6	Р
32	Rhamnus prinoides L'Herit.	Rhamnaceae	Xaddo	Shrub	Leaves	Skin infection	11	Р
33	Rumex nepalensis Spreng.	Polygonaceae	Sharbicho	Herb	Leaves/root	Ear problem, body Swelling	9	Α
34	Ruta chalepensis L.	Rutaceae	Sunkurta	Herb	Leaves	Stomach problem	98	Α
35	Ricinus communis L.	Euphorbiaceae	Qomboho	Tree	Root	Pneumonia	14	Р
36	Sesbania sesban (L.) Merr.	Fabaceae	Arbeti	Shrub	Root	Body swelling	10	Р
37	Solanum incanum L.	Solanaceae	Borbodho	Shrub	Root	Intestinal parasities	12	Р
38	Solanum nigrum L.	Solanaceae	Xunaye	Herb	Leaves	Intestinal parasites	21	Α
39	Vernonia amygdalina Del.	Asteraceae	Hecho	Tree	Leaves	Malaria	69	Р
40	Vernonia auriculifera Hiern.	Asteraceae	Rejicho	Shrub	Leaves	Wound	9	Р
41	Withania somnifera (L.)Dunal.	Solanaceae	Gizawa	Herb	Root	Pneumonia	39	Р

Appendix IV. Timber (Furniture) tree species encountered in the study area.

No.	Scientific name	Local name	Family	Habit	Frequency	Duration
1	Acacia albida		Fabaceae	Tree	12	Р
2	Acacia melanoxylon R.Br.	Omedlla(Amh)	Fabaceae	Tree	67	Р
3	Acacia tortilis (Forssk.)	Teddecha	Fabaceae	Tree	14	Р
4	Albiza gummifera (J.F.Gmel.	Matticho	Fabaceae	Tree	5	Р
5	Albizia schimperiana var. schimperiana	Mukarba(Or)	Fabaceae	Tree	4	Р
6	Aningeria adolfi-friedericii	Kararo	Sapotaceae	Tree	6	Р
7	Arundo donax	Shenbeko	Poaceae	Shrub	9	Р
8	Azadirachta indica	Neem	Meliaceae	Tree	3	Р
9	Casuarina equisetifolia L.	Shewshewe	Casuarinaceae	Tree	87	Р
10	Celtis africana Burm.f	Xoqono(Shisho)	Ulmaceae	Tree	8	Р
11	Cordia africana Lam.	Wadicho	Boraginaceae	Tree	97	Р
12	Croton macrostachyus Del.	Masincho	Euphorbiaceae	Tree	33	Р
13	Cupressus Iusitanica Mill.	Homme	Cupresaceae	Tree	98	Р
14	Eucalyptus camaldulensis Dehn.	Duumo bahirzafe	Myrtaceae	Tree	29	Р
15	Eucalyptus globulus	Waajoo bahirzafe	Myrtaceae	Tree	4	Р
16	Eucalyptus saligna Smith.	Duumo bahirzafe	Myrtaceae	Tree	54	Р

# Appendix IV. Contd.

17	Ficus sur Forssk.	Odakko	Moraceae	Tree	8	D
		Ouakko			_	<u>'</u>
18	Ficus vasta		Moraceae	Tree	14	Р
19	Grevillea robusta R.Br.	Temenjazaf	Proteaceae	Tree	99	Р
20	Hagenia abyssinica (Bruce)J.F.Gmel.	Dadako	Rosaceae	Tree	6	Р
21	Juniperus procera HochstexEngl.	Honcho	Cupresaceae	Tree	13	Р
22	Melia azedarch L.	Neem,	Meliaceae	Tree	118	Р
23	Olea europaea L.ssp. Cuspidata(Wall.ex G.Don) Cif.	Ejersu	Oleaceae	Tree	7	Р
24	Pinus patula L.	Patula	Pinaceae	Tree	16	Р
25	Pinus radiata		Pinaceae	Tree	5	Р
26	Podocarpus falcatus (Thunb.) Mirb.	Dagucho	Podocarpaceae	Tree	11	Р
27	Prunus africana (Hook.f.)Kalkm	Garbicho	Rosaceae	Tree	9	Р
28	Syzygium guineense (Wild.)DC.	Duwancho	Myrtaceae	Tree	3	Р
29	Balanites aegyptiaca (L.) Del.	Badana	Balanitaceae	Tree	4	Р

# Appendix V. List of plants used as fire wood.

No.	Scientific name	Local name	Family	Habit	Fr.	Duration
1	Acacia abyssinica Hochst.Ex Benth	Wacho	Fabaceae	Tree	12	Р
2	Acacia albida	Grar	Fabaceae	Tree	24	Р
3	Acacia etbaica Schweinf.	Grar	Fabaceae	Tree	10	Р
4	Acacia mearnsii DeWild.	Yefereji grar	Fabaceae	Tree	32	Р
5	Acacia nilotica	Cheba	Fabaceae	Tree	6	Р
6	Acacia seyal Del.	Wachu	Fabaceae	Tree	15	Р
7	Acacia melanoxylon R.Br.	Omedella	Fabaceae	Tree	55	Р
8	Acacia tortilis (Forssk.)Hayne	Deweni grar	Fabaceae	Tree	20	Р
9	Albiza gummifera (J.F.Gmel.	Maxicho	Fabaceae	Tree	4	Р
10	Albizia schimperiana var. schimperiana	Gorbe	Fabaceae	Tree	3	Р
11	Azadirachta indica	Neem	Meliaceae	Tree	2	Р
12	Senna didymobotrya (Fresen.) Irwin & Barneby	Hamashaqa	Caesalpinioideae	Shrub	4	Р
13	Casuarina equisetifolia L.	Arezelibanos	Casuarinaceae	Tree	77	Р
14	Celtis africana Burm.f	Amalaka	Ulmaceae	Tree	4	Р
15	Combretum collinum Fresen.		Combretaceae	Tree	5	Р
16	Cordia africana Lam.	Wanza	Boraginaceae	Tree	114	Р
17	Croton macrostachyus Del.	Masincho	Euphorbiaceae	Tree	68	Р
18	Cupressus Iusitanica Mill.	Homme	Cupresaceae	Tree	76	Р
19	Dodonaea angustifolia L.	Etancha	Sapindaceae	Tree	7	Р
20	Eucalyptus camaldulensis Dehn.	Duumebahirzafe	Myrtaceae	Tree	78	Р

# Appendix V. Contd.

21	Eucalyptus globulus	Duume bahirzafe	Myrtaceae	Tree	2	Р
22	Ficus sur Forssk.	Odakko	Moraceae	Tree	7	Р
23	Grevillea robusta R.Br.	Temenjazaf	Proteaceae	Tree	89	Р
24	Hagenia abyssinica (Bruce)J.F.Gmel.	Dadako	Rosaceae	Tree	12	Р
25	Jacaranda mimosifolia D.Don.	Jacaranda	Bignoniaceae	Tree	79	Р
26	Justicia schimperiana (Hochst ex.Nees	Cikkicho	Acanthaceae	Shrub	23	Р
27	Maytenus arbutifolia (A.Rich.)Wilczek	Cucco	Cleastraceae	Tree	5	Р
28	Melia azedarch L.	Neem,	Meliaceae	Tree	116	Р
29	Millettia ferruginea (Hochst.)Bak.	Hengedicho	Fabaceae	Tree	10	Р
30	Olea europaea L.ssp. Cuspidata (Wall.ex G.Don) Cif.	Ejersu	Oleaceae	Tree	19	Р
31	Pinus patula L.	Patula	Pinaceae	Tree	36	Р
32	Podocarpus falcatus (Thunb.) Mirb.	Dagucho	Podocarpaceae	Tree	24	Р
33	Prunus africana (Hook.f.)Kalkm	Garbicho	Rosaceae	Tree	18	Р
34	Schinus molle L.	Kunde berbere	Anacardiaceae	Tree	63	Р
35	Sesbania sesban (L.) Merr.	Arbeti	Fabaceae	Shrub	39	Р
36	Spathodea campanulata P.Beauv. ssp. nilotica.	Spathoda	Bignoniaceae	Tree	45	Р
37	Syzygium guineense (Wild.)DC.	Duwancho	Myrtaceae	Tree	5	Р
38	Vernonia amygdalina Del.	Hecho	Asteraceae	Shrub	74	Р
39	Balanites aegyptiaca (L.) Del.	Badano	Balanitaceae	Tree	12	Р

Cl, climbers; P, Perennial; A, Annual; C, Cultivated; W, Wild; T, Tree; Sh, Shrub; H, Herb.

# academicJournals

Vol. 10(10), pp. 234-237, October 2016

DOI: 10.5897/AJP\$2016.1430 Article Number: 11E5A4C61082

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# Short Communication

# Performance of yield attributes, yield and economics of teff (*Eragrostis tef*) influenced by various row spacing, nitrogen and phosphorus fertilizers

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Received 24 May, 2016; Accepted 22 August, 2016

Teff [Eragrostis tef (Zucc.)] is the most important cereal crop of Ethiopia, occupying 31% area and 20% production of the total cereal crops. However, its productivity is almost stagnant, with a national average yield of meager 1.46 tonnes ha mainly due to inadequate modern agronomic practices and technologies in the present production system. The urgent necessity for its higher production and productivity as a staple food of the country is increasing year after year. Therefore, in order to investigate the effect of three levels of rows spacing, four levels of nitrogen and phosphorus fertilizer rates and their interaction on growth and yield attributes as well as yield of teff, a field experiment was conducted on kora variety of teff. The response of growth parameters to the treatments were significant in all parameters, except lodging percentage due to main effect of rows spacing. The highest increments recorded were 317, 49.09 and 28.18% in effective tillers, panicle length and plant height plant<sup>-1</sup>, respectively in their interaction effects in treatments of 80/80 and 70/70 kg of N/P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup> with 10 cm over the lowest result recorded from treatment of 50/50 kg of N/P<sub>2</sub>O<sub>5</sub>ha<sup>-1</sup> with 30 cm spacing. While, the response of yield and yield components were found significant and the highest result in interaction effects were an increment of 113.21, 35.28, 55.45 and 41.7% kg ha<sup>-1</sup> for grain yield, straw yield, biomass yield and harvest index, respectively from 80/80 kg of N/P₂O₅ with 10 cm spacing over the lowest result reported from 50/50 kg of N/P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup> with 30cm spacing. Economic partial budget analysis of the study depicted that at row spacing of 10 cm with 80/80 kg of N/P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup> fertilizer dose resulted in maximum relative net return of ETB 50178 ha<sup>-1</sup> followed by ETB 48017 ha<sup>-1</sup> for 70/70 kg of N/P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup>. The study concluded that, higher productivity and net economic return can be achieved for kora variety of teff by applying 80/80 kg of N/P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup> with 10 cm row spacing as compared to the other treatments.

**Key words:** Economics, nitrogen, phosphorus, row spacing, teff, yield.

### INTRODUCTION

Teff [Eragrostis tef (Zucc.)] is the most important staple food crop in Ethiopia, occupying 31% area and 20%

production of the total cereal crops (CSA, 2014). Teff grain and straw has good demand and fetch relatively

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	Plant height (cm)				Effectiv	e tiller n	number plants <sup>-1</sup>		
Fertilization	Row spacing (cm)								
N/P <sub>2</sub> O <sub>5</sub> (kg ha <sup>-1</sup> )	10	20	30	Mean	10	20	30	Mean	
50/50	86.6 <sup>f</sup>	88 <sup>f</sup>	97.3 <sup>e</sup>	90.66 <sup>d</sup>	3.37 <sup>de</sup>	3.63d <sup>e</sup>	3.2 <sup>e</sup>	3.4 <sup>c</sup>	
60/60	98.5 <sup>de</sup>	97.4 <sup>e</sup>	97.2 <sup>e</sup>	97.73 <sup>c</sup>	7.91 <sup>b</sup>	5.73 <sup>c</sup>	5.5 <sup>c</sup>	6.38 <sup>b</sup>	
70/70	104.3 <sup>b</sup>	103 <sup>cd</sup>	100.1 <sup>cd</sup>	101.59 <sup>b</sup>	10.1 <sup>a</sup>	8.33 <sup>b</sup>	5.43 <sup>c</sup>	7.96 <sup>a</sup>	
80/80	101.7 <sup>c</sup>	111.0 <sup>a</sup>	106.1 <sup>b</sup>	106.30 <sup>a</sup>	10.9 <sup>a</sup>	8.3 <sup>b</sup>	5.9 <sup>c</sup>	8.37 <sup>a</sup>	
Mean	97.8 <sup>c</sup>	99.19 <sup>b</sup>	100.22 <sup>a</sup>		8 <sup>a</sup>	6.5 <sup>b</sup>	5.00 <sup>c</sup>		
LSD (5%) RS = $0$	.93				L	SD(5%)	RS = 0.	.5	

**Table 1.** Main and interaction effects on plant height at 90% physiological maturity plant<sup>-1</sup> and effective tiller number plants<sup>-1</sup>.

Means of the same letter along columns and rows are not significantly different.

higher price as compared to other cereal crops. Moreover, teff has excellently adapted to the changing agro-climatic conditions in Ethiopia; thus, reduces production risks. The major limitation for teff production in the country has been its low productivity. Teff has the lowest yield per hectare as compared to other major cereals, as the national average yield is meager, 1.46 tonnes ha<sup>-1</sup> (CSA, 2014). One of the reasons for the low yield is ineffective traditional sowing methods and inadequate application of nitrogen (N) and phosphorus (P2O5) fertilizers. Most farmers practice the traditional broadcasting method of sowing, which leads to excess crop density and increases competition among plants for nutrients, water, sunlight and CO2. Further, broadcasting methods requires additional seed rate as compared to row sowing method hence increases cost of production of teff.

LSD (5%) FR = 1.08

CV% 1.11

LSD (5%) RS X FR = 1.87

Therefore, present cultivation system of teff is unable to satisfy the consumers demand, due to the fact that most Ethiopian farmers practice traditional farming system. Production system is not efficiently supported by modern technology due to research gap in choosing most feasible modern technology. The research gaps in most effective sowing methods and most appropriate amount of N/P<sub>2</sub>O<sub>5</sub> fertilizers has been identified as significant constraints for the low productivity of teff in West Showa zone among others. The above modern agronomic practices and technologies have the potential to substantially enhance teff productivity to ensure food security in West Showa zone. The experiment was conducted with the objectives to study the effect of three levels of rows spacing, four levels of N/P2O5 fertilizer rates and their interaction on growth parameters, yield components and yield.

### **MATERIALS AND METHODS**

A field experiment was carried out at the experimental plots in Ambo University Research Site, Ambo. Ambo is located 114 km

away from Addis Ababa towards the west of the country. The research was done on 'Kora' variety of teff during the main cropping season of 2015 on Vertisols of Ambo district in West Shoa Zone, Western Oromia, Ethiopia.

LSD (5%) FR = 0.58LSD (5%) RS X FR = 1.0

CV% 9.23

The experiment was laid out in randomized complete block design with three replications. The treatments were: 10, 20 and 30 cm row spacing and 50/50 kg of N/P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup>, 60/60 kg of N/P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup>, 70/70 kg of N/P<sub>2</sub>O<sub>5</sub>ha<sup>-1</sup> and 80/80 kg of N/P<sub>2</sub>O<sub>5</sub>ha<sup>-1</sup> with application of di-ammonium phosphate (DAP) as basal dose and urea 21 days after sowing. Each harvestable plot had an area of 1.6 m by 1.25 m (2 m²) with 0.5 m spacing between plots and 1 m between blocks. The treatments were assigned to plots by randomization method.

### **RESULTS AND DISCUSSION**

The results of analysis of the observations revealed that the highest number of tillers plant (10.9) were recorded from the treatment combination of 80/80 kg of N/P<sub>2</sub>O<sub>5</sub>ha<sup>-1</sup> with 10 cm row spacing, followed by 10.1 from 70/70 kg of N/P<sub>2</sub>O<sub>5</sub>ha<sup>-1</sup> with 10 cm, respectively. However, the lowest number of tillers (2.6) was recorded from the combination of 50/50 kg of N/P<sub>2</sub>O<sub>5</sub>ha<sup>-1</sup> with 30 cm rows spacing (Table 1). The highest number of tillers was recorded from the treatment of 10 cm rows spacing with  $80/80 \text{ kg of N/P}_2\text{O}_5\text{ha}^{-1}$  and  $70/70\text{kg of N/P}_2\text{O}_5\text{ha}^{-1}$  may be due to less plant density which creates favorable environment for plant tillering. The effective number of tillers was increased by 240.6% in plots treated with 80/80 kg of N/P<sub>2</sub>O<sub>5</sub>ha<sup>-1</sup> with 10 cm rows spacing as compared to the lowest result obtained from the treatment combination of 50/50 kg of N/P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup> with 30 cm space between the rows. As the number of productive tillers plant 1 increased, the yield per hectare also increased. Therefore, analysis of effective number of tillers counted at 90% physiological maturity of teff was significantly (p<0.001) affected between 12 treatments of rows spacing and the combination of N and P<sub>2</sub>O<sub>5</sub> fertilizer rates (Table 1). Debebe (2005) reported that productive

**Table 2.** Main and interaction effects on teff grain yield in kg ha<sup>-1</sup> and harvest index (%).

		Grain yiel	d (kg ha <sup>-1</sup>	Harvest Index (%)							
Fertilization	Row spacing (cm)										
N/P(kg ha <sup>-1</sup> )	10	20	30	Mean	10	20	30	Mean			
50/50	2150 <sup>d</sup>	1966 <sup>e</sup>	1766.6 <sup>f</sup>	1960.86 <sup>c</sup>	27 <sup>d</sup>	26 <sup>d</sup>	24 <sup>e</sup>	26 <sup>c</sup>			
60/60	3036.7 <sup>b</sup>	2650 <sup>c</sup>	2050d <sup>e</sup>	2588.89 <sup>b</sup>	32 <sup>ab</sup>	31b <sup>c</sup>	27 <sup>d</sup>	31 <sup>b</sup>			
70/70	3633.4 <sup>a</sup>	2933.4 <sup>b</sup>	2600 <sup>c</sup>	3055.56 <sup>a</sup>	34 <sup>a</sup>	32 <sup>ab</sup>	30°	32.3 <sup>a</sup>			
80/80	3766.7 <sup>a</sup>	3050 <sup>b</sup>	2633.3 <sup>c</sup>	3150 <sup>a</sup>	34 <sup>a</sup>	33 <sup>ab</sup>	30°	32.6 <sup>a</sup>			
Mean	3146.7 <sup>a</sup>	2649.8 <sup>b</sup>	2262 <sup>c</sup>		32 <sup>a</sup>	31 <sup>b</sup>	28 <sup>c</sup>				
LSD (5%) RS	= 89.6				LS	D (5%)	RS =	0.79			
LSD (5%) FR	= 103.5			LSD $(5\%)$ FR = 0.92							
LSD (5%) RS	X FR = 80	LSD (5	5%) RS	X FR	= 0.1.5						
CV% 3.95						CV%	3.09				

Means of the same letter along columns and rows are not significantly different.

tiller numbers were affected by plant densities and available nutrients in his study on teff.

Likewise, the analysis of plant heights measured at 90% physiological maturity of teff were significantly (p<0.001) affected among 12 treatments of row spacing and combination of N/P2O5 fertilizer rates (Table 1). The highest plant height plant 1 (111.0 cm) was recorded from the combination of 80/80 kg of N/P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup> with 20 cm, followed by 104.3 cm from 70/70 kg of N/P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup> with 10 cm row spacing and 106.1 cm from 80/80 kg of N/P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup> with 30 cm row spacing, respectively. However, the lowest (86.6 cm) was recorded from the combination of 50/50 kg of N/P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup> with 10 cm row spacing (Table 1). The highest mean value of plant height of the treatment was significantly taller by 28.18% over the shortest plant height. This might be due to a series of intra-row competition for nutrients such as N/P2O5 ha-1, which retarded elongation of stems. Legesse (2004) also reported that, as applied N rates increased, the grain uptake also increased which also reflected in the plant height, yield and yield components like panicle length, grain yield, straw yield and biomass yield.

Further, the grain yield ha<sup>-1</sup> was significantly (p<0.001) affected among 12 treatments of row spacing and combination of N/P<sub>2</sub>O<sub>5</sub> fertilizer rates (Table 2). The highest grain yield (3766.67kg ha<sup>-1</sup>) was recorded from the combination of 80/80 kg of N/P<sub>2</sub>O<sub>5</sub>ha<sup>-1</sup> with 10 cm rows spacing followed by 3633.4 kg ha<sup>-1</sup> from 70/70 kg of N/P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup> with 10 cm rows spacing. While, moderate grain yield of 3050, 2933 and 3036.7 kg ha<sup>-1</sup> were recorded from 80/80 kg of N/P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup> with 20 cm row spacing, 70/70 kg of N/P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup> with 20 cm row spacing and 60/60 kg of N/ P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup> with 10cm spacing between the rows, respectively. However, the lowest grain yield of 1766.6 kg ha<sup>-1</sup> was recorded from the combination of 50/50 kg of N/P<sub>2</sub>O<sub>5</sub>ha<sup>-1</sup> with 30 cm rows spacing (Table 2). Kassahun (2001) reported that adequate nitrogen fertilization had substantial influence on yield and related traits of teff.

Similarly, the highest mean value of grain yield was increased by 113.22 and 105.67% in the treatments of 80/80kg of  $N/P_2O_5$  ha<sup>-1</sup> with 10 cm rows spacing and 70/70 kg of  $N/P_2O_5$  ha<sup>-1</sup> with 10 cm row spacing, respectively as compared to 50/50 kg of  $N/P_2O_5$  ha<sup>-1</sup> with 30 cm row spacing. This might be due to the fact that plants supplied with adequate amount of  $N/P_2O_5$  fertilizers had better ability for absorbing nutrients and water which enhanced vegetative growth and grain filling as compared to plants grown under lower fertilizer rates. Tekalign and Teklu (2000) reported yield improvement due to effective management of soil fertility and plant nutrition in their research conducted on teff.

Further, analysis of the harvest index of teff was significantly (p<0.001) affected by 12 treatments of rows spacing and combination of N/P<sub>2</sub>O<sub>5</sub> fertilizer rates (Table 2). The highest harvest index (34%) was recorded from the combination of 80/80 kg of N/P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup> with 10 cm row spacing. The moderate harvest index (33, 32 and 32%) was recorded from the combination of 80/80 kg of  $N/P_2O_5$  ha<sup>-1</sup> with 20 cm row spacing, 70/70 kg of  $N/P_2O_5$ ha<sup>-1</sup> with 20 cm row spacing and 60/60 kg of N/P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup> with 10 cm row spacing, respectively, which were statically at par with each other. On the other hand, the lowest harvest index (24%) was recorded from the combination of 50/50 kg of N/P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup> with 30 cm row spacing. Plots treated with 80/80 kg of N/P $_2$ O $_5$  ha $^{-1}$  with 10 cm and 70/70 kg of N/P $_2$ O $_5$  ha $^{-1}$  with 10 cm row spacing showed the harvest index increment of 41.7 and 41.7%, respectively as compared to 50/50 kg of N/P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup> with 30 cm. Liben et al. (2004) reported higher harvest index with adequate nitrogen fertilizer rate in teff.

### Assessment of the benefit

Cost ratio associated with different treatments, the partial budget technique of CIMMYT was applied on yield and straw yield. Based on this technique, the highest net

**Table 3.** Partial budget analysis for variable cost on mean yield of grain and straws.

Parameters	30 cm RS &50/50 kg N/P ha <sup>-1</sup>	30 cm RS &60/60 kg N/P ha <sup>-1</sup>	20 cm RS &50/50 kg N/P ha <sup>-1</sup>	10 cm RS &50/50 kg N/P ha <sup>-1</sup>	30 cm RS &80/80 kg N/P ha <sup>-1</sup>	30 cm RS &70/70 kg N/P ha <sup>-1</sup>	20 cm RS &60/60 kg N/P ha <sup>-1</sup>	20 cm RS &70/70 kg N/P ha <sup>-1</sup>	20 cm RS &80/80 kg N/P ha <sup>-1</sup>	10 cm RS &60/60 kg N/P ha <sup>-1</sup>	10 cm RS &70/70 kg N/P ha <sup>-1</sup>	10 cm RS &80/80 kg N/P ha <sup>-1</sup>
AGY kgh <sup>-1</sup>	1767	2050	1966	2150	2633	2600	2650	2933	3050	3037	3633	3767
SY kg ha-1	5433	5500	6233	5583	5667	5900	5617	5867	6150	6233	6983	7350
GR ETB ha-1	27389	31775	30473	33325	40812	40300	41075	45462	47275	47074	56312	58389
SR ETB ha <sup>-1</sup>	8150	8250	9350	8374.5	8501	8850	8426	8801	9225	9350	10475	11025
TR ETB ha <sup>-1</sup>	35539	40025	39823	41670	49313	49150	49501	54263	56500	56424	66787	69414
Cost of DAP ETB ha-1	1535	1842	1535	1535	2456	2149	1842	2149	2456	1842	2149	2456
Cost of Urea ETB ha-1	8o1	961	801	801	1280	1121	961	1121	1280	961	1121	1280
Variable cost ETB ha-1	15500	15500	15500	15500	15500	15500	15500	15500	15500	15500	15500	15500
Total cost ETB ha-1	17836	18303	17836(-)	17836(0)	19236	18770(-)	18303(-)	18770	19236	18303(-)	18770	19236
NB (ETB ha-1)	17703	21722	21987	23834	30077	30380	31198	35493	37264	38121	48017	50178
MRR%		861%			445.9%			919.7%	380%		2119%	463.7%

Where, RS= Row Spacing, \*Sale of grain = ETB 15.5 kg<sup>-1</sup> and Sale of straw = ETB 1.5 kg<sup>-1</sup>. \*Variable costs = Cost from land preparation to yield and straw yield transportation (ETB 15125) + cost of seed (ETB 375) were equivalent for all treatments. Where, GY kg/ha (in kilo gram/hectare) = Grain yield, AGY = adjusted grain yield, SY = straw yield, GR Birr/ha= grain revenue in Birr/ha, TR Birr/ha = total revenue in Birr/ha, TVct Birr/ha= total variable cost in Birr/ha, NB Birr/ha = net benefit Birr/ha, MRR%= marginal return rate in percentage.

return was estimated from 80/80 kg/ha of N/P $_2$ O $_5$  with 10 cm (Table 3). The partial budget analysis estimated that the treatment of 10 cm row spacing with 80/80 kg of N/P $_2$ O $_5$  ha $^1$  fertilizer rate resulted in maximum relative net return of ETB 50178 ha $^1$  followed by ETB 48017 ha $^1$  for 10 cm row spacing with 70/70 kg of ha $^1$  and ETB 38121 ha $^1$  for 10 cm row spacing with 60/60 kg of N/P $_2$ O $_5$  ha $^1$  fertilizer rate, respectively (Table 3). Similar higher net return was also reported by Tefera and Ketema (2001).

It can be concluded that from the present study, application of 80/80 kg of N/P<sub>2</sub>O<sub>5</sub>ha<sup>-1</sup> with 10 cm rows spacing followed by 70/70 kg of N/P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup> with 10 cm row spacing with the same amount of 25 kg ha<sup>-1</sup> of seed rate resulted in better economical return with maximum grain yield production for kora variety of teff in the field experiment. Further, the research also suggests that there is potential to conduct future studies

under varying amounts of seed rate below 25 kg ha<sup>-1</sup> with the studied N/P<sub>2</sub>O<sub>5</sub> fertilizer rate and rows spacing.

### Conflict of Interests

The authors have not declared any conflict of interests.

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