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Performance Evaluation and Identification of Highland Quality Protein Maize Hybrids in Ethiopia

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Authors' contributions

This work was carried out in collaboration between both authors. Author GMM collected the data, performed the analysis, and developed the manuscript. Author ATC reviewed and made editorial comments on the draft manuscript. Both authors read and approved the final manuscript.

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ABSTRACT

Background: Maize plays an indispensable role in meeting high food demand. It is globally one of the most widely adopted and cultivated crops. Hybrid development from fixed inbred lines is one of the strategies for the improvement of maize production. The national average maize yield is low in East Africa; thus, the selection of promising germplasm has a great role to meet the high food demand of growing the population. Forty-two Quality Protein Maize (QPM) crosses (21 inbred lines each crossed with two testers) along with three popular standard hybrids checks were evaluated in two replications using alpha lattice during the 2017 cropping season at Ambo, Arsi-Negele, and Kulumsa. The objective of this study was to evaluate and select the best new QPM hybrids for grain yield, and other agronomic and morphological characteristics.

Results: Significant difference among crosses was observed for 19 traits at Ambo, 14 traits at Arsi-Negele, and 19 traits at Kulumsa. Out of 28 traits studied, six of them did not show genotype by location interaction but they showed a significant genotypic effect. These traits were: Days to Silking (DS), Number of Ears per plant (EPP), Ear Length (EL), Kernels Per Row (KPR), Ear Diameter (ED), and Thousand Seed Weight (TSW). Based on the mean performance in the combined analysis, from 42 new QPM crosse, six of them (L8xT2, L7xT1, L8xT1, L19xT1, L6xT2, and L18xT1) were scored higher grain yield compared with the mean of the three standard checks and best conventional maize (CM) check (AMH853). L8xT2, L7xT1, L8xT1 crosses showed a higher grain yield advantage over the best check (AMH853) by 20.87, 14.13, and 13.63%, respectively.

Conclusion: The study implicated the existence of a difference between the newly developed hybrids and the standard check varieties. In general, the study enabled us to identify promising crosses that could be forwarded for further use in maize breeding programs in future work.

Keywords: Maize; quality protein; cross; conventional maize.

1. INTRODUCTION

Maize is one of the cereal crops which has adapted to different agro-ecologies and produces widely around the globe. In 2021, worldwide production of maize was around 1,205.35 MT with a world average of 5.95 t ha 1 . The world maize production area also covered around 202.72 million hectares [1]. Maize is a very productive, adaptable, versatile, and most important food security crop in Sub Saharan Africa (SSA); Eastern and Southern Africa use 85% of maize produced as food while Africa as a whole use 95% as food (Bekele et al*.,* 2011). In 2017, worldwide production of maize was around 1042.4 MT. Its' production took 40% share of all cereals and 25% of the land allocated for cereals [2]. The largest share, 37% (384.8 MT) was held by the USA which continued to be the largest maize producing country in the world. In Africa, in terms of maize production area coverage, Nigeria (6 million hectares) took the leading and followed by Tanzania (4.1 mh) and South Africa (3mh). Ethiopia (2.4 mh) is the fourth maize producer in Africa in terms of area coverage [1]. Ethiopia is the third leading country for the production of maize in Africa next to South Africa and Nigeria [2].

Despite its importance, maize yield in Africa is lower (4.30 t/ha) which is computed from major maize producing African countries compared to the world average of 5.95 t/ha [1]. In Ethiopia, too, the national average maize yield is low compared to the world average grain yield [2]. This is due to several biotic and abiotic stresses that limit maize productivity across countries in sub-Saharan Africa [3]. Among abiotic stresses, drought and low soil fertility are the most important stresses that affect maize production [4,5,6].

Maize is one of the five strategic crops for food security in Ethiopia. In 2018, maize was grown on 21% of the total cereals area and it ranked 2nd following teff (30%) in terms of total production contributing 31% of the total cereals grain produced in the country. Wasihun and Desu [7] also reported the increasing trend for maize production area from the 2007-2018 production season according to the central statics authority (CSA) data. About 8.4 MT of maize is produced from 2.1 million hectares with an average yield of 3.94 t ha⁻¹ [8]. Of all the smallholder cereals framers in the country, 70% grow maize in variable scales [8].

The maize crop is an important source of protein, although its protein is low in essential amino acids such as Lysine (Lys) and Tryptophan (Trp) [9,10]. It is also a source of minerals, vitamin B, iron, and carbohydrate [11]. Maize grain protein has long been known to have low nutritional value and research on improving the nutritional value of maize grain protein was started before one century [12]. While millions of people worldwide are overly dependent on maize as a staple food, this nutritional deficiency caused for kwashiorkor is a concern in the area where maize is a staple food, particularly for people with high protein requirements [13,14]. The nutritional superiority of Quality Protein Maize (QPM) to Conventional Maize (CM) has been amply demonstrated in rats [15,16], pigs [17,18,19], infants and small children [20,19,21] as well as adults [22], (Clark *et al.,* 1977), dairy cattle (Glover, 1992), pregnant or lactating women, and the ill [21] in countries where maize is a staple and is the main protein source. The term QPM refers to maize genotypes whose Lys and Trp levels in the endosperm of the kernels are about twice higher than in CM varieties [22]. QPM has a nutritional advantage over CM. QPM contains the *o2* mutation, which alters the protein composition of the maize endosperm, resulting in increased concentrations of Lys and Trp [23]. Consumption of QPM may help alleviate human malnutrition problems in regions with maizebased diets [22,24] because of the 60 to 100% increase in the concentration of Lys and Trp [22].

In Sub-Saharan Africa, where maize is the major source of calories and the existence of malnutrition, emphasis has been given to the introduction and development of QPM varieties as a means to solve malnutrition caused due to heavy dependence on maize as a source of protein. In Africa or elsewhere the most followed QPM breeding strategy relies on the conversion of existing adapted genotypes to QPM [25,26].

Adapted CM genotypes that resist major biotic and abiotic stresses are converted to QPM mostly following backcrossing or modified backcross breeding methods [27].

Elite QPM inbred lines well adapted to eastern and southern African regions are being developed by CIMMYT-Zimbabwe, CIMMYT-Kenya, and CIMMYT-Ethiopia (at Ambo, EIAR). Dagne [28] and Adefris et al. [27] pointed out that as converted QPM inbred lines, OPVs and hybrids can be available to national research programs, and other private and public research organizations from CYMMYT, and other parts of the world. So that, using this genetic resource, QPM hybrid development efforts can be fruitful in developing the nutritionally balanced maize varieties to contribute to solutions to the problem of food and nutrition insecurity. The relative performance of inbred lines in test crosses with appropriate testers has proven useful to select inbred lines with good combining abilities and superior performance in hybrids [29]. Although currently available elite QPM inbred lines have been tested in hybrid combinations with selected lines and testers, selection of suitable parents for hybridization, identification of promising hybrids, and the development of stable improved maize hybrids and open-pollinated synthetic varieties with high yield for diverse agroecologies [30]

The national maize improvement program of Ethiopia has been working to address maize technologies' need for the mid-altitude, low moisture-prone, and highland agro-ecologies. Highland maize breeding program in collaboration with CIMMYT has developed a large number of highland elite maize inbred lines. Recently 21 new elite QPM inbred lines have been developed by the highland maize improvement section at Ambo-EIAR. Using these lines 42 single cross hybrids were generated but never tested these hybrids' performance before for any parameters. Therefore, this study was designed to evaluate and compare newly developed 42 F1 crosses generated from new lines crossed using line by tester mating design using CMLL144 and CML 159 as testers.

2. MATERIALS AND METHODS

2.1 Description of Experimental Sites

The study was conducted at three locations in the highland agroecology of Ethiopia including; Ambo, Arsi-Negele, and Kulumsa Agriculture Research Centers in the 2017 main season.

Ambo Agriculture Research Center is located at 8° 57' N latitude, 38 $^{\circ}$ 07' E longitude at an altitude of 2225 masl. It represents the highland subhumid maize growing agroecology of Ethiopia. The soil type is heavy clay (vertisol) with a pH of 7.8 for most topsoil $(0 - 30$ cm) [31]. The longterm total annual rainfall is 1115 mm, and average minimum and maximum temperatures are 11.7 $\mathrm{^{\circ}C}$ and, 25.5 $\mathrm{^{\circ}C}$, respectively with an average value of 18.6 °C.

Arsi-Negele is located at $7^{\circ}19'$ N latitude and 38 $^{\circ}$ 39' E longitude at an altitude of 1960 masl. The long-term annual rainfall is 886 mm with erratic and uneven distribution. The site had mean minimum and maximum temperatures of 9.1 $^{\circ}$ C and 26 \degree C, respectively with an average value of 17.6 $^{\circ}$ C. The soil texture is clay loam with a pH of 6.5-7.5 [32].

Kulumsa is located at 8° 5' N latitude, $39^{\circ}10'$ E longitude at an altitude of 2200 masl. The dominant soil type is luvisol/eutric nitosols with good drainage and a pH of = 6.0 [33]. The total long-term annual rainfall is 830 mm. The mean minimum and maximum temperatures are 10 \degree C and 23.2 \degree C, respectively with an average value of 16.6 \degree C.

2.2 Experimental Materials

Twenty-one highland QPM inbred lines and two elite QPM inbred line testers (CML159 and CML144) constituted the basic genetic materials of this experiment (Table 1). From the 21 inbred lines and the two testers, 42 F1 hybrids were generated in Ambo Highland Maize Breeding Program (AHMBP). Three standard checks: one QPM (AMH852Q) and two CM (Jibat and AMH853), designated as hybrid checks, were tested.

2.3 Experimental Design and Crop Husbandry

The trial was laid out using an alpha lattice design consisting of one-row plots replicated twice. Each plot consisted of a 5.25 m long row with 0.75 and 0.25 cm inter-and intra-row spacing. The plot was hand-planted with two seeds per hill and later was thinned to one plant per hill to attain the final plant density of 53,333 plants per hectare. Diammonium phosphate (DAP) fertilizer was applied all at planting at the rate of 150 kg ha⁻¹ while 200 kg ha⁻¹ of urea was applied in partition 1/3 at planting, 1/3 at knee height, and 1/3 at flowering at Ambo and Kulumsa. At Arsi-Negele, 100 kg ha⁻¹ DAP and

150 kg ha $^{-1}$ urea fertilizer were applied based on the site recommendation following the same time of application mentioned for Ambo and Kulumsa above.

2.4 Data Collected

Data on morphological, phenological, yield, and related yield traits were recorded and presented as follows. Days to tasseling (DT), Days to silking (DS), Anthesis silking interval (ASI), Days to maturity (MD), Plant aspect (PAS), Disease score: gray leaf spot (GLS), turcicum leaf blight (TLB), and common leaf rust (CLR), Ear aspect (EAS), Number of ears per plant (EPP), Kernel Modification (MOD), Grain yield (GY), Number of leaves per plant (LFPP), Number of leaves above uppermost ear per plant (LFAE), Number of leaves bellow uppermost ear per plant (LFBE), Leaf angle (LANG), Leaf length (LL), Leaf width (LW), Leaf area (LFAR), Plant height (PH), ear height (EH), ear length (EL), Ear diameter (ED), Number of kernel rows (NKR), Number of kernels per row (KPR), Thousand seed weight (TSW), Biomass (BIOM) and Harvest index (HI).

Grain yield (t ha⁻¹) = $\frac{fresh \, cob \, weight \, x(100 - MC) \, 0.8 \, x10}{87.5 \, x \, 3.94}$

Where, fresh ear weight $=$ fresh weight of the cob from the plot in kg , 0.8 = shelling percentage, 87.5 = standard value of grain at the moisture content of 12.5% from the total grain mass, $MC = grain$ moisture content (%) at harvest, 3.94 = plot area harvested in meter square (m2).

2.5 Data Analysis

The data obtained from field measurements were organized and analyzed using SAS statistical package [34]. Analysis was conducted using the model of RCBD after confirming the nonsignificance of the block effect which implied there was uniformity among blocks. Accordingly, to testify to the presence of variation among crosses for the trait in question variance was carried out for individual locations and across locations.

2.5.1 Analysis of variance

Individual and across locations data were subjected to analysis of variance using PROC GLM procedure in SAS software version 9.0 (SAS, 2002). In the analysis, treatments were used as a fixed factor while replications and locations were considered random factors. This was specified using the RANDOM statement in the PROC GLM model. A mean separation test was done for traits that expressed differences among treatments using LSD. Combined analysis was done for the significant traits that showed significant differences in each location analysis and testing homogeneity of error variances. Whenever traits were found to be significant at three locations combined based on the ratio of error [35]. In the combined analysis, the variation among genotypes crosses, and checks effects were tested against their respective interaction effect with the location. The interaction effect of each source of variation by location was tested as per the expected mean square (MS) of the error estimate.

3. RESULT AND DISCUSSION

3.1 Analysis of Variance and Mean Contrast

3.1.1 Analysis of variance

Analysis of Variance (ANOVA) for the hybrid trial showed a significant genotypic difference for Grain Yield (GY), Days to Tasseling (DT), Day to Silking, (DS) Plant Height (PH), Ear Height (EH), Ear Per Plant (EPP), Ear Length (EL), Kernel Per Row (KPR), Ear Diameter (ED), Thousand Seed Weight (TSW) and Biomass yield (BIOM) at each of the three locations (Table 2). A similar result was also reported by (Berhanu, 2009). The genotypic difference for Gray Leaf Spot (GLS) and Leaf above uppermost Ear (LFAE) was not significant in any of the three locations.

Variances due to genotype were significant only at Kulumsa for Common Leaf Rust (CLR), Leaf Angle (LANG), and Leaf Area (LFAR), while for Turcicum Leaf Blight (TLB) and Harvest Index (HI) these differences were significant only at Arsi-Negele. For Anthesis Silking Interval (ASI), Kernel Modification (MOD), Plant Aspect (PAS), and Number of Kernel Rows per ear (NKR) difference between the crosses was significant only at Ambo (Table 2). For days to maturity (MD), leaves per plant (LFPP), and leaves below the uppermost ear (LFBE) these differences were significant at two of the three locations.

Genotype by Location interaction was significant for GY, DT, ASI, PH, EH, CLR, TLB, PAS, BIOM, LFPP, and LFBE. It was non-significant for the remaining traits. The result observed in this study is in line with previous reports [36,37,38], (Bitew, 2016) indicating a significant difference among maize genotypes for phenology, yield, yield related, and other agronomic traits. In contrast to the current finding for LFPP, Berhanu [36] reported significant MS. The interaction was significant for hybrids for GY, DT, CLR, TLB, LFPP, and non-significant MD, MOD, EPP, EL, NKR, KPR, ED, HI, and LFAR. Genotype performance in-ear traits (EPP, EL, NKR, KPR, and ED) seem to be stable across environments while performance in GY, DT, CLR, TLB, and LFPP is unstable across environments (Table 2). A significant difference among locations was observed for most of the traits except, for MOD, EAS, NKR, LFPP, and LFBE. Similarly, genotype by location (GxL) interaction was significant for GY, DT, ASI, CLR, PAS and BIOM at $(p < 0.05)$ for EH and TLB at (p <0.01) and for PH, LFPP and LFBE at (p <0.001). Berhanu (2009) also reported similar findings for grain yield, grain yield-related, and other agronomic traits. Gudeta et al. [10] also reported significant GxL interaction for GY and EH. The result in this study contradicted to significant MS difference reported for TSW among the genotypes and the non-significant difference for DT and PH [10]. The non-significant MS for GxL confirmed that genotypes performed consistently across a location for EPP imply that the location effect was not that much which is similar to the report of Berhanu [36].

Check x location interaction showed significant MS for GY (data not shown) indicating the performance of standard checks was not consistent across the location. Significant variation among the environments for most of the traits indicates that each of the target locations

was a unique and similar report by a different scholar [28,36] for yield and yield-related traits were reviewed.

3.1.2 Sum square contribution

Percent contributions of the genotype (G), the Location (L), and GxL interaction to the total sum of the squares of treatment for various traits are presented in the following (Table 3). When the total sum of the square is partitioned to its various sources, the sum square due to location constitutes a preponderance amount for most of the traits followed by genotype. In all traits except, LFPP sum square due to location was a major component contributing to the total sum of squares. The effect of GxL was far less than the location and genotype effect except for LFPP and LFBE. Except for LFBE and LFPP, the sum of squares due to genotype was the 2^{nd} most important contributor to the total sum of the squares. DT showed a high percent sum square contribution for genotype as compared to the remaining traits (Table 3). Single crosses are more sensitive to environments than other types of crosses and open-pollinated cultivars [39]. Dange (2008) also reported a higher effect of location on QPM crosses studied.

3.1.3 Mean comparison in combined data analysis

In the combined analysis, GY ranged from 3.79 (L13xT1) to 9.67 t ha⁻¹ (L8 xT2) with an overall mean of 7.00 t ha⁻¹. EPP ranged from 1.01 to 1.91 with a mean of 1.32. The mean EPP of crosses was similar to the mean EPP of hybrid checks (1.3 EPP for cross and 1.3 for check) (Table 4). The mean EPP of the top five crosses was higher than the mean of checks (AMH852Q, AMH853, and Jibat), mean of CM checks (Jibat and AMH853), best CM check (AMH853), and QPM check (AMH852Q) by 12%, 15%, 21%, and 8%, respectively. Cross (L8xT2) which had the highest EPP) also had higher EPP by 8%, 11%, 17%, and 4% as compared with the EPP value of the mean of all hybrid checks, mean of hybrid CM checks, best CM check and QPM check (AMH852Q), respectively. EPP was not affected by the environment and similarly, Berhanu [36] reported non-significant GxL for this trait.

L16xT2 had the lowest DS (94.0 days) while L1xT1 had the highest (106 days). The mean value of standard checks was less than the mean of crosses for DS indicates that checks are earlier than crosses in general. This result

Table 2. MS due to genotypes and error for grain yield and other traits in 21x2 LxT crosses of QPM evaluated at Ambo, Arsi-Negele, and Kulumsa Agricultural Research Centers, 2017

Table 2. (Continued)

** = significant at 0.05 probability level, **= significant at 0.01probabilty level and *** = significant at 0.001probabilty level, DF = Degree of freedom, GxL= Genotype by location interaction, GY = Grain yield (t/ha), DT = Days to tasseling (days), DS = Days to silking (days), ASI = Anthesis Silking Interval (days), MD = Days to Maturity (days), PH = Plant Height (cm), EH = Ear Height (cm), MOD = Kernel Modification (1-5 scoring), GLS = Gray Leaf Spot (1-5 scoring), CLR = Common Leaf Rust (1-5 scoring), TLB = Turcicum Leaf Blight (1-5 scoring), EAS = Ear Aspect (1-5 scoring), PAS = Plant Aspect (1-5 scoring), EPP = Ear Per Plant (number), EL= Ear Length (cm), NKR = Number of Kernel Rows (number), KPR = Kernel Per Row (number), ED = Ear Diameter (cm), TSW = Thousand Seed Weight (gram), BIOM = Biomass yield (t/ha), HI = Harvest Index (%), LANG = Leaf Angle (degree), LL = Leaf Length (cm), LW = Leaf Width (cm), LFAR = Leaf Area (cm²), LFPP =Leaf Per Plant (number), LFAE = Leaf above upper most ear (number), LFBE = Leaf bellow upper most ear (number)*

Table 3. Percent sum square contribution by genotype, GxL interaction, location, rep (location), and error in combined analysis for traits showed a significant GxL effect

Source of variation	DF	GY	DT	ASI	РH	EН	CLR	TLB	PAS	BIOM	∟FPP	LFBE
Genotype	44	29.38	40.47	21.73	21.64	25.18	11.70	3.75	21.93	18.73	19.48	13.10
Genotype*location	88	12.96	5.52	22.34	6.16	4.65	10.87	10.10	24.33	15.58	36.57	23.67
∟ocation		47.14	47.66	34.34	66.81	65.07	65.20	74.29	28.54	47.90	0.43	27.58
Rep (location)		. . 71	0.51	0.10	l.14	0.64	.36	2.66	1.27	2.05	14.27	17.10
Model	137	91.18	94.16	78.51	95.75	95.55	89.13	90.79	76.07	84.25	70.74	81.45
Error	132	8.82	5.84	21.49	4.25	4.45	10.87	9.20	23.93	15.75	29.26	18.55
Total	269	100.00	100.00	100.00	100.00	100.00	100.00	100.00	99.99	100.00	100.00	100.00

Table 4. Selected traits for the10 top-yielding QPM hybrids and standard checks were evaluated across three locations in Ethiopia

Table 5. Mean Square contrast between new crosses and standard checks and the estimated value of mean difference for selected traits which are not included in the combined analysis in LxT crosses of maize by location (Ambo, Arsi-Negele, and Kulumsa Agricultural Research Centers), 2017

agreed with the report of Elmyhum [40] and Demissew [31] who reported that QPM hybrids were late in flowering than the CM hybrid check. With the prevalence of frost and inadequate amount and distribution of rainfall in tropical highlands, breeders should focus on developing early flowering maize varieties with acceptable grain yield [31]. Earliness is one of the main breeding objectives in areas with limited water resources and short growing seasons [41].

The highest and lowest TSW mean values were 214.1 (L3xT1) to 424.9 (L17xT2) with an overall mean of 313.2 g (Table 4). Generally, the mean of checks showed higher performance than the mean of crosses (Appendix 1). The overall mean of genotypes in this study was almost similar in magnitude to the overall mean result reported by Berhanu [36] but a little smaller than reported by Demissew [31] and Beyene [38]. The latter two authors reported 347.7 g and 358.7g TSW in a different set of crosses, but while considering among the crosses, the highest TSW was obtained in this study as compared to the report made by Berhanu [36], Demissew [31] and Beyene [38]. Out of 42 crosses, 57.1% had a higher mean TSW than the overall mean TSW of genotypes.

For ED, the mean of crosses (4.42 cm) and mean of standard checks (4.42cm) were almost equal whereas for EL the mean of standard checks (16.65 cm) was higher than the mean of crosses (14.29 cm). The highest EL (18.1cm) was obtained from the standard check (Jibat) (Table 4). The mean of KPR for checks (35.69) was greater than the mean of crosses (32.14). KPR mean value ranged from 25.67 to 39.44 kernels with an overall mean of 32.37. Cross L8xT2 which is a high yielder and also took the 1st rank for KPR with the value of 39.44. Most of the crosses which are included in the top-yielding crosses also had high KPR (Table 4). The higher mean of checks for KPR than the mean of crosses is in line with the report of Abiy [33] who reported the higher mean performance by standard checks than the mean of crosses.

3.1.4 Comparison of test-crosses mean with mean of hybrid checks

A) Individual location

In the one-degree freedom analysis (orthogonal contrast); GY, DT, ASI, EH, PH, MOD, EAS, PAS, NKR, BIOM, and LFBE showed significant MS for cross vs mean of checks. Except, for ASI

and PAS, NKR, and LFBE the same traits mentioned above showed significant MS for cross vs best check at Ambo (Table 5). At Arsi-Negele, only DT and LFPP had significant MS for cross vs mean of check whereas, for cross vs best check, DT, EH, PH, and TLB showed significant MS. At Kulumsa, cross vs means of check showed significant differences for DT, EH, PH, CLR, LANG, LL, LFAR, and LFPP.GY, DT, LANG, and LFPP showed significant differences for cross vs best check (Table 5).

For GY, the mean of crosses had less performance than the mean of hybrid checks and best check by 1.61 and 3.21 t ha⁻¹ at Ambo. At Kulumsa, the mean of crosses lowered by 0.37 t ha $^{-1}$ and 2.25 t ha $^{-1}$ compared with the mean of checks and best check, respectively. At Ambo, the mean of crosses was inferior to the mean of checks and best check by 4.11 and 8.04 t ha $^{-1}$ for BIOM, respectively (Table 5). This result is in line with the lower performance of the mean of crosses compared with the mean of checks reported by [33]. Regarding HI, in terms of magnitude, the mean of crosses has exceeded the mean of checks by 8.16% and is lower than the best check by 9.81% at Arsi-Negele.

The mean of crosses was higher than the mean of checks by 6.0, 5.18, and 10.54 days at Ambo, Arsi-Negele, and Kulumsa for DT, respectively. This implies that crosses were relatively late for male flowering as compared to the mean of checks in overall observation. This finding is in agreement with the result of Demissew [31]. Late maturity is not a favorable trait to look for because early maturity could help to escape some natural stresses like random drought. The mean difference of crosses over best check was 9.83, 7.35, and 13.71 days. The estimated mean difference of crosses over the best check was almost zero (0.07days) but it was higher than the mean of checks by 1.23 days for ASI at Ambo (Table 5).

The difference between the mean of crosses with mean checks best indicated that the crosses are shorter for EH and PH in general at three locations. Based on this, the crosses' mean was shorter by 5.98, 2.96, and 13.25 cm compared with the mean of checks at Ambo, Arsi-Negele, and Kulumsa, respectively similarly the mean of crosses was shorter than the best check by 25.65, 17.63 and 6.58cm at three locations. For PH, the mean of crosses was shorter by 1.82,1.72 and 14.15 cm compared with the mean of checks whereas it was shorter by 31.82,21.55 and 9.15 cm compared with the best check at Ambo, Arsi-Negele, and Kulumsa respectively. Generally, the result indicates the breeding program is successful in reducing the plant stature compared to recently released varieties (Table 5).

For MOD, the mean of cross exceeded the mean of checks (by 1.30) and the best check (by 1.05) at Ambo. This shows that checks were good for kernel outside appearance but there were crosses with better endosperm modification than the standard checks while evaluating each genotype separately. For PAS and EAS, the mean of crosses exceeded the mean of checks and best check (AMH853) by the magnitude of 0.38 over the mean of checks and best check. The magnitude of the mean difference was 0.35 and 0.68 over the mean of checks and best check for EAS, respectively. This implies that the mean of checks and best check were better than the mean of crosses which is manifested by the lower value of PAS and EAS at Ambo. The mean of crosses was higher by 2.36 and 1.02 kernel rows over the mean of checks and best check variety (AMH853), respectively at Ambo. This indicates the breeding program is successful in the improvement of kernel row number (Table 5) which is one of the yield components.

At Arsi-Negele, the mean of crosses was less in magnitude by 0.61 for TLB indicating that crosses were more tolerant as compared to the best check (Jibat). The mean of crosses exceeded the mean of checks by 0.23 meaning that crosses were slightly attacked by this disease as compared to standard checks in overview observation. At Kulumsa, the mean of crosses was higher than the mean of checks and best check by 0.51 and 0.35 for CLR, respectively. This over view result also explain that crosses were slightly attacked by CLR than standard check varieties (Table 5).

At Kulumsa, the mean of checks and the best check (Jibat) had a higher mean value over the mean of crosses by 12.8 and 13.17° for LANG, respectively. This implies that crosses were characterized by narrow leaf angles in general. For LL, LW, and LFAR, the mean of crosses had a higher value than the mean of checks by 6.06 cm, 0.29 cm, and 69.01 cm², respectively. The mean of crosses also exceeded the best check (Jibat) by 4.23 cm and 34.83 cm² for LL and LFAR, respectively but it was almost equal in magnitude to the best check for LW (Table 5). For LFPP, the mean of crosses was higher than the mean of checks by 0.91 leaves but it was

almost equal to the value of 0.42 as compared to the value of the best check at Ambo. At Kulumsa, the mean of crosses was higher than the mean of checks and best check by 0.61 and 0.95 respectively. For LFBE, the mean of crosses was lower in magnitude by 1.29 and 0.45 leaves compared with the mean of checks and best check, respectively at Ambo (Table 5).

B) Across locations

Orthogonal contrast between the mean of the 42 test-crosses and the mean of the three check hybrids and with the best check revealed a significant difference in combined analysis for GY, DT, DS, PH, EH, EAS, MOD, EL, NKR, KPR, and TSW. MS of cross vs means of checks also showed significant differences for CLR, PAS, and LANG. Cross vs best check showed significance for ASI, LW, and LFAE. In contrast, the orthogonal contrast between cross vs means of checks and cross vs best check was non-significant for GLS, TLB, EPP, HI, ED, LL, LFPP, LFAR, and LFBE. Moreover, cross vs means of checks showed non-significant variation for ASI and LFAE. CLR, PAS, and LANG had also a non-significant MS for cross vs best check. Similarly, Berhanu [36] reported significant cross vs mean checks similar to this study result for DT, DS, EH, and TSW. Abiy [33] also reported a significant difference between crosses vs mean of checks for BIOM, HI, TSW, NKR, EL, and ED. Due to the presence of significant MS for GxL interaction effect for most of the traits, orthogonal contrast was carried out only in six traits (DS, EPP, EL, KPR, ED, TSW) considered in the combined analysis of variance (Table 6).

In the combined analysis, the estimate of the mean difference is presented in Table 7 for six traits. The mean of crosses was higher than the mean of checks and the best check for DS. This indicates that the crosses were late for female flowering than the checks and best check. The mean of the crosses exceeded the mean of checks and the best check by 7.46 for and 6.31 days for DS, respectively (Table 7).

For TSW, the mean of checks and best check exceeded the mean of crosses by 45.27 and 51.58g, respectively. This result is in contrast to the report of Abiy [33] who reported relatively higher mean permeance by crosses than the mean of checks TSW (4.4 g). The mean of checks and the best check have exceeded the mean of crosses by 3.54 and 2.47 for KPR. The

Source of Variation	DF	MS							
		GY	DS	EPP	EL	KPR	ED	TSW	
Rep (Location)		$7.96***$	17.90*	0.13	$12.57**$	15.69	$0.70***$	2634.36	
Location		$329.7**$	1159.35**	$4.61**$	294.57*	669.62**	$8.57*$	235157.73**	
Genotype	44	$9.34***$	$71.47***$	$0.27***$	$18.31***$	59.84***	$0.50***$	13595.74***	
Cross	41	$9.67***$	50.82***	$0.29***$	16.89***	57.99***	$0.53***$	13699.93***	
Check		0.38	11.56	0.04	$9.82*$	21.86	0.06	1039.38	
Cross vs Check		$13.59***$	1038.19***	0.001	93.55***	211.59***	0.08	34436.88***	
Cross vs best check		$6.62**$	233.01***	0.07	24.50**	35.9	0	15592.55***	
Genotype x Location	88	$2.05***$	6.79	0.05	2.97	15.61	0.06	1519.21	
Cross x Location	82	$1.81***$	5.82	0.05	3.09	15.64	0.07	1451.95	
Check x Location	4	$4.85*$	12.81	$0.11*$	0.65	8.26	0.03	2948.70*	
pooled error crosses	123	0.87	5.13	0.06	2.34	12.68	0.05	1350.83	
pooled error genotypes	132	0.93	5.05	0.05	2.42	12.81	0.05	1320.95	
pooled error checks	6	0.67	3.61	0.02	4.7	18.93	0.07	630.48	

Table 6. ANOVA table for mean square combined across three locations for tested genotypes, 2017

= significant at 0.05 probability level, **= significant at 0.01probabilty level, * = significant at 0.001probabilty level, DF = Degree of freedom, GY Grain Yield, MS =Mean Square, DS = Days to Silking, EPP = Ear Per Plant, EL= Ear Length, KPR = Kernels Per Row, ED = Ear Diameter, TSW = Thousand Seed Weight*

Table 7. MS of contrast between new crosses and standard checks and the estimated value of mean difference for grain yield and other traits in combined analysis from LxT crosses of maize evaluated at Ambo, Arsi-Negele, and Kulumsa Agricultural Research Centers, 2017

mean difference of crosses and mean of checks and mean of crosses with that of the best check showed that the mean difference value was in the negative direction meaning that the mean of crosses was less by 2.35 cm and 2.04 cm than the mean of checks and best check for EL (Table 7). Abiy [33] also reported a lower mean of crosses than the mean of checks for KPR, EL, and narrow ED.

4. CONCLUSION

In the combined analysis, grain yield, yield related, and agronomic parameters showed significant difference variation between genotypes except for MD, GLS, TLB, NKR, HI, LANG, LW, LFAR, LFPP, LFAE, and LFBE. GxL interaction was significant for grain yield and
indicates the existence of inconsistent the existence of inconsistent performance by genotypes across locations. For grain yield-related traits, the absence of a significant interaction effect highlights that the genotypes were consistently performed across a location for these traits. In all traits except, LFPP sum square due to location was a major component contributing to the total sum of squares. The effect of GxL was far less than the location and genotype effect except for LFPP and LFBE. From the new hybrids, L8XT2, L7XT1, and L8XT1 were well performed compared with the standard QPM and CM hybrid check varieties. For grain yield and most yieldrelated traits, the orthogonal contrast between cross vs means of checks and cross vs best checks showed that crosses had inferior performance however for flowering data higher value by mean of the cross against the standard checks highlighted the new crosses were relatively late. However, while considering each new cross with the standard checks in the pairwise matrix, some crosses were better than the checks. In general, the study result indicates that the breeding program is in good progress in terms of generating new QPM hybrids which might be released for commercial use in the future.

DISCLAIMER

The products used for this research are commonly and predominantly used products in our area of research and country. There is no conflict of interest between the authors and producers of the products because we do not intend to use these products as an avenue for any litigation but the advancement of knowledge. Also, the research was not funded by the

producing company rather it was funded by the personal efforts of the authors.

ETHICS APPROVAL

The researchers have obtained permission from funding institutions CIMMYT and EIAR. Accordingly, the information under this article had been developed in collaboration with CIMMY, Ethiopian Institute of agriculture research investigators.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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APPENDIX

Appendix 1. Mean of each hybrid by 28 traits combined over three locations Ambo, Arsi-Negele, and Kulumsa Agricultural Research Centers, in 2017

Appendix1 (Continued)

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Appendix1 (Continued)

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