Ethiop. J. Biol. Sci. **17(2): 99–116, 2018**
 ISSN: 1819-8678

RESEARCH ARTICLE

ANALYSIS OF GRAIN YIELD STABILITY AND QUALITY TRAITS OF MALT BARLEY GENOTYPES UNDER TERMINAL MOISTURE-STRESSED ENVIRONMENTS OF NORTH WOLLO

Arega Gashaw [1,*], Agegnehu Mekonnen[1], Eyeberu Abere[1] and Akalu Gebru[1]

ABSTRACT: Fifteen malt barley genotypes, along with the standard check (Miscal 21), were evaluated at Geregera, Kon and Estayish testing sites of Sirinka Agricultural Research Centre during 2013 and 2014 cropping seasons, with the objectives of evaluating the performance of malt barley genotypes for grain yield, yield stability and grain quality traits. The experiment was carried out using Randomized Completely Block Design (RCBD), replicated three times. Data were recorded and subjected to Analysis of Variance (ANOVA) using Genstat 18th software package. Analysis of Genotype by Environment Interaction (GEI) was carried out using Additive Main effects and Multiplicative Interaction (AMMI) model. ANOVA depicted significant variations among malt barley genotypes for all the traits considered. AMMI1 and AMMI2 analyses plainly assigned genotypes and environments in biplots graphs. G6 (Libra T95/Diamalt) and G11 (*E.Acacia*/ Defra//Atah92/Gob) were plotted at the far right hand side of the AMMI1 biplot graph, indicating their best performance in grain yield over the rest of the tested malt barley genotypes. G6 was plotted with Geregera 2014 and Kon 2014 environments while G11 was plotted with Estayish 2014 in AMMI2 biplot graph, showing their specific adaptability to the environments where they were plotted with. However, G11 was found susceptible to scald and was excluded from further evaluation. On the other hand, G6 was advanced to variety verification trial and evaluated for agronomic and quality traits at North and South Wollo major barley growing areas in 2017 and 2018 cropping seasons. G6 yielded above average grain yield and fulfilled the minimum TKW, HLW, PC and EC quality standards, and was relatively tolerant to Scald and Net Bloch diseases. Therefore, G6 (Libra T95/Diamalt) was found superior to the rest of the tested malt barley genotypes for both agronomic and malt quality traits under terminal moisture-stressed barley growing areas of North Wollo and similar environments, and officially released by the National Variety Releasing Committee (NVRC) for commercial production in 2019 cropping season with a local name "Waro".

Key words/phrases: AMMI, GEI, Grain quality, Malt barley.

[1] Sirinka Agricultural Research Centre, P.O. Box 74, Woldia, North Wollo, Ethiopia. E-mail: argonlacomolza@gmail.com
* Author to whom all correspondence should be addressed.

INTRODUCTION

Malt barley (*Hordeum distichon* L.) is the most important cereal crop in malting industries (Biadge Kefale and Yadesa Abushu, 2017). Because of its husk content, barley is highly preferred for malting. The husk protects the acrospire during germination process and provides firm texture grain and higher amylase activity (Kumar *et al*., 2013). Barley is rich in β-glucan and glycemic index, both are useful in lowering the risk of cardio-vascular diseases (Kumar *et al*., 2013). Moreover, malt barley is being utilized for baby foods, confectioneries, malt drinks and for medicinal syrups (Verma *et al*., 2011).

Malt barley possessed both α- and β-amylase that cleave the starch chains during malting, converting to maltose during malting. The presence of α- and β-amylase in barley grains makes barley the noble and universal grain for malters and breweries. The end quality of malt depends on grain and malt parameters. Several grain quality traits are being used to grade malt barley grain. Kernel plumpness, kernel boldness (TKW), hectoliter weight (HLW), grain protein content (PC) affect the end quality of malt (Kumar *et al*., 2013). Low grain protein content (PC<9.0%) decreases enzymatic activities and slows down the conversion process of starch, whereas higher grain protein content (PC>11.5%) lowers the proportion of grain starch content and decreases extract yield. The minimum national standard for grain protein content ranged from 9.0–11.0% for two-rowed and 9.0–11.5% for six rowed barley (Kumar *et al*., 2013).

Brewing industries are booming in Ethiopia, demanding more than 0.23 million tons of malt barley grains per annum. In spite of agro-ecological suitability for barley cultivation, production and productivity of malt barley in Ethiopia remained very low and didn't meet the growing demand of breweries. It is, therefore, imperative to develop high-yielding malt barley varieties that fulfill the minimum quality standards. The development of high-yielding and stable cultivars is the primary target of any plant breeders. Grain yield, a complex polygenic trait, exhibits Genotype x Environment Interactions (GEI) (Fan *et al*., 2007). Thus, because of the existence of GEI, selection based on grain yield alone may not always be effective, rather complicates the identification of superior genotypes for a range of environments. GEI reduces the correlation between phenotype and genotype values, resulting in inconsistent performance of genotypes in different environments and decreases selection efficiency (Kang, 1997; Karimizadeh *et al*., 2013).

Hence, identifying genotypes that perform relatively consistent across diverse environments or specifically-adapted genotypes to defined environment would minimize performance inconsistencies. Breeders use stability analysis to single-out stable or specific- adapted genotypes. Several statistical models have been proposed to analyze GEI and stability of genotypes in multi-environment trials (MET) (Ding *et al*., 2007; Karimizadeh *et al*., 2013; Yan *et al*., 2007). Of the various stability models, Additive Main effects and Multiplicative Interaction (AMMI) (Kadhem and Baktash, 2016; Karimizadeh *et al*., 2013) and Genotype by Genotype-Environment interaction (GGE) biplot (Ding *et al*., 2007; Karimizadeh *et al*., 2013) is the most recent and efficient stability analytical tool. However, information on malt barley genotypic grain yield stability and malt grain quality traits at terminal moisture-stressed environment of North Wollo is limited. Hence, this paper presents the results of grain yield stability and grain quality traits of malt barley genotypes at moisture-stressed environments of North Wollo.

MATERIALS AND METHODS

Description of the study areas

Field experiments were conducted at Geregera, Kon and Estayish testing sites of Sirinka Agricultural Research Centre (SARC), North Wollo Administrative zone, Ethiopia during 2013 and 2014 main cropping seasons. The testing sites lie between 11°36′51″–11°49′34″ N (latitude) and 38°44′57″–39°07′36″ E (longitude) at an altitude ranging from 2870 to 3270 meters above sea level (masl). Description of the testing sites is presented in Table 1.

Monthly rainfall from June-October over the last five years (2010–2014 cropping seasons) is presented in Fig. 1. Maximum precipitation (60–70% of the total annual rainfall) was recorded in July and August with maximum rainy days (25–31 days), and then showed a decreasing trend after September onwards and registered the lowest in October (0.4–55.1 mm) with extended dry spells in the growing season. Depending on the evapo-transpiration rate, barley requires 450–650 mm water throughout the growing season (Brouwer and Heibloem, 1986), of which about 45% is needed at late growth stage (Deo *et al*., 2017). Grain filling, translocation of photosynthesis assimilates from source-sink, starts in September and October in the testing locations. However, the total rainfall in September and October is lower than the optimum water requirement for grain filling, resulting in moisture-stress at late crop growth stage. Thus, the low

precipitation and few rainy days at grain filling stage might disturb the source-sink translocation pathway. The testing locations are therefore categorized as terminal moisture-stressed environments. Early cessation of rainfall at the critical grain filling stage became the major abiotic constraints that significantly affect crop production and productivity.

Table 1. Description of the testing sites.

Locations	Altitude (masl)	Rainfall* (mm)	Soil description**				
			Texture	PH	OM (%)	Total N (%)	Available P (ppm)
Geregera	2870	1060	Clay	5.8	2.3	0.15	13.8
Kon	2878	720	Silty clay loam	5.3	2.8	0.14	13.5
Estayish	3270	NA	Clay loam	5.3	4.6	0.24	16.5

Source: * Effective rainfall from June-October, National Meteorology Agency, Kombolcha Meteorological Station, NA = not available, ** SARC soil laboratory analysis

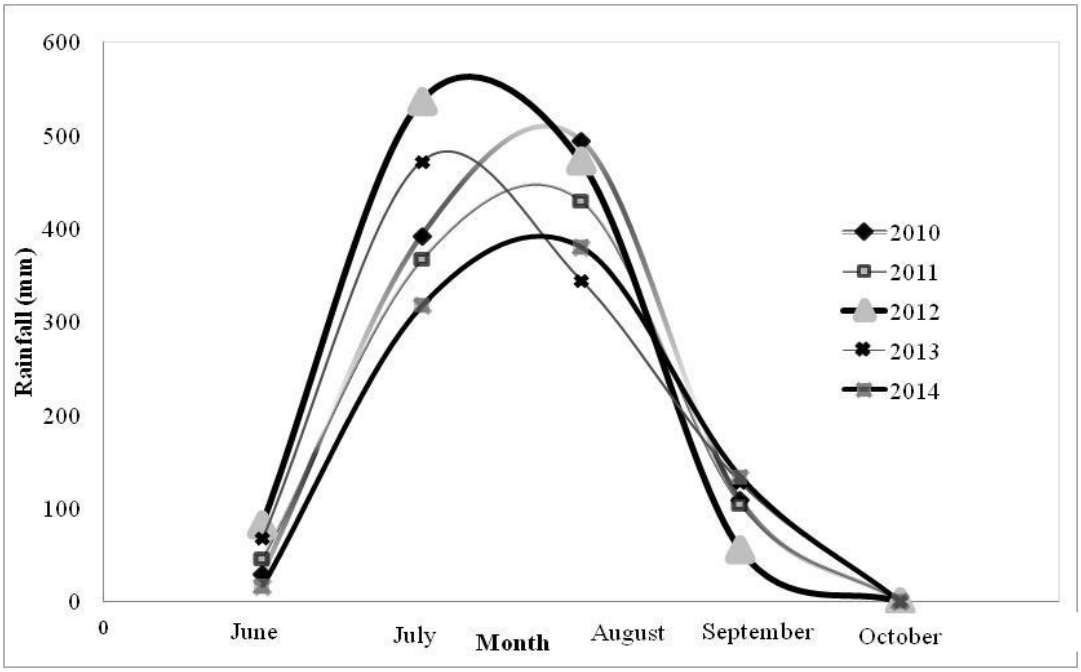

Fig. 1. Monthly rainfall from June-October over the five years (2010-2014 cropping seasons).

Experimental materials and experimental design

Fifteen malt barley genotypes, along with the standard check (*Miscal* 21) (Table 2) were evaluated for grain yield and quality traits. The experiment was carried out in Randomized Complete Block (RCB) design with three replications. The genotypes were row planted at a seed rate of 85 kg ha^{-1}. A plot size of $3m^2$ (six rows of 2.5 m length with an inter-spacing of 0.2 m)

was used.

Table 2. Pedigree of tested malt barley genotypes.

Code	Pedigree
G_1	Msel/Conlon
G_2	Canela/Estanzuela Jacaranda
G_3	E.Acacia/Defra//Atah 92/Gob
G_4	Libra T95/Af9216
G_5	Ibcb-Spring09/10#115
G_6	Libra T95/Diamalt
G_7	Merit,B/Ac Metcalfe
G_8	Bsi 58
G_9	Ibl Sgp09/10#14
G_{10}	E.Acacia/Defra/3/Svanhals/Bar/Msel//Azaf/Gob24dh
G_{11}	E.Acacia/Defra//Atah92/Gob
G_{12}	Condor-Bar/3/Ptt.B/Ruda//Aleli/4/Aleli/5/Foster
G_{13}	Fnc I 22/Defra/6/Zhedar#1/4/Shyri//Gloria-Bar/Copal/3/Shyri/Grit/5/Aruf
G_{14}	Nd19929/Prisna//Canela
G_{15}	Miscal 21

Nitrogen and phosphorous fertilizers were applied in the form of Urea (46% N) and Di-Ammonium Phosphate (DAP) (18% N and 46% P_2O_5) at the rate of 41 kg ha^{-1} N and 46 kg ha^{-1} P_2O_5, respectively. Full dose of P_2O_5 was applied at planting time, while N was applied in split, half at planting time and the remaining half at tillering stage. The experiment was hand-weeded twice at 25 and 45 days after emergence and other management practices were done as required.

Data collection and statistical analysis

Days to maturity (DM), number of days to reach physiological maturity, was recorded from plot basis. Grain yield (GY), obtained from the central four rows leaving a border rows from both sides of the plot, was measured using precision balance, adjusted to standard cereal moisture content (12.5%). Grain moisture content was measured using Dickey-John Grain Analysis Computer (GAC) 2100 moisture tester. Then, grain moisture content was corrected using the following formula;

$$GY_{Adj} = \left(\frac{100-y}{100-x}\right) * GY_{Unadj}$$

where, GY_{Adj} = adjusted grain yield at 12.5% moisture content,

y = the actual grain moisture content of the sample measured using GAC 2100 moisture tester,

x = the standard moisture content of cereal crops (12.5%) and

GY_{Unadj} = unadjusted grain yield. Then, adjusted grain yield was converted

to kg ha^{-1}.

Hectolitre weight (HLW) was measured using GAC 2100 moisture tester. A liter of grains were loaded on GAC 2100 moisture tester and the weight of grains was recorded, adjusted at 12.5% moisture content and recorded as kg hL^{-1}. Thousand kernels weight (TKW) was recorded from randomly taken pure thousand kernels. Thousand kernels were counted and weighed using precision balance. The data was adjusted to the standard moisture content (12%). On the other hand, grain protein content (PC%) and starch content (SC%) of each genotypes was estimated using non-destructive and robust Infratec^{TM} 1241 Grain Analyzer, Near-Infrared Reflectance Spectroscopy (NIRS), and recorded in percentage. To examine the agronomic and malt quality traits, the candidate genotype along with the standard check were further evaluated at North and South Wollo major barley-growing areas in 2017 and 2018 cropping seasons. Extract and protein contents of the candidate and the standard check were examined using NIRS.

Data were subjected to analysis of variance (ANOVA) using Genstat 18th software package as per Gomez and Gomez (1984). Duncan Multiple Range Test (DMRT) was used to separate treatment means. Before pooling the grain yield data from each environment, homogeneity of error variance was tested using Bartlett's test (Steel and Torrie, 1980). Stability of genotypes across diverse environments was analyzed using Additive Main effects and Multiplicative Interaction (AMMI) model of Genstat 18th software package as described in Gauch (1992).

RESULTS AND DISCUSSION

Genotypic variation of malt barley genotypes for days to maturity

Terminal moisture-stress is one of the major abiotic production constraints that significantly impair production and productivity of field crops in Eastern Amhara. Developing early maturing varieties that escape terminal moisture stress is becoming the primary objective of breeders. Thus, the tested malt barley genotypes were evaluated for days to heading at different environments that were characterized with different degree of stress. Genotypes showed highly significant variation ($p<0.01$) for days to maturity, implying remarkable genotypic maturity difference (Table 3). Mean days to maturity (Table 4) ranged from 133 days (G5) to 149 days (G3), where G6 had moderate maturity group (141 days). The tested malt barley genotypes, except G3 and G10, had an average physiological maturity of <145 days, making them suitable for terminal moisture-stressed environments.

Genotypic variation of malt barley genotypes for grain yield

The pooled data for grain yield was homogeneous ($p>0.19$), indicating the possibility of combining the grain yield data of all the testing environments. Combined analysis of variance (ANOVA) depicted highly significant variations among malt barley genotypes for grain yield and grain quality traits (Table 3), implying the presence of varietal variations for all the traits considered. Mean grain yield of malt barley genotypes (pooled over locations and over years) is presented in Table 4. Mean grain yield ranged from 1814 kg ha^{-1} (G_1) to 2854 kg ha^{-1} (G_6). Of the tested malt barley genotypes, G_6 and G_{11} consistently out-smarted in grain yield over the rest of the malt barley genotypes in all the testing environments, except at Estayish 2014. G6 recorded about 21% mean grain yield advantage over the check (*Miscal* 21).

The presence of seasonal and location variations influenced the performance of malt barley genotypes for grain yield (Table 3). Maximum mean grain yield was recorded at Kon 2014 (2934 kg ha^{-1}) followed by Estayish 2014 (2601 kg ha^{-1}) and Geregera 2014 (2532 kg ha^{-1}), indicating 2014 cropping season was conducive environment for malt barley production.

Moreover, genotype by environment interaction (GEI) exhibited significant variation for grain yield (Table 3), showing differential yield performance of malt barley genotypes across environments and necessitating developing stable or specific adaptable malt barley genotype. As grain yield is a polygenic trait, its phenotypic expression is controlled by genetic factors and highly influenced by environmental variations. Similar to the current finding, Muluken Bantayehu (2013) reported differential response of malt barley genotypes for grain yield across diverse testing environments. Similarly, Sardana and Zhang (2003) reported that grain yield and malt barley grain quality traits were greatly varied with genotypes and influenced by environmental variations and GEI.

Table 3. Analysis of variance (ANOVA) of days to maturity, grain yield and grain quality traits of malt barley genotypes evaluated at five environments.

Source of variation	DF	Mean squares					
		DM	GY	TKW	HLW	PC (%)	SC (%)
Replication	2	0.12	501357	64.3	4.6	0.2	1.0
Genotype (G)	14	270.2**	117012**	195.3**	35.9**	2.9**	8.1**
Environment (E)	4	5249.0**	15079695**	353.7**	447.2**	54.0**	58.7**
GxE	56	26.5**	496941**	16.4ns	21.6**	0.9**	1.0*
Residual	148	1.2	193124	22.7	11.5	0.3	0.5

DF=Degree of Freedom, DM=Days to maturity, GY=Grain Yield, TKW=Thousand Kernels Weight, HLW=Hectolitre Weight, PC=Protein Content and SC=Starch Content

Table 4. Mean performance of malt barley genotypes for days to heading, grain yield and malting grain quality grown at five environments.

Genotypes	DM (days)	GY (kg ha^{-1})	Malt barley grain quality traits			
			TKW (g)	HLW (kg hL^{-1})	PC (%)	SC (%)
G1	144^{i}	1814^{g}	47.4^{b}	58.7^{a-d}	10.6^{c-f}	69.7^{b-e}
G2	138^{d}	2527^{a-c}	35.3^{h}	54.0^{e}	10.4^{ef}	69.2^{c-e}
G3	149^{k}	2176^{c-f}	42.3^{f}	60.1^{ab}	10.1^{f}	72.3^{a}
G4	136^{b}	1960^{fg}	45.4^{b-e}	60.7^{a}	11.6^{ab}	69.9^{b-e}
G5	133^{a}	2215^{c-f}	48.1^{b}	59.1^{a-d}	11.7^{ab}	69.4^{c-e}
G6	141^{g}	2854^{a}	47.1^{bc}	56.8^{d}	11.2^{a-d}	69.9^{b-e}
G7	140^{e}	2255^{c-f}	39.4^{g}	60.4^{a}	11.3^{a-c}	70.4^{b}
G8	144^{i}	2443^{b-d}	42.0^{f}	59.1^{a-d}	11.2^{a-d}	70.2^{bc}
G9	138^{d}	2435^{b-d}	50.7^{a}	59.4^{a-c}	11.8^{a}	69.1^{e}
G10	147^{j}	2232^{c-f}	43.6^{d-f}	57.1^{cd}	10.5^{d-f}	69.7^{b-e}
G11	143^{h}	2713^{ab}	44.6^{c-f}	59.2^{a-c}	10.7^{c-f}	70.5^{b}
G12	137^{c}	1991^{e-g}	43.9^{d-f}	59.7^{ab}	11.6^{ab}	69.1^{e}
G13	140^{f}	2110^{d-g}	43.7^{d-f}	59.6^{ab}	11.0^{b-e}	70.1^{b-d}
G14	142^{g}	2192^{c-f}	46.3^{b-d}	57.9^{b-d}	11.8^{a}	69.4^{c-e}
G15	140^{ef}	2354^{c-e}	42.8^{ef}	59.6^{ab}	11.8^{a}	69.2^{de}
Mean	141	2285	44.0	58.8	11.1	69.9
CV %	0.7	8.9	7.5	3.2	6.4	1.2

GxE interaction analysis using AMMI model

AMMI analysis of variance (AMMI ANOVA) for grain yield of fifteen malt barley genotypes at five testing environments is presented in Table 5. AMMI ANOVA showed highly significant variations ($p<0.01$) due to genotypes, environments and GEI, accounting 15.7%, 57.7% and 26.6% of the total treatment sum squares, respectively. The significant GEI indicates the differential performance of malt barley genotypes across diverse environments. Environmental variation contributed more than half of the total treatment sum of squares. The highest sum of squares attributed due to environmental effects explains the presence of diverse environments with large differences in environmental means. In agreement with the current finding, Kadhem and Baktash (2016), Kang and Gauch (1996), Oliveira *et al*. (2014) reported large environmental variations, accounting for 60-80% of the total variations.

The GEI sum of square was further partitioned into three Interaction Principal Component Axes (IPCA) scores, predicting about 88% of the GEI sum of squares (Table 5). The first principal component axis (IPCA1) accounted about 40% of the GEI sum of squares in 30.3% of the GEI degrees of freedom, while the second principal component axis (IPCA2) captured about 32% of the GEI sum of squares (Table 5). Whereas, IPCA3 explained about 16% of the GEI sum of squares. IPCA1 and IPCA2, which

jointly accounted 72% of the GEI sum of squares, explained the interaction sum of squares and accurately predicted the model, whereas IPCA3 captured mostly noise and couldn't explain the interaction (Kadhem and Baktash, 2016).

Table 5. Additive Main Effect and Multiplicative Interactions (AMMI) Analysis of Variance (ANOVA) for malt barley grain yield across environments.

Sources of variations	DF	SS	MS	SS explained (%)	SS accumulated (%)
Total	224	136580092	609733		
Treatments	74	104529164	1412556**		
Genotypes (G)	14	16381689	1170121**	15.7	
Environments (E)	4	60318779	15079695**	57.7	
Block	10	5013568	501357**		
GE Interactions	56	27828696	496941**	26.6	
• IPCA 1	17	11131478	654793**	40	40
• IPCA 2	15	8905183	593679**	32	72
• IPCA 3	13	4452591	342507*	16	88
• Residuals	24	3339444	139144		
Pooled error	140	27037360	193124		

DF= degree of freedom, IPCA= Interaction Principal Component Analysis, SS= Sum of squares and MS= Mean of squares

AMMI selection of malt barley genotypes across environments

The top four AMMI selection of malt barley genotypes across the testing environments is presented in Table 6. Of the tested malt barley genotypes, G_6 ranked first in all the environments, except at Estayish 2014, showing its consistent performance at Geregera and Kon areas. On the other hand, G_{11} ranked first once (at Estayish 2014) and second twice (at Geregera 2013 and Geregera 2014), implying its best adaptability at Estayish areas. Therefore, G_6 is adaptable for Geregera, Kon and similar environments, and hence advanced to variety verification trial for further evaluation. On the other hand, G_{11} showed relative tolerance to cold stress and thus out-performed in grain yield at Estayish. G_{11} was found susceptible to scald (major barley leaf disease) and thus was excluded from further evaluation under variety verification trial.

Table 6. First four AMMI selections per environment.

Environment	GY(kg ha^{-1})	IPCA score	Genotypic rank			
			1st	2nd	3rd	4th
Geregera 2013	1574	9.75	**G_6**	**G_{11}**	G_8	G_9
Kon 2013	1783	15.84	**G_6**	G_8	G_7	G_{10}
Geregera 2014	2532	-19.78	**G_6**	**G_{11}**	G_5	G_{15}
Kon 2014	2934	-27.13	**G_6**	G_2	G_4	G_8
Estayish 2014	2601	21.32	**G_{11}**	G_2	G_9	G_{14}

AMMI biplot analysis

AMMI biplot analysis of grain yield for AMMI1 (main effects versus IPCA1) and AMMI2 (IPCA1 vs IPCA2) of 15 malt barley genotypes evaluated at five environments is graphically presented in Fig. 2 and Fig. 3, respectively. AMMI1 biplot was constructed using main effects (mean grain yield of genotypes over all environments and mean grain yield of environments of all genotypes) on the x-axis (abscissa) and interaction (IPCA1 of both genotypes and environments for grain yield) on y-axis (ordinate). AMMI1 sensibly clustered the additive main effects into four quadrants. Genotypes and environments which are located at the left hand side of the ordinate (2^{nd} and 3^{rd} quadrant of the biplot) yielded below average, while those located at the right hand side of the ordinate (1^{st} and 4^{th} quadrant of the biplot) yielded above average. G_6, G_{11}, G_2, G_8, G_9 and G_5 are located at the right hand side of the biplot graph, implying their adaptability and yielded above average. The rest of the tested malt barley genotypes yielded below average (Fig. 2). Similarly, environments plotted at the 1^{st} and 4^{th} quadrant of the biplot are considered high yielding environments. Accordingly, Geregera 2014, Kon 2014 and Estayish 2014 were plotted at the right hand side of the ordinate, showing 2014 cropping season was good environment for malt barley production compared to 2013 cropping season.

With regard to the IPCA1 score, genotypes were plotted near or away from the abscissa, indicating the degree of their GEI. The closer the genotypes to the abscissa of the biplot graph, the lower the GEI contribution and the stable the genotype. Accordingly, G_2, G_9, G_{11}, G_8 and G_5 had fairly low IPCA1 score and thus plotted near the abscissa, confirming small interaction with variable environments. On the other hand, G_{13}, G_4, G_{10}, G_1, G_3 and G_6 had sizable IPCA1 score (either positive or negative) and hence plotted away from the abscissa (Fig. 2), indicating their sensitivity to environmental variability. Therefore, the performance of G_{13}, G_4, G_{10}, G_1, G_3 and G_6 for grain yield was significantly affected by variable environments. With regard to the environments, Geregera 2013 and Kon 2013 exhibited relatively low IPCA1 score, indicating their small contribution to the GEI. Other environments showed strong GEI (Fig. 2).

AMMI2 biplot was plotted using IPCA1 (on x-axis) and IPCA2 (on y-axis). Genotypes and environments plotted near the origin of the AMMI2 biplot had low IPCA1 and IPCA2 scores and regarded as stable (Oliveira *et al*., 2014). Thus, G_5 and G_9 relatively had low IPCA1 and IPCA2 scores and

plotted near the origin of the AMMI2 biplot (Fig. 3), indicating their grain yield stability. On the other hand, G_4, G_2, G_{12}, G_7 and G_{11} possessed substantial IPCA1 and IPCA2 scores, implying their sensitivity to environmental variability. Both AMMI1 and AMMI2 analysis commonly identified G_5 and G_9 as the most stable and relatively high-yielding malt barley genotypes under variable environments. From this study, the result of AMMI1stability was not consistent with what was found in AMMI2 stability model (Fig. 2 and Fig. 3). Unlike AMMI1 (which considers only IPCA1), AMMI2 stability model contains both IPCA1 and IPCA2 scores. Therefore, the result of AMMI2 stability might be more accurate than AMMI1 stability. The current finding is in agreement with previous report of Oliveira *et al*. (2014). Based on AMMI1 and AMMI2 stability analyses model, G_6 was found high-yielding and plotted closely with Geregera 2014 and Kon 2014 environments (Fig. 3), indicating their specific adaptability to Geregera, Kon and similar environments. On the other hand, G_{11} (high-yielding genotypes) was plotted with Estayish 2014. Because of its relative cold tolerance, G_{11} out-performed in grain yield over the rest of the tested malt barley genotypes and thus it was the winning genotype at Estayish and similar areas. However, G_{11} was susceptible to scald disease and thus excluded from variety verification trial.

The vector length of the environmental markers from the origin of AMMI2 biplot reveals the contribution of the environment to GEI. Kon 2014 and Estayish 2014 relatively had long vectors and plotted away from the origin of the AMMI2 biplot (Fig. 3), indicating their main contribution to GEI.

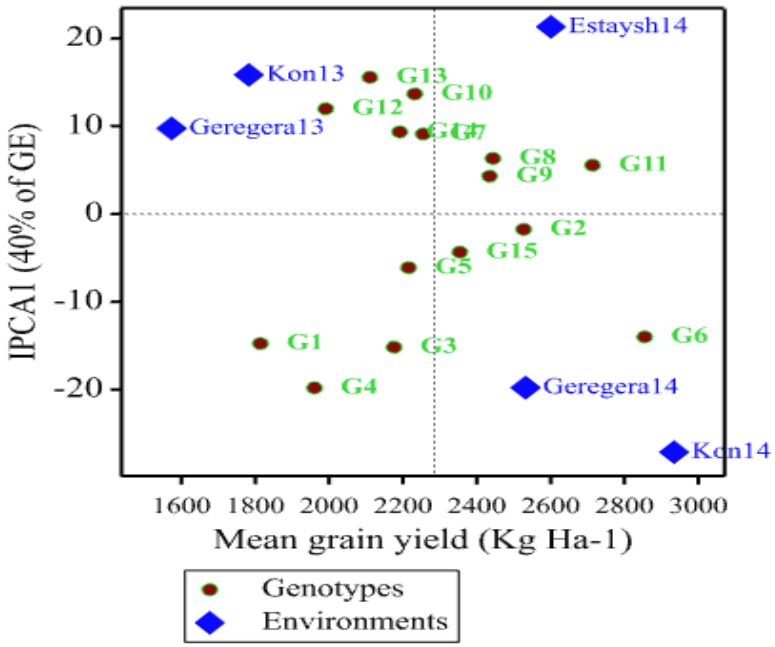

Fig. 2. Biplot analysis of Malt barley genotypes using AMMI1 model.

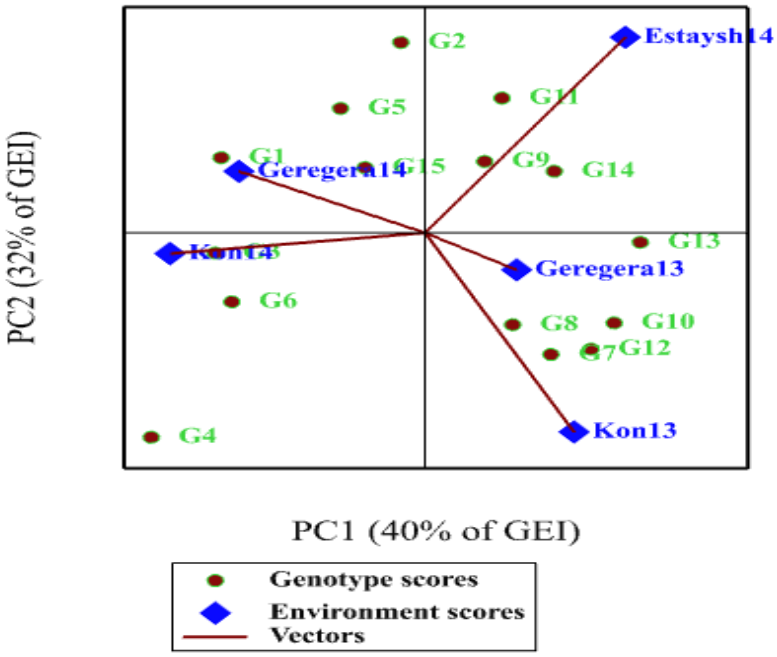

Fig. 3. Biplot analysis of Malt barley genotypes using AMMI2 model.

Malt barley grain quality-related traits

Grain quality is the major criterion to select malt barley genotypes. Barley grains that didn't meet the minimum grain malting standard will not qualify for malting. Grain quality is quantitatively inherited complex trait that is determined by genetic and environmental factors (Pržulj *et al*., 2013). The tested malt barley genotypes were evaluated for thousand kernels weight (TKW), hectolitre weight (HLW), grain protein (PC%) and starch (SC%) contents.

Thousand kernels weight

Kernel plumpness and kernel size is one of the major malt barley grain quality parameters. Often, export certificates require TKW as an indicator of kernel size. Uniform and plumped kernels allow for consistent processing and for high malt extract (Kumar *et al*., 2013). Based on the spike architecture, barley genotypes can be grouped in to two-row or six-row. Both two-rowed and six rowed barley types have three spikelets at each node of the rachis. Unlike six-rowed barley varieties, only the central spikelet is fertile in two-rowed barley. Six-rowed barley usually has smaller and variable kernel size and shape than two-rowed barley varieties. Because of uniform kernel size and symmetrical kernel shape, two-rowed barley grain is generally preferred for malting though six-rowed barley varieties are preferred in some markets (Kumar *et al*., 2013). The plumped the kernel is, the higher the quality of the malt barley grain.

Genotypes exhibited highly significant ($p<0.01$) variation for thousand kernels weight (Table 3). Mean thousand kernels weight (TKW) of the tested malt barley genotypes ranged from 35.3 g (G_2) to 50.7 g (G_9) (Table 4). With regard to TKW, malt barley grains are divided into three groups; bold kernels (>45 g), medium kernels (41–44 g) and small kernels (37–40 g) (Kunze, 2004). In this study, most of the tested malt barley genotypes were classified either bold or intermediate kernel size (Table 4). The minimum international standard of TKW for two- and six-rowed malt barley grain should be >45 g and >42 g, respectively (Biadge Kefale and Yadesa Abushu, 2017; Kumar *et al*., 2013). Smaller grain generally has higher grain protein but low starch contents, yielding low extract. On the other hand, bold kernel has low grain protein and higher starch contents, resulting in more extracts (Edney *et al*., 2005; Fox *et al*., 2003; Kumar *et al*., 2013). In this study, G_9 (50.7 g), G_5 (48.1 g), G_1 (47.4 g), G_6 (47.1 g), G_{14} (46.3 g) and G_4 (45.4 g) fulfilled the minimum malt barley grain TKW standard. The phenotypic expression of TKW was significantly influenced by

environmental variations (Table 3). In harmony with the current finding, Muluken Bantayehu (2013) reported the presence of genotypic variation among malt barley genotypes and the significant influence of environmental variations in expressing malt barley TKW.

Hectolitre weight

Hectolitre weight (HLW) measures the density of grains. It is an important predictor of flour extraction rate of cereal grains. It indicates the effect of pre-harvest sprouting. The higher the HLW is, the sound the grain quality is and the higher the extract yield is. HLW could be effectively used to shortlist large number of malt barley breeding lines at initial screening stage (Verma *et al*., 2008).

In this study, ANOVA depicted the presence of highly significant variation ($p<0.01$) among malt barley genotypes for HLW (Table 3). HLW of malt barley genotypes ranged from 54 kg hL^{-1} (G_2) to 60.7 kg hL^{-1} (G_7). The minimum Ethiopian national standard for malt barley grains HLW ranged from 48–62 kg hL^{-1} (Amare Aleminew and Adane Legas, 2015; Minale Liben *et al*., 2011), indicating all the tested malt barley genotypes fulfilled the minimum Ethiopian malt barley grains HLW standard.

Protein content

Protein content (PC) is one of the most important malt grain quality traits in selecting superior genotypes for malting and brewing. Grain protein is strongly correlated with other malt quality parameters. Thus, there is a need to select genotypes possessing stable grain protein content (Cai *et al*., 2013; Fox *et al*., 2003). The tested malt barley genotypes showed highly significant variation for PC% ($p<0.01$) (Table 3). Grain PC% ranged from 10.1% (G_3) to 11.8% (G_{14}) (Table 4). Malt barley with high grain PC% yields lower extracts (Biadge Kefale and Yadesa Abushu, 2017) and slows down water uptake during steeping and potentially affecting final malt quality. On the other hand, malt barley genotypes with very low grain protein content is deficient in enzymatic activity necessary to modify the barley kernel and to break down the starch during brewing and impairs the brewing performance due to poor yeast amino acid nutrition. The desirable national standard authority for malt barley grains protein content to qualify for malting ranged from 9.0–11.0% for two-rowed and 9.0–11.5% for six rowed barley (Kumar *et al.*, 2013). Protein content lower than 9.0% and greater than 11.5% for two-rowed barley grains is undesirable for malting.

Based on the grain protein standard, G_1, G_2, G_5, G_6, G_7, G_8 and G_{12} fulfilled the minimum specific standard at all environments, except at Estayish 2014. The rest of the tested malt barley genotypes, however, were inconsistent in grain protein content across environments and hence undesirable for malting. As PC is a quantitative trait, environmental variations significantly influence the grain protein content (Muluken Bantayehu, 2013; Cai *et al*., 2013). It is affected by genotype, cultural practices and growing environments (Kumar *et al*., 2013). Grain protein content at Estayish was generally higher than the malting grain PC quality standard. Since Estayish is cold-prone area, the high grain PC might be associated with the prevalence of severe cold stress, especially at grain filling stage. Abiotic stress generally triggers the expression of defense chemicals such as amino-acids, proteins and other physiological defense mechanisms. In response to low temperature, genotypes might tend to synthesize protectant amino-acids like proline that elevates the grain protein content beyond the minimum standard (Dar *et al*., 2016; Verbruggen and Hermans, 2008).

Starch content

The breaking down of starch by enzymatic reaction during the malting process resulted in fermentable sugars. Highly significant variations ($p<0.01$) was observed among malt barley genotypes for grain starch content. Moreover, grain starch content was significantly ($p<0.01$) influenced by environmental variations (Table 3). Mean grain starch content ranged from 69.1–72.3% (Table 4). In agreement with the current finding, Fox *et al*. (2003) reported the significant influence of growing environments on barley grain starch content and extract yield. Mean grain yield, thousand kernels weight, hectolitre weight, grain protein content and extract yield of the candidate genotype and standard check are presented in Table 7.

Table 7. Summary of grain yield of candidate genotype over the standard check at different environments (at Geregera, Kon and Estayish in 2013 and 2014 cropping seasons).

Genotypes	**GY (Kg ha^{-1})**	**TKW (g)**	**HLW (Kg hL^{-1})**	**PC (%)**	**EC (%)**
G_6	20–34	43–52	52–62	7–10.2	76–79
Standard check	15–30	39–49	53–60	8.2–11	76–77
Ethiopian standard		**25–30**	**48–62**	**9.12**	**76 min**
World standard		**35–45**	**60–65**	**8.5–12**	**79–82**

Where, GY = Grain Yield, TKW = Thousand Kernels Weight, HLW = Hectolitre Weight, PC = Protein Content and EC = Extract Content

CONCLUSION

Currently, brewing industries are booming in Ethiopia. The fast growing malting and brewing industries in Ethiopia necessitates the development of high yielding and stable malt barley varieties, possessing acceptable malting grains quality traits. Fifteen malt barley genotypes were evaluated for grain yield and grain quality traits at major barley growing areas of North Wollo, Ethiopia for two consecutive cropping seasons. Significant variations were observed among genotypes for grain yield and grain quality traits. AMMI stability analysis for grain yield depicted that G_{11} was relatively tolerant to cold stress and thus out-smarted in grain yield over the tested malt barley genotypes at Estayish. However, G_{11} was susceptible to scald disease and thus excluded from further evaluation. On the other hand, G_6 was found agronomically superior in grain yield at Geregera and Kon areas and fulfilled the minimum TKW, HLW and PC quality standards. Moreover, it took 135–145 days to reach physiological maturity, making it suitable for terminal moisture-stressed environments.

G_6 (candidate genotype) along with the standard check was further evaluated for agronomic and malt quality traits at North and South Wollo major barley growing areas in 2017 and 2018 cropping seasons. Both G_6 and the standard check recorded extract content ranging from 76–79% at different locations. Thus, the candidate genotype met the minimum Ethiopian malt barley grain standard for extract content. Therefore, G_6 (Libra T95/Diamalt) was found superior over the rest of the tested malt barley genotypes for both agronomic and malt barley grain quality traits under terminal moisture-stress environments of North Wollo and similar environments, and officially released by the National Variety Releasing Committee (NVRC) in 2019 cropping season for commercial production with a local name called "Waro".

ACKNOWLEDGEMENTS

The research was funded by the Ethiopian Institute of Agricultural Research (EIAR) through the Sirinka Agricultural Research Centre.

REFERENCES

Amare Aleminew and Adane Legas (2015). Grain quality and yield response of malt barley varieties to Nitrogen fertilizer on brown soils of Amhara Region, Ethiopia. *World J. Agric. Sci.* **11**(3): 135–143.

Biadge Kefale and Yadesa Abushu (2017). Malt quality profile of malt barley varieties grown in the Central Highlands of Ethiopia. *Int. J. Bioorg. Chem*. **2**(3): 130–134.

Brouwer, C. and Heibloem, M. (1986). **Irrigation Water Management: Irrigation Water Needs**. Training manual no. 3. FAO.

Cai, S., Yu, G., Chen, X., Huang, Y., Jiang, X., Zhang, G. and Jin, X. (2013). Grain protein content variation and its association analysis in barley. *BMC Plant Biol*. **13**(35): 1–11.

Dar, M.I., Naikoo, M.I., Rehman, F., Naushin, F. and Khan, F.A. (2016). Proline accumulation in plants: roles in stress tolerance and plant development In: **Osmolytes and Plants Acclimation to Changing Environment**. Emerging Omics Technologies, 155–166 (Iqbal, N., Nazar, R. and Khan, N.A., eds.). Springer.

Deo, K. Mishra, S., Singh, A. Mishra, A. and Singh, A. (2017). Water requirement of wheat crop for optimum production using CROPWAT model. *J. Med. Plants Stud.* **5**(3): 338–342.

Ding, M., Tier, B. and Yan, W. (2007). Application of GGE biplot analysis to evaluate genotype (G), environment (E) and GxE interaction on *P. radiata*: A case study. Australasian Forest Genetics Conference 11–14 April 2007, the Old Woolstore, Hobart, Tasmania.

Edney, M., Izydorczyk, M.S., Symons, S. and Woodbeck, N. (2005). Measuring barley kernel colour and size to predict and use malt quality. Grain Research Laboratory/Industry services collaborative study on valuing barley, Canadian Grains Commission, Winnepeg.

Fan, X.-M., Kang, M.S., Chen, H., Zhang, Y., Tan, J. and Xu, C. (2007). Yield stability of maize hybrids evaluated in multi-environment trials in Yunnan, China. *Agron. J.* **99**(1): 220–228.

Fox, G.P., Panozzo, J.F., Li, C., Lance, R., Inkerman, P.A. and Henry, R.J. (2003). Molecular basis of barley quality. *Austral. J. Agric. Res.* **54**(12): 1081–1101.

Gauch H.G. (1992): **Statistical Analysis of Regional Yield Trials: AMMI Analysis of Factorial Designs**. Elsevier, Amsterdam.

Gomez, A.K. and Gomez, A.A. (1984). **Statistical Procedures for Agricultural Research**. Second edition. John Wiley and Sons, New York.

Kadhem, F. and Baktash, F. (2016). AMMI analysis of adaptability and yield stability of promising lines of bread wheat (*Triticum aestivum* L.). *Iraqi J. Agric. Sci.* **47**(7): 35–43.

Kang, M.S. (1997). Using genotype by environment interaction for crop cultivar development. *Adv. Agron*. **62**: 199–252.

Kang, M.S. and Gauch, Jr, H.G. (1996). **Genotype-By-Environment Interaction**. CRC press, Boca Raton.

Karimizadeh, R., Mohammadi, M., Sabaghni, N., Mahmoodi, A. A., Roustami, B., Seyyedi, F. and Akbari, F. (2013). GGE biplot analysis of yield stability in multi-environment trials of lentil genotypes under rainfed condition. *Not. Sci. Biol.* **5**(2): 256–62.

Kumar, D., Kumar, V., Verma, R., Kharub, A. and Sharma, I. (2013). Quality parameter requirement and standards for malt barley: A Review. *Agric. Rev*. **34**(4): 313–317.

Kunze, W. (2004). **Technology Brewing and Maltin**g. Third edition. VLB Berlin.

Minale Liben, Alemayehu Assefa and Tilahun Tadesse (2011). Grain yield and malting quality of barley in relation to Nitrogen application at mid and high altitude in Northwest Ethiopia. *J. Sci. Develop*. **1**(1): 75–88.

Muluken Bantayehu (2013). Study on malting barley genotypes under diverse agroecologies of North Western Ethiopia. *Afr. J. Plant Sci.* **7**(11): 548–557.

Oliveira, E., Freitas, J. and Jesus, O. (2014). AMMI analysis of the adaptability and yield

stability of yellow passion fruit varieties. *Sci. Agr*. **71**(2): 139–145.

Pržulj, N., Momčilović, V. and Crnobarac, J. (2013). Path coefficient analysis of quality of two-row spring barley. *Genetika* **45**(1): 21–30.

Sardana, V. and Zhang, G. (2003). Important quality attributes of malt barley (*Hordeum vulgare* L.): Effect of genetics, environments and agronomic factors. *Agric. Sci. China* **2**: 815–824.

Steel, R.G.D. and Torrie, J. H. (1980). **Principles and Procedures of Statistics: A Biometrical Approach**. Second edition, McGraw-Hill Kogakusha, New York.

Verbruggen, N. and Hermans, C. (2008). Proline accumulation in plants: A review. *Amino acids.* **35**(4):753–759.

Verma, R., Kharub, A., Kumar, D., Sarkar, B., Selvakumar, R., Singh, R., and Sharma, I. (2011). Fifty years of coordinated barley research in India. Directorate of Wheat Research, Research Bulletin, 27:46.

Verma, R., Sarkar, B., Gupta, R. and Varma, A. (2008). Breeding barley for malting quality improvement in India. *Cereal Res. Comm*. **36**(1): 135–145.

Yan, W., Kang, M.S., Ma, B., Woods, S. and Cornelius, P.L. (2007). GGE biplot vs. AMMI analysis of genotype-by-environment data. *Crop Sci.* **47**(2): 643–653.