

# Per se performance evaluation and identification of new quality protein maize inbred lines

## Abstract

**Background:** Maize plays a critical role in meeting high food demand. It is a globally widely adopted and cultivated crop. Hybrid and open-pollinated varieties development from fixed inbred lines is one of the strategies for the improvement of maize production. Compared with the world average, the national average maize yield is low in Ethiopia. According to this development and selection of promising germplasm has indispensable value for developing high-yielding maize varieties. The study consists of 21 Quality Protein Maize (QPM) lines, two QPM testers lines, and one Conventional Maize (CM) line check (FS67). They were evaluated in RCBD with two replications at Ambo and Arsi-Negele. The objective was to identify new lines with good performance compared with released QPM checks and CM lines. **Result:** There is a significant difference between the lines for 28 traits in Ambo and 24 traits at Arsi-Negele. In combined mean performance analysis, the highest yielding line (L8) exceeded the mean of all line checks, mean QPM checks, CM line check (FS67), and best QPM line check (CML144) by 34.89%, 54.80%, 7.30%, and 25.31%, respectively for GY. The value of EPP ranged from 0.91 (L14) to 1.85 (L3) with an overall mean of 1.19). The highest yielding line (L8) had the 2nd highest EPP (1.63). Mean EPP of the top five QPM lines was less by 5.45%, 4.96%, 22.41%, and 6.41% compared with the mean of all checks, mean of QPM line checks, best QPM check, and CM line check, respectively. The high yielder line (L8) had a higher mean value than the mean of lines checks (CML144, CML159, and FS67), mean of QPM lines checks, best QPM check (CML144), and FS67 by 25.71%, 26.36%, 3.16%, and 24.43%, respectively.

**Conclusion:** In general, the study confirmed the existence of promising new QPM parental lines. These promising lines can be used as source material in the breeding program for further improvement.

**Keywords:** Quality protein maize, conventional maize, maize inbred lines

## 1. INTRODUCTION

Maize (*Zea mays* L.  $2n = 20$ ) belongs to the family Gramineae, and the tribe Maydeae (Andropogoneae) (Norman *et al.*, 1995). 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 (FAO, 2017). The largest share, 37% (384.8 MT) was held by the USA and continued to be the

40 largest maize-producing country in the world. Africa contributes 7.6% of the global maize  
41 production area; Nigeria, Egypt, Ethiopia, and South Africa are the front-runner countries in  
42 order of importance (FAO, 2015). Ethiopia is the third leading country for the production of  
43 maize in Africa next to South Africa and Nigeria (FAO, 2017).

44 Despite its importance, maize yield in sub-Saharan Africa has stagnated at less than 2 t ha<sup>-1</sup>  
45 compared to the world average of more than 5 t ha<sup>-1</sup>. In Ethiopia, too, the national average  
46 maize yield is low compared to the world average (5.85 t ha<sup>-1</sup>) (FAO, 2017). This is due to  
47 several biotic and abiotic stresses that limit maize productivity across countries in sub-  
48 Saharan Africa (Badu-Apraku *et al.*, 2011). Drought and low soil fertility are the most  
49 prominent abiotic stresses which affect maize production (Mosisa *et al.*, 2007; Lobell *et al.*,  
50 2011; Weber *et al.*, 2012).

51 Maize is one of the five strategic crops for food security in Ethiopia. In 2018, maize covers  
52 21% of the total cereals area of production. It ranked 2<sup>nd</sup> following teff (30%) in terms of  
53 total production contributing 31% of the total cereals grain produced in the country. About  
54 8.4 MT of maize is produced in 2.1 million hectares with an average yield of 3.94 t ha<sup>-1</sup>  
55 (CSA, 2018). Of all the smallholder cereals framers in the country, 70% grow maize in  
56 variable scales (CSA, 2018).

57 The maize is an important source of protein, although its amount of protein is low in  
58 essential amino acids such as Lysine (Lys) and Tryptophan (Trp) (Mbuya *et al.*, 2011;  
59 Gudeta *et al.*, 2015). It is also a source of minerals, vitamin B, iron, and carbohydrate (Rouf  
60 Shah *et al.*, 2016). While millions of people worldwide are overly dependent on maize as a  
61 staple food, this nutritional deficiency caused for kwashiorkor is a concern in areas where  
62 maize is a staple food, particularly for people with high protein requirements (Bain *et al.*,  
63 2013; Morley, 2016). The nutritional superiority of Quality Protein Maize (QPM) to  
64 Conventional Maize (CM) has been amply demonstrated in rats (Bressani *et al.*, 1969; Gupta  
65 *et al.*, 1970), pigs (Lopez-Pereira, 1992; Osei *et al.*, 1994a, and Atlin *et al.*, 2011), infants  
66 and small children (Bressani, 1995; Atlin *et al.*, 2011 and Tekeba, 2017) as well as adults  
67 (Bressani,1992), dairy cattle (Glover, 1992), pregnant or lactating women, and the ill  
68 (Tekeba, 2017) in countries where maize is a staple and is the main protein source. The term

69 QPM refers to maize genotypes whose Lys and Trp levels in the endosperm of the kernels  
70 are about twice higher than in CM varieties (Bressani, 1992). QPM has a nutritional  
71 advantage over CM. QPM contains the *o2* mutant gene, which alters the protein composition  
72 of the maize endosperm, resulting in increased concentrations of Lys and Trp (Mertz *et al.*,  
73 1964). Consumption of QPM may help alleviate human malnutrition problems in regions  
74 with maize-based diets (Bressani, 1999; Mertz, 1992) because of the 60 to 100% increase in  
75 concentration Lys and Trp (Bressani, 1992).

76 In Sub-Saharan Africa, where maize is the major source of calories and the existence of  
77 malnutrition, emphasis has been given to the introduction and development of QPM varieties  
78 as a means to solve malnutrition. This is caused due to heavy dependence on maize as a  
79 source of protein. In Africa or elsewhere the most followed QPM breeding strategy relies on  
80 the conversion of existing adapted genotypes to QPM (CIMMYT, 2004b, and Krivanek *et*  
81 *al.*, 2007). Adapted CM genotypes that resist major biotic and abiotic stresses are converted  
82 to QPM mostly following backcrossing or modified backcross breeding methods (Adefris *et*  
83 *al.*, 2015).

84 Elite QPM inbred lines well adapted to eastern and southern African regions are being  
85 developed by CIMMYT-Zimbabwe, CIMMYT-Kenya, and CIMMYT-Ethiopia (at Ambo,  
86 EIAR). Dagne (2008) and Adefris *et al.* (2015) pointed out that as converted QPM inbred  
87 lines, OPVs and hybrids can be available to national research programs, and other private  
88 and public research organizations from CYMMYT, and other parts of the world. Thus, using  
89 such genetic resources and the development of QPM hybrids development can help develop  
90 nutritionally balanced maize varieties to reduce food and nutrition insecurities. Although  
91 currently available elite QPM inbred lines have been tested in hybrid combinations with  
92 selected lines and testers, systematic classification of the continuously introduced lines into  
93 different heterotic groups has not been conducted. Line's information regarding mode of  
94 gene action, combining abilities, heritability and heterosis would be useful in the  
95 development of inbred lines and selection of suitable parents for hybridization for the  
96 development of suitable maize hybrids and open-pollinated synthetic varieties with high  
97 yield adaptable to diverse agro-ecologies (Abebe *et al.*, 2004).

98 Highland maize breeding program in collaboration with CIMMYT has developed a large  
99 number of highland elite maize inbred lines. The effort is aimed at identifying better  
100 combining inbred lines for the development of hybrids for highland areas of the country. To  
101 initiate an effective hybrid breeding program, information on the combining ability of inbred  
102 lines and identifying the lines' heterotic group is an essential and critical factor. Recently 21  
103 new elite QPM inbred lines have been developed by the highland maize improvement  
104 section at Ambo-EIAR. Thus, the objective of this study was to assess the *per se*  
105 performance of new QPM lines and their level of tolerance to biotic stresses in comparison  
106 with the well-known QPM and CM line checks.

## 107 **2. Materials and Methods**

### 108 **2.1. Description of Experimental Sites**

109 The study was conducted at two locations in the highland agroecology of Ethiopia Ambo and  
110 Arsi-Negele (transition highland) Agriculture Research Centers during the 2017 main  
111 cropping season.

112 **Ambo** Agriculture Research Center is located at 8° 57' N latitude, 38° 07' E longitude at an  
113 altitude of 2225 masl. It represents the highland sub-humid maize growing agroecology of  
114 Ethiopia. The soil type is heavy clay (vertisol) with a pH of 7.8 for most topsoil (0 - 30 cm)  
115 (Demissew, 2014). The long-term total annual rainfall is 1115 mm, and average minimum  
116 and maximum temperatures are 11.7°C and, 25.5°C, respectively with an average value of  
117 18.6 °C.

118 **Arsi-Negele** is located at 7°19' N latitude and 38° 39' E longitude at an altitude of 1960  
119 masl. The long-term annual rainfall is 886 mm with erratic and uneven distribution. The site  
120 had mean minimum and maximum temperatures of 9.1 °C and 26 °C, respectively with an  
121 average value of 17.6 °C. The soil texture is clay loam with a pH of 6.5-7.5 (Etagegnehu and  
122 Amare, 2016).

### 123 **2.2. Experimental Materials**

124 Twenty-one highlands new QPM inbred lines named hereafter as lines (L1 to L21) and two  
125 elite QPM inbred lines checks (CML159 and CML144), named hereafter as QPM checks (T1  
126 and T2, respectively), and one elite CM highland inbred line (FS67) check was evaluated  
127 into two locations (Table 1).

### 128 **2.3 Experimental Design and Crop Husbandry**

129 The trial was laid out using a randomized complete block design (RCBD) consisting of one-  
130 row plots replicated twice. Each plot consisted of a 5.25 m long row with 0.75 and 0.25 cm  
131 inter-row and intra-row spacing. The plot was hand-planted with two seeds per hole and later  
132 was thinned to one plant per hole to attain the final plant density of 53,333 plants per hectare.  
133 Diammonium phosphate (DAP) fertilizer was applied all at planting at the rate of 150 kg ha<sup>-1</sup>  
134 while 200 kg ha<sup>-1</sup> of urea was applied in partition 1/3 at planting, 1/3 at knee height, and 1/3  
135 at flowering at Ambo. At Arsi-Negele, 100 kg ha<sup>-1</sup> DAP and 150 kg ha<sup>-1</sup> urea fertilizer were  
136 applied based on the site recommendation following the same time of application mentioned  
137 for Ambo above.

### 138 **2.4. Data Collected**

139 Data on morphological, phenological, yield and related yield traits were recorded on plot and  
140 individual plant basis at each location as specified below.

141 Days to tasseling (DT), Days to silking (DS) Anthesis silking interval (ASI) Days to maturity  
142 (MD), Plant aspect (PAS) Disease score: gray leaf spot (GLS), turicum leaf blight (TLB),  
143 and common leaf rust (CLR), Ear aspect (EAS), Number of ears per plant (EPP), Kernel  
144 Modification (MOD) Grain yield (GY), Number of leaves per plant (LFPP), Number of  
145 leaves above uppermost ear per plant (LFAE), Number of leaves bellow uppermost ear per  
146 plant (LFBE), Leaf angle (LANG), Leaf length (LL), Leaf width (LW), Leaf area (LFAR),  
147 Plant height (PH), ear height (EH), ear length (EL), Ear diameter (ED), Number of kernel  
148 rows (NKR), Number of kernels per row (KPR), Thousand seed weight (TSW), Biomass  
149 (BIOM) and Harvest index (HI).

150 Grain yield (t ha<sup>-1</sup>) =  $\frac{\text{fresh cob weight} \times (100 - MC) \times 0.8 \times 10}{87.5 \times 2.81}$

151 *Where, fresh ear weight = fresh weight of the cob from the plot in kg, 0.8 = shelling*  
152 *percentage, 87.5 = standard value of grain at a moisture content of 12.5% from the total grain*  
153 *mass, MC = grain moisture content (%) at harvest, 2.81 = plot area harvested in meter square*  
154 *(m<sup>2</sup>).*

## 155 **2.5 Data Analysis**

156 The data obtained from field measurements were organized and analyzed using SAS  
157 statistical package (SAS, 2002). Accordingly, to test the presence of variation among inbred  
158 lines for the trait in question analysis was carried out for individual locations and across  
159 locations.

### 160 **2.5.1. Analysis of variance**

161 Before data analysis, the anthesis-silking interval (ASI) was normalized using  $\ln(\text{ASI} + 10)$   
162 as suggested by Bolaños and Edmeades (1996). Individual and across locations data were  
163 subjected to analysis of variance using PROC GLM procedure in SAS software version 9.0  
164 (SAS, 2002). In the analysis, treatments were used as fixed factors while replications and  
165 locations were considered as random factors. This was specified using the RANDOM  
166 statement in the PROC GLM model. A mean separation test was done for traits that  
167 expressed differences among treatments using LSD. Combined analysis was done for the  
168 significant traits that showed significant differences in each location analysis and test  
169 homogeneity of error variances. Whenever traits were found to be significant at two  
170 locations combined based on the ratio of error (Gomez and Gomez, 1984). In the combined  
171 analysis, the variation among genotypes and checks effects were tested against their  
172 respective interaction effect with a location. The interaction effect of each source of variation  
173 by location was tested as per the expected mean square (MS) of the error estimate.

174

175

176

177

178 **Table 1.** List of parental inbred lines and standard checks used.

Code	Pedigree	Remark
L1	[CML144/[CML144/CML395] F2-8sx]-1-2-3-2-B*5-1-B-B-B-#	QPM
L2	[CML144/[CML144/CML395] F2-8sx]-1-2-3-2-B*5-2-6-B-B-#	QPM
L3	(CLQRCWQ50/CML312SR)-2-2-1-BB-1-B-B-B-#	QPM
L4	[CML144/[CML144/CML395] F2-8sx]-1-2-3-2-B*5-1-B-B-B-#	QPM
L5	([NAW5867/P49SR(S2#)]/NAW5867] F#-48-2-2-B*/CML511) F2)-B-B-39-1-B-#	QPM
L6	(CML197/(CML197/[(CLQRCWQ50/CML312SR)-2-2-1-BB/CML197]-BB) F2)-B-B-9-1-B-#	QPM
L7	(CML197/(CML197/[(CLQRCWQ50/CML312SR)-2-2-1-BB/CML197]-BB) F2)-B-B-35-2-B-#	QPM
L8	(CML197/(CML197/[(CLQRCWQ50/CML312SR)-2-2-1-BB/CML197]-BB) F2)-B-B-44-2-B-#	QPM
L9	(CML197/(CML197/(CLQRCWQ50/CML312SR)-2-2-1-BBB) F2)-B-B-18-2-B-#	QPM
L10	(CML197/(CML197/(CLQRCWQ50/CML312SR)-2-2-1-BBB) F2)-B-B-30-1-B-#	QPM
L11	(CML197/(CML197/(CLQRCWQ50/CML312SR)-2-2-1-BBB) F2)-B-B-35-2-B-#	QPM
L12	(CML395/(CML395/[NAW5867/P49SR(S2#)]/NAW5867] F#-48-2-2-B*4) F2)-B-B-30-1-B-#	QPM
L13	[CML144/[CML144/CML395] F2-8sx]-1-2-3-2-B*5-2-6-B-B-#	QPM
L14	(CML395/(CML395/[CML144/[CML144/CML395] F2-8sx]-1-2-3-2-B*5) F2)-B-B-46-1-B-#	QPM
L15	(CML395/(CML395/[CML144/[CML144/CML395] F2-8sx]-1-2-3-2-B*5) F2)-B-B-50-1-B-#	QPM
L16	(CML395/(CML395/S99TLWQ-B-8-1-B*4-1-B) F2)-B-B-10-3-B-#	QPM
L17	(CML395/(CML395/S99TLWQ-B-8-1-B*4-1-B) F2)-B-B-14-1-B-#	QPM
L18	(CML395/(CML395/S99TLWQ-B-8-1-B*4-1-B) F2)-B-B-29-1-B-#	QPM
L19	(CML395/(CML395/CML511) F2)-B-B-7-2-B-#	QPM
L20	(CML395/(CML395/CML511) F2)-B-B-11-2-B-#	QPM
L21	(CML395/(CML395/CML511) F2)-B-B-37-1-B-#	QPM
T1	CML144-Check1	QPM
T2	CML159-Check2	QPM
	FS67-Check3	CM

179

### 180 3. RESULT AND DISCUSSION

#### 181 3.1. Analysis of variance

182 Individual location ANOVA showed significant differences among genotypes for all traits at  
 183 both locations except for GLS, CLR, PAS, NKR, and KPR at Ambo and MOD and TLB at  
 184 Arsi-Negele (Table 2). The difference between the 24 lines was non-significant for MD and  
 185 HI at both locations. In the combined analysis, only HI showed a non-significant difference  
 186 among genotypes (Table 2) which is in line with the report of Dufera et al. (2018). In the  
 187 combined analysis, the difference between 21 QPM lines was significant for GY, DT, DS,  
 188 ASI, PH, EH, TLB, EL, KPR, ED, LFPP, and LFBE. Regarding QPM checks, the difference  
 189 was significant in combined analysis for DT, DS, ASI, EH, CLR, EPP, EL ED, TSW, and

190 LW but for the rest of the traits the difference was non-significant (Table 2). Similarly,  
191 Demissew (2014) reported significant differences in grain yield and yield-related and  
192 phenological traits among QPM inbred lines. Dagne et al. (2008) also reported significant  
193 MS for PH, MD, and GY.

194 The Genotype by Location interaction was significant for GY, DT, DS, GLS, CLR, TLB,  
195 EAS, TSW, LANG, LFPP, and LFAE and non-significant for the remaining traits. This  
196 indicates that with the traits with a significant difference in interaction, the performance was  
197 unstable across the location. Berhanu (2009) and Dufera et al. (2018) also reported similar  
198 findings for grain yield, grain yield-related, and other agronomic traits. Gudeta et al. (2015)  
199 also reported significant GxL interaction for GY. The result in this study contradicted to  
200 significant MS difference reported for TSW among the genotypes and the non-significant  
201 difference for DT and PH (Gudeta et al., 2015).

202

203

204

205

206

207 Table 2. MS due to genotype (lines) and error for grain yield and other traits of maize evaluated at Ambo and Arsi-Negele Agricultural Research  
 208 Centers, 2017.

Trait	Mean Square									
	Ambo		Arsi-Negele		Across locations (Ambo and Arsi-Negele)					
	Genotype (DF=23)	Error (DF=23)	Genotype (DF=23)	Error (DF=23)	Location (DF=1)	Genotype (DF=23)	G X L (DF=23)	Error (DF=46)	QPM line (DF=20)	QPM check (DF=1)
GY	1.19**	0.32	1.60***	0.23	14.23*	2.28***	0.53*	0.28	13.59*	6.00
DT	35.77***	3.00	17.39***	5.30	5265.84*	44.30***	8.86*	4.15	62.59*	1102.1***
DS	41.59***	6.65	18.08***	4.26	4401.04**	45.72***	13.95**	5.45	50.74*	644.48***
ASI	5.39*	2.04	4.38*	1.96	38.76	6.30***	3.48	2.00	5.78*	61.01***
MD	5.91	3.25	7.74	3.87	19238.34***	8.28**	5.37	3.56	11.82	2.29
PH	506.02***	57.86	567.20***	55.78	6600.17	998.58***	74.64	56.82	3092.17***	682.12
EH	380.83***	48.34	347.82***	47.33	2223.38	684.75***	43.92	47.84	1641.54***	4484.00**
MOD	0.69**	0.25	0.59	0.48	5.27	0.77*	0.51	0.36	0.73	0.40
GLS	0.01	0.01	0.11*	0.05	3.56*	0.06*	0.06*	0.03	0.04	0.05
CLR	0.09	0.09	0.92**	0.25	64.19**	0.68***	0.34*	0.17	0.63	5.43**
TLB	0.15*	0.08	0.21	0.14	15.84*	0.16	0.20*	0.11	0.16*	0.12
EAS	0.42*	0.17	0.88***	0.21	3.76	0.96***	0.35*	0.19	0.46	1.83
PAS	0.24	0.13	0.51*	0.19	1.76*	0.56***	0.19	0.16	0.38	1.08
EPP	0.15**	0.05	0.08***	0.02	0.60	0.20***	0.04	0.03	0.18	5.99***
EL	4.21**	1.16	4.21*	1.59	5.59	6.93***	1.49	1.38	24.65**	61.01**
NKR	2.35	1.54	4.83**	1.30	0.51	5.49***	1.68	1.42	3.44	4.59
KPR	17.81	13.31	28.84**	7.97	59.38	34.69***	11.96	10.64	95.06**	4.15
ED	0.28***	0.05	0.22***	0.03	0.09	0.46***	0.05	0.04	0.62*	4.81***

209

210

211

212

213 **Table 2 (Continued)**

Trait	Mean Square									
	Ambo		Arsi-Negele		Across Locations Ambo and Arsi-Negele)					
	Genotype (DF=23)	Error (DF=23)	Genotype (DF=23)	Error (DF=23)	Location (DF=1)	Genotype (DF=23)	G X L (DF=23)	Error (DF=46)	QPM line (DF=20)	QPM check (DF=1)
TSW	7613.35***	1425.51	5446.06***	610.3	51666.65	10306.21***	2753.21**	1017.91	10496.97	234328.2**
BIOM	9.83*	4.48	6.37*	2.68	13.77	10.72***	5.5	3.58	30.86	44.84
HI	143.51	160.28	97.84	89.69	4220.75	119.83	121.51	124.98	221.59	0.63
LANG	81.19***	13.7	220.03***	16.09	580.17	243.68***	57.55***	14.9	23.08	2.61
LL	118.70***	4.69	157.11***	34.52	4842.67*	244.21***	31.6	19.61	85.97	1.59
LW	1.76***	0.3	2.08**	0.63	126.42***	2.89***	0.96*	0.47	0.39	3.14*
LFAR	8076.17***	1715.4	14656.32**	4851.32	1198421.01**	18097.12***	4635.38	3283.36	7006.99	15019.4
LFPP	1.99***	0.33	1.32*	0.55	9.17	2.39***	0.92*	0.44	2.32*	0.37
LFAE	0.72***	0.1	0.64***	0.08	0.12	1.19***	0.18*	0.09	0.29	0.02
LFBE	1.33***	0.33	1.15**	0.31	7.22	2.01***	0.48	0.32	1.67**	0.37

214  
 215 \*= significant at 0.05 probability level, \*\*= significant at 0.01 probability level and \*\*\* = significant at 0.001 probability level, DF = Degree of freedom,  
 216 GxL= Genotype by location interaction, GY = Grain yield (t/ha), DT = Days to tasseling (days), DS = Days to silking (days), ASI = Anthesis Silking Interval  
 217 (days), MD = Days to Maturity (days), PH = Plant Height (cm), EH = Ear Height (cm), MOD = Kernel Modification (1-5 scoring), GLS = Gray Leaf Spot  
 218 (1-5 scoring), CLR = Common Leaf Rust (1-5 scoring), TLB = Turicum Leaf Blight (1-5 scoring), EAS = Ear Aspect (1-5 scoring), PAS = Plant Aspect (1-5  
 219 scoring), EPP = Ear Per Plant (number), EL= Ear Length (cm), NKR = Number of Kernel Rows (number), KPR = Kernel Per Row (number), ED = Ear  
 220 Diameter (cm), TSW = Thousand Seed Weight (gram), BIOM = Biomass yield (t/ha), HI = Harvest Index (%), LANG = Leaf Angle (degree), LL = Leaf  
 221 Length (cm), LW = Leaf Width (cm), LFAR = Leaf Area (cm<sup>2</sup>), LFPP = Leaf Per Plant (number), LFAE = Leaf above upper most ear (number), LFBE = Leaf  
 222 below upper most ear (number).  
 223  
 224

225

### 226 3.2. Sum Square Contribution

227 The percent sum contribution of the genotype (G), Location (L), and GxL interaction to the  
 228 total sum of the square of treatment for various traits is presented in Table 3. When the total  
 229 sum of squares is partitioned to its various sources, the sum square due to location constitutes  
 230 a preponderance amount for DT, DS, GLS, CLR, and TLB and followed by Genotype for  
 231 DT, DS, GLS, and TLB. However, the sum square contribution was higher for GY, EAS,  
 232 TSW, LFAG, LFPP, and LFAE for genotype. The effect of GxL was far less than the  
 233 location and genotype for DT and DS (Table 3). The higher sum square contribution by the  
 234 interaction (genotype\*location) component cannot be considered a good result because, in  
 235 this case, genotype\*location can be considered an error term. The experiment result having  
 236 results with a small genotype\*location interaction effect is preferable. Similarly, the sum  
 237 square contribution by location to the total sum square contribution will not be good because  
 238 this indicates the existence of higher variability between locations used to test our  
 239 experiment. The higher sum square contribution by genotype effect to the total sum square is  
 240 the one that the researcher going to get from the experiment.

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

Source of variation	DF	GY	DT	DS	GLS	CLR	TLB	EAS	TSW	LANG	LFPP	LFAE
Genotype	23	56.37	14.94	17.27	17.87	16.15	12.33	51.34	58.01	67.20	50.83	76.38
Genotype*location	23	13.07	2.99	5.27	17.87	8.04	15.70	18.51	15.50	15.87	19.44	11.79
Location	1	15.32	77.19	72.28	46.42	66.61	52.46	8.71	12.64	6.96	8.46	0.32
Rep (location)	2	1.62	2.09	1.05	1.70	1.11	1.76	1.21	2.39	1.76	2.44	0.17
Model	49	86.37	97.20	95.88	83.86	91.90	83.20	79.77	88.54	91.78	81.18	88.67
Error	46	13.63	2.80	4.12	16.21	8.10	16.79	20.23	11.46	8.22	18.82	11.33
Total	95	100.00	100.00	100.00	100.00	100.00	99.99	99.99	100.00	100.00	100.00	100.00

243

### 244 3.3. Mean comparison in combined data analysis

245 In the combined analysis, GY ranged from 2.10 to 5.0 t ha<sup>-1</sup> with an overall mean value of  
 246 3.04 t ha<sup>-1</sup> (Table 4). The mean of the top five lines had higher mean performance over the  
 247 means of all checks (CML144, CML159, and FS67) and the mean of QPM checks (CML  
 248 144 and CML 159) by 5.32% and 20.87%, respectively. The mean of the top five QPM lines

249 had an inferior performance of 2.16% and 16.22% over the best QPM line check (CML144)  
250 and CM line check (FS67), respectively. The highest yielding line (L8) exceeded the mean of  
251 all line checks, mean QPM checks, CM line check (FS67), and best QPM line check  
252 (CML144) by 34.89%, 54.80%, 7.30%, and 25.31%, respectively for GY.

253 The value of EPP ranged from 0.91 (L14) to 1.85 (L3) with an overall mean of 1.19 (Table  
254 4). The highest yielding QPM line (L8) had the 2<sup>nd</sup> highest EPP (1.63) (Table 4). The mean  
255 EPP of the top five QPM lines was less by 5.45%, 4.96%, 22.41%, and 6.41% compared  
256 with the mean of all line checks, mean of QPM line checks, best QPM check, and CM line  
257 check, respectively. The high yielder line (L8) had a higher mean value than the mean of  
258 lines checks (CML144, CML159, and FS67), mean of QPM lines checks, best QPM check  
259 (CML144), and FS67 by 25.71%, 26.36%, 3.16%, and 24.43%, respectively. This indicates  
260 there was a promising line that can be used as source material in the breeding program for  
261 further research.

262 For EL the value ranged from 9.17 g (L13) to 14.50 cm (L10) (Table 4). The mean of new  
263 QPM lines and the mean of QPM line checks had almost equal EL values of 11.29 and 11.27  
264 cm, respectively. For ED, the values ranged from 2.91g (L3) to 4.41 cm (L13). Like that of  
265 the value for EL, the mean of QPM line checks and QPM line checks had almost equal  
266 performance (Table 4).

267 At Ambo, considering the new QPM lines for TSW the highest and lowest value was  
268 obtained from L3 (139.23 g) and L17 (318.55 g) but from the overall genotypes tested, the  
269 highest value was obtained from FS67 with values of 364.16 g and 410.99 g at Ambo and  
270 Arsi-Negele, respectively (Table 5 and 6). The mean of new QPM lines and the overall mean  
271 showed almost equal values in magnitude. From the new QPM lines, the highest value  
272 (353.50 g) was obtained from L18 at Arsi-Negele (Table 6). For NKR and KPR, the highest  
273 value was obtained from L15 with the value of 14 and 31.83, respectively (Table 6). This  
274 line was also good for grain yield 2<sup>nd</sup> in grain yielding ability highlighting the existence of an  
275 association of these traits with GY (Table 6).

276 ASI ranged from 1.25 to 6.75 with a mean value of 3.4 days. The highest yielding line (L8)  
277 performed consistently across locations and had a reasonable ASI (2.5 days) (Table 4). Low

278 ASI was shown by L1 (1.75), L13 (1.25), and T1 (1.75 days). L1 is among the top 10 higher-  
279 yielding lines and T1 also recorded a high GY (Table 4). Lower ASI is a preferred trait to  
280 synchronize to males and females for better fertilization.

281 L8 had the highest BIOM of 10.59 t ha<sup>-1</sup> and was followed by FS67 (10.55 t ha<sup>-1</sup>). The  
282 lowest BIOM (4.84 t ha<sup>-1</sup>) was obtained from L11 compared with an overall mean of 6.90 t  
283 ha<sup>-1</sup>. The mean of BIOM obtained in this study was relatively smaller than the mean  
284 performance of QPM lines tested by Berhanu (2009) and Beyene (2016). They reported 8.37  
285 t ha<sup>-1</sup> and 10.8 t ha<sup>-1</sup>, respectively. Out of the 21 new QPM lines tested, 52.38 % had higher  
286 BIOM than the mean of the QPM checks indicating that the new coming lines had higher  
287 performance in converting inputs into different sinks which finally contribute to the total  
288 biomass (Table 4). The mean BIOM of new QPM lines (6.73 t ha<sup>-1</sup>) was almost equal to the  
289 mean of the QPM line checks (6.87 t ha<sup>-1</sup>) (Table 4). The line which was superior for BIOM  
290 (L8) also had a high GY in the combined analysis (Table 4).

291 Among new QPM lines, L15 attained the maximum PH (156.25 cm) while L13 was the  
292 shortest (95.75 cm) (Table 4). EH mean value ranged from 36.25 to 89.50 cm with a mean  
293 value of 62.06 cm. The mean of QPM new lines and the mean of QPM line checks were  
294 almost equal in value for both PH and EH (Table 4).

295 For LFBE, the lowest (6.08) was recorded by L13, and the highest (8.83) was obtained from  
296 L8 with an overall mean value of 7.34 for LFBE (Table 4). LL ranged from 53.83 cm (FS67)  
297 to 90.75 cm (L5) with an overall mean value of 71.95 cm. For LFAR the mean value of lines  
298 ranged from 408.94 cm<sup>2</sup> (FS67) to 654.4 cm<sup>2</sup> (L5) with an overall mean value of 538.62  
299 cm<sup>2</sup>. These lowest and highest LFAR scored QPM line and CM line check also had relatively  
300 high GY. The L8 which had the highest GY at both locations and in the combined analysis  
301 ((Tables 4, 5, and 6)) showed relatively high LFAR (530.68 cm<sup>2</sup>) in the combined analysis  
302 (Table 4).

303

304

305

307 Table 4. Mean of each line for traits combined over two locations Ambo and Arsi-Negele in  
 308 2017.

Code	GY	ASI	PH	EH	EPP	EL	ED	BIOM	LL	LFAR	LFBE
L1	2.88	1.75	111.75	52.75	1.02	10.75	4.02	7.15	69.92	595.95	7.25
L2	2.52	2.75	104.00	41.75	1.04	9.83	3.74	5.41	68.92	554.43	6.83
L3	2.58	2.75	144.50	73.25	1.85	11.08	2.91	7.81	79.25	513.28	6.50
L4	2.74	3.00	106.25	47.25	1.13	10.00	3.91	5.81	70.08	579.82	6.75
L5	3.58	2.75	131.75	59.50	1.11	11.46	3.89	7.54	90.75	654.40	7.75
L6	3.62	4.25	137.50	74.00	1.23	11.92	3.51	9.27	76.00	527.73	7.25
L7	3.03	4.50	131.25	62.75	1.29	11.50	3.55	6.17	76.58	500.95	7.08
L8	5.00	2.50	151.75	89.50	1.63	12.58	3.63	10.59	75.83	530.68	8.83
L9	2.87	3.25	143.75	75.25	1.24	13.08	3.43	7.23	72.17	490.43	7.92
L10	2.76	3.50	131.25	64.50	1.25	14.50	3.66	5.97	72.33	514.23	6.83
L11	2.44	4.50	130.25	59.25	1.17	11.08	3.87	4.84	70.25	461.59	6.67
L12	3.19	2.75	152.00	69.75	0.94	10.17	4.40	8.60	82.67	651.02	7.42
L13	2.33	1.25	95.75	36.25	1.09	9.17	3.72	5.01	68.92	569.13	6.08
L14	2.46	6.75	143.00	68.50	0.91	9.33	4.41	6.19	80.58	598.61	7.83
L15	3.86	2.75	156.25	80.25	1.08	12.25	4.19	7.82	73.00	621.16	8.33
L16	3.46	4.50	120.25	50.50	1.08	11.42	4.21	6.14	64.83	478.71	6.50
L17	2.53	4.25	123.25	53.50	1.03	10.58	4.25	5.21	65.42	485.98	7.50
L18	2.62	3.00	135.50	68.00	1.29	12.08	3.76	6.44	56.71	434.15	8.33
L19	2.10	4.00	138.75	50.25	1.27	13.17	3.51	6.58	73.17	540.36	6.67
L20	2.24	5.25	132.25	63.00	1.01	10.13	4.11	5.35	75.50	580.73	7.67
L21	2.91	4.25	118.50	59.75	1.09	11.00	3.83	6.25	66.25	494.01	8.00
T1	3.99	1.75	130.75	60.00	1.58	11.17	3.82	8.34	75.42	642.69	7.58
T2	2.47	3.75	119.50	49.25	1.00	11.38	3.62	5.40	68.42	497.84	6.50
FS67	4.66	2.00	145.25	80.75	1.31	13.17	3.64	10.55	53.83	408.94	8.00
Grand Mean	3.04	3.41	130.63	62.06	1.19	11.37	3.81	6.90	71.95	538.62	7.34
CV (%)	17.28	41.51	5.77	11.14	15.49	10.32	5.21	27.42	6.15	10.64	7.69
F-Test	***	**	***	***	***	***	***	**	**	***	***
LSD	0.75	2.01	10.73	9.84	0.26	1.67	0.28	2.69	6.30	81.56	0.80
Minimum (QPM line)	2.10	1.25	95.75	36.25	0.91	9.17	2.91	4.84	56.71	434.15	6.08
Maximum (QPM line)	5.00	6.75	156.25	89.50	1.85	14.50	4.41	10.59	90.75	654.40	8.83
Mean (QPM lines)	2.94	3.54	130.45	61.88	1.18	11.29	3.83	6.73	72.82	541.78	7.33
Mean (QPM checks)	3.23	2.75	125.13	54.63	1.29	11.27	3.72	6.87	71.92	570.27	7.04

309

310

311 **Table 5.** The mean of each line for yield, yield-related parameters, phenological, disease, and  
 312 morphological traits evaluated and significant only at Ambo, 2017.

Code	GY	DT	DS	MOD	TLB	EAS	TSW	LANG	LW	LFPP	LFAE
L1	2.65	113.00	114.00	1.00	1.00	3.00	270.84	43.33	11.87	13.17	6.33
L2	2.01	111.50	113.50	2.00	1.00	2.75	248.92	31.17	11.58	13.17	6.17
L3	2.15	108.00	110.00	1.00	1.25	4.00	181.49	27.50	9.25	14.33	7.67
L4	2.30	112.00	114.00	1.50	1.50	3.25	210.78	37.50	12.08	12.83	6.00
L5	3.39	114.50	120.00	1.50	1.00	2.75	303.93	30.00	9.83	15.33	6.83
L6	2.92	107.50	111.00	1.00	1.75	3.25	202.88	43.33	10.00	13.33	5.83
L7	2.59	103.50	107.00	1.00	1.00	3.75	139.23	36.67	9.92	13.00	5.67
L8	4.38	107.00	109.00	1.00	1.50	2.25	175.75	45.00	10.08	14.83	6.00
L9	2.71	108.00	111.00	1.00	1.25	3.50	199.58	38.33	9.75	15.00	6.50
L10	2.74	104.00	106.50	2.50	1.00	3.75	160.19	38.33	10.50	13.00	6.00
L11	2.15	104.00	107.50	2.50	1.00	3.50	166.77	40.00	10.33	14.17	6.67
L12	3.18	117.00	119.00	2.00	1.00	2.50	295.69	29.17	11.47	15.50	7.67
L13	2.24	110.00	109.00	1.50	1.00	2.75	216.45	33.33	12.00	13.33	6.83
L14	2.15	111.00	117.00	2.50	1.00	3.00	248.93	28.33	10.92	15.00	6.67
L15	2.20	110.00	112.00	1.00	1.00	3.00	314.17	34.17	12.58	14.67	6.33
L16	3.13	101.00	106.00	2.00	1.75	2.75	220.91	32.50	11.00	12.50	6.33
L17	2.24	112.00	116.00	2.50	1.25	3.25	318.55	30.83	10.67	14.50	6.67
L18	2.52	109.00	111.00	1.00	1.00	2.75	286.89	40.00	12.08	15.67	6.83
L19	1.37	106.00	108.50	1.00	1.75	3.75	295.90	51.67	10.92	14.50	7.67
L20	2.15	110.50	116.00	1.00	1.50	3.00	290.90	40.00	11.83	15.00	6.83
L21	2.62	108.50	112.00	1.00	1.00	3.25	243.50	34.17	10.42	14.00	6.33
T1	3.39	105.00	105.50	2.00	1.00	2.50	164.63	36.67	12.33	15.83	7.33
T2	1.68	107.50	109.50	1.50	1.00	3.50	172.75	30.83	11.08	13.17	6.50
FS67	4.74	99.00	101.00	1.00	1.00	2.75	364.16	47.50	11.67	14.00	5.50
Grand Mean	2.65	108.31	111.08	1.50	1.19	3.10	237.24	36.68	11.01	14.16	6.55
CV (%)	21.45	1.60	2.32	33.09	23.23	13.29	15.91	10.09	4.96	4.08	4.80
F-Test	**	***	***	**	*	*	***	***	***	***	***
LSD	1.18	3.58	5.34	1.03	0.57	0.85	78.10	7.66	1.13	1.19	0.65
Minimum (QPM line)	1.37	101.00	106.00	1.00	1.00	2.25	139.23	27.50	9.25	12.50	5.67
Maximum (QPM line)	4.38	117.00	120.00	2.50	1.75	4.00	318.55	51.67	12.58	15.67	7.67
Mean (QPM lines)	2.56	108.95	111.90	1.50	1.21	3.13	237.73	36.44	10.91	14.13	6.56
Mean (QPM checks)	2.53	106.25	107.50	1.75	1.00	3.00	168.69	33.75	11.71	14.50	6.92

313

314

315

316

317 **Table 6.** The mean of each line for yield, yield-related parameters, phenological, agronomic, disease, and morphological traits were evaluated  
 318 and significant by Arsi-Negele, 2017.

Code	GY	DT	DS	GLS	CLR	EAS	PAS	NKR	KPR	TSW	LANG	LW	LFPP	LFAE
L1	3.11	96.50	99.00	1.25	2.50	3.25	2.50	13.00	17.83	310.93	40.83	10.83	14.17	6.50
L2	3.03	95.00	98.50	1.25	2.50	2.50	3.00	13.00	20.83	270.09	35.83	9.58	13.00	6.33
L3	3.01	95.00	98.50	1.25	2.25	3.00	3.00	10.00	20.17	205.75	28.33	7.83	13.50	7.17
L4	3.18	94.50	98.50	1.50	2.00	3.25	2.75	13.00	16.67	324.45	38.33	9.83	13.50	6.83
L5	3.76	99.00	99.00	1.50	2.00	2.75	3.00	14.00	28.83	208.64	27.50	9.33	13.67	6.67
L6	4.33	92.00	97.00	1.75	3.00	2.25	2.75	13.00	22.00	261.00	48.33	8.33	12.83	5.83
L7	3.46	93.00	98.50	1.25	3.50	3.25	3.00	14.00	25.50	187.91	39.17	7.17	12.50	5.67
L8	5.62	95.00	98.00	1.25	3.25	2.00	3.00	12.00	26.67	299.23	53.33	8.50	15.00	6.17
L9	3.03	95.50	99.00	1.00	4.50	3.00	3.50	11.00	25.17	232.62	45.83	8.17	13.83	6.50
L10	2.77	94.50	99.00	1.00	3.50	4.25	3.25	14.00	22.50	256.25	45.83	8.33	12.83	6.17
L11	2.73	91.00	96.50	1.00	3.25	2.50	3.50	14.00	22.00	243.11	61.67	6.58	12.00	6.17
L12	3.19	95.50	99.00	1.50	3.00	1.50	2.25	14.00	25.00	275.81	32.50	9.33	14.67	7.67
L13	2.42	96.00	99.50	1.25	2.50	2.75	3.00	12.00	18.00	287.01	40.83	9.63	12.50	6.83
L14	2.77	95.00	102.50	1.50	1.75	3.25	2.50	13.00	18.50	225.92	30.00	8.83	14.17	6.83
L15	5.52	92.00	95.50	1.75	2.75	1.25	1.75	14.00	31.83	293.77	40.00	10.00	15.33	7.00
L16	3.80	86.50	90.50	1.50	2.00	1.75	2.00	14.00	23.67	316.59	45.83	8.33	13.33	6.50
L17	2.83	92.50	97.00	1.25	3.00	2.50	2.25	11.00	20.67	335.00	39.17	9.00	13.17	6.00
L18	2.72	92.00	96.00	2.00	2.00	3.25	2.50	10.00	20.67	353.50	47.50	7.33	14.00	6.17
L19	2.83	93.50	99.00	1.50	2.00	2.75	2.00	10.00	24.83	319.02	43.33	8.50	13.67	7.17
L20	2.32	94.00	99.00	1.50	3.25	2.50	2.75	12.00	20.33	309.45	40.00	8.17	13.33	6.17
L21	3.21	93.50	98.50	1.50	2.50	2.50	2.00	11.00	23.83	337.00	43.33	9.33	14.33	6.00
T1	4.59	95.50	98.50	1.50	3.25	2.50	1.75	13.00	26.67	266.83	30.83	10.12	13.50	6.83
T2	3.26	91.50	97.00	1.25	2.75	3.50	2.50	13.00	23.00	276.51	28.33	8.00	13.50	7.17
FS67	4.57	85.50	87.50	1.25	1.75	3.00	2.50	9.00	28.33	410.99	71.67	8.00	12.67	5.17

319

320

321 **Table 6. Continued**

Code	GY	DT	DS	GLS	CLR	EAS	PAS	NKR	KPR	TSW	LANG	LW	LFPP	LFAE
Grand Mean	3.42	93.50	97.54	1.39	2.70	2.71	2.63	12.38	23.06	283.64	41.60	8.71	13.54	6.48
CV %)	13.93	2.46	2.12	16.79	18.52	16.89	16.81	9.22	12.24	8.71	9.64	9.14	5.50	4.29
T-Test	***	**	***	*	**	**	*	**	**	***	***	**	*	***
LSD	0.99	4.76	4.27	0.48	1.03	0.95	0.91	2.36	5.84	51.11	8.30	1.65	1.54	0.57
Minimum (QPM line)	2.32	86.50	90.50	1.00	1.75	1.25	1.75	10.00	16.67	187.91	27.50	6.58	12.00	5.67
Maximum (QPM line)	5.62	99.00	102.50	2.00	4.50	4.25	3.50	14.00	31.83	353.50	61.67	10.83	15.33	7.67
Mean (QPM lines)	3.32	93.88	98.00	1.39	2.71	2.67	2.68	12.48	22.64	278.72	41.31	8.71	13.59	6.49
Mean (QPM checks)	3.93	93.50	97.75	1.38	3.00	3.00	2.13	13.00	24.83	271.67	29.58	9.06	13.50	7.00

322

323

324

325

326

327

328

329

330

### 331 **3.4. Comparison of mean of lines with mean of line checks**

#### 332 **A. Across locations**

333 Of the total of 28 traits, 13 traits: GY, DT, DS, ASI, MOD, CLR, TLB, EAS, TSW, LANG,  
334 LW, LFPP, and LFAE had significant. Due to this, the contrast analysis was conducted for  
335 the individual location.

#### 336 **3.4.1. Comparison between the 21 QPM lines and the two QPM checks across locations**

337

338 In the combined ANOVA, MS of QPM line vs QPM check was significant for DS, MD, EH,  
339 TSW, LANG, and LFAE but this orthogonal analysis showed a non-significant difference for  
340 GY, DT, ASI, PH, MOD, GLS, CLR, TLB, EAS, PAS, EPP, EL, ED, NKR, KPR, BIOM,  
341 HI, LL, LW, LFAR, LFPP, and LFBE. In contrast, Pavan et al. (2011) reported significant  
342 MS for DT, PH, EH, KPR, NKR, and GY but his report is in line with the finding in this  
343 study for TSW, DS, and EL. The overall observation of the combined analysis of this study  
344 shows that QPM lines vs QPM check had a positive magnitude (above zero) for DT, DS,  
345 ASI, MD, PH, EH, GLS, TLB, PAS, EL, ED, TSW, LANG, LL, and LFBE. Whereas GY,  
346 MOD, EAS, EPP, NKR, KPR, BIOM, HI, LW, LFAR, LFPP, and LFAE had a negative  
347 magnitude (below zero).

348 The orthogonal mean difference between QPM lines had lower GY performance by 0.29 t/ha  
349 compared with QPM checks. Similarly, Dufera et al. (2018) reported promising QPM lines.  
350 This highlights the existence of the possibility to generate promising hybrids development.  
351 The mean difference indicates that the mean of QPM lines had less value by 0.14 t ha  
352 compared to the mean of QPM checks for BIOM. For EL, the mean of QPM lines mean had  
353 almost equal to the mean of QPM checks in magnitude (0.02 cm). QPM lines were taller  
354 than QPM checks by 5.33 and 7.26 cm for PH and EH, respectively. QPM lines had a higher  
355 LL value than the mean of QPM checks. However, for LFAR, the new QPM lines are lower  
356 than the mean of QPM checks by 28.49 cm<sup>2</sup> (Table 7).

#### 357 **3.4.2. Comparison between the 21 QPM lines and the QPM check line FS67 across** 358 **locations**

359

360 The MS of the QPM line vs FS67 showed significant MS for GY, DT, DS, ASI, PH, EH,  
361 CLR, TLB, EL, NKR, TSW, BIOM, LANG, LL, LFAR, and LFAE (P<0.001, P<0.01 or  
362 P<0.05) whereas the MS of MD, MOD, GLS, EAS, PAS, EPP, KPR, ED, HI, LW, LFPP, and  
363 LFBE was non-significant (data not shown). For QPM lines vs FS67: DT, DS, ASI, GLS,  
364 CLR, TLB, EAS, NKR, ED, HI, LL, LFAR, LFPP, and LFAE had positive values (above

365 zero) but GY, MD, PH, EH, MOD, PAS, EPP, EL, KPR, TSW, BIOM, LANG, LW and LFBE  
 366 inferior performance than FS67(data not shown).

367 The mean of QPM lines had a lower performance by 1.72 t ha<sup>-1</sup> and 3.82 t ha<sup>-1</sup> compared  
 368 with CM FS67 for GY and BIOM, respectively. For EL, the QPM lines mean had shorter by  
 369 1.88 cm compared with the value of FS67. Dufera et al. (2018) also reported higher  
 370 performance by testers for yield-related traits compared with new QPM lines except for  
 371 TSW.QPM lines were shorter than FS67 by 14.80 and 18.87 cm for PH and EH,  
 372 respectively. For LL and LFAR, the mean of QPM lines had a higher value than FS67 by  
 373 18.98 cm 132.82 cm<sup>2</sup>, respectively. QPM Lines mean had wider days of ASI interval  
 374 compared with FS67 with the value of 1.54. Based on this, it can be concluded that QPM  
 375 lines had a problem with male and female flower synchronization (Table 7) but there are  
 376 QPM lines with an acceptable range.

377 Table 7. MS of contrast between lines, testers, and CM line check (FS67) and estimated value of mean  
 378 difference for grain yield and other traits in combined analysis evaluated at Ambo and Arsi-Negele  
 379 Agricultural Research Centers, 2017.

		MS of Contrast							
Source of Variation	DF	GY	ASI	PH	EH	EL	BIOM	LL	LFAR
QPM Lines vs QPM checks	1	0.62	4.51	207.3	384.57**	0.001	0.14	5.9	5927.24
QPM Lines vs FS67	1	11.2***	9.00*	836.07**	1359.4***	13.45*	55.69 **	1375.77***	67378.25 **
		Estimate of Mean Difference							
QPM Lines vs QPM checks	1	-0.29	0.79	5.33	7.26**	0.02	-0.14	0.9	-28.49
QPM Lines vs FS67	1	-1.72	1.54*	-14.80**	-18.87***	-1.88*	-3.82**	18.98***	132.84**

380

## 381 B. Individual Location

### 382 3.4.3. Comparison between the 21 QPM lines and the two QPM checks at each location

383

384 The MS for QPM line vs QPM checks and QPM lines vs FS67 and the mean difference of  
 385 the contrast is presented in (Table 8) for traits considered at an individual location. At Ambo,  
 386 the mean of QPM lines had a higher value by 0.03 t ha<sup>-1</sup> than the mean of QPM checks for  
 387 GY however at Arsi-Negele mean of QPM checks showed higher than the mean of QPM  
 388 lines by 0.61 t ha<sup>-1</sup>. But while considering the performance of each line with the checks,  
 389 some lines had higher performance (Tables 5 and 6). For TSW the mean of QPM lines  
 390 exceeded the mean of QPM checks by 69.04 and 7.05 g at Ambo and Arsi-Negele,  
 391 respectively (Table 8). This indicates there are new QPM lines are better than the QPM  
 392 checks in the breeding program. So that the breeding program can have the chance to get  
 393 better QPM lines after further evaluation. At Arsi-Negele, the mean of QPM lines was  
 394 delayed by 2.87 days for maturity compared with the mean of QPM checks (Table 8). For  
 395 DT and DS, QPM lines were late by 2.70 and 4.4 days compared with QPM checks at Ambo.

396 At Arsi-Negele, the smaller value of QPM lines compared with QPM checks by 0.29 for  
397 CLR indicates that the QPM lines are more tolerant to this disease. The poorness of QPM  
398 checks for PAS is manifested by the higher value of 0.55 compared with the mean value of  
399 QPM lines (Table 8). The mean of QPM lines had wider LANG ((by  $11.73^\circ$ ) and thinner LW  
400 (by 0.80 cm) than the mean of QPM checks at Arsi-Negele and Ambo, respectively.  
401 Regarding LFAE, QPM lines also showed higher performance than QPM checks by 0.51  
402 leaves at Arsi-Negele (Table 8).

#### 403 **3.4.4. Comparison between the 21 QPM lines and the CM check line FS67 at each** 404 **location**

406 At both locations (Ambo and Arsi-Negele), the performance of FS67 was higher by 2.17 t  
407  $\text{ha}^{-1}$  and 1.26 t  $\text{ha}^{-1}$  compared with the mean of QPM lines, respectively (Table 8). But while  
408 considering each line pairwise there are QPM lines (L8 and L15) that had higher GY than  
409 FS67 (Table 6).

410 Regarding NKR, the mean of QPM lines gave higher performance than FS67 by 2.31 and  
411 3.48 kernel rows at Ambo and Arsi-Negele, respectively. Regarding TSW, the mean of QPM  
412 lines had a lower value of 126.43 and 132.27 g compared with the FS67 at Ambo and Ars-  
413 Negele, respectively. The mean of QPM lines was lower by 126.43 g compared with FS67 at  
414 Ambo for TSW (Table 8). In agreement with this finding, a higher TSW value by QPM lines  
415 compared with the testers was obtained in the study of Dufera et al. (2018). QPM lines  
416 mature earlier by 1.88 days compared with FS67 at Arsi-Negele (Table 8). For DT and DS,  
417 QPM lines were late for flowering by 9.95 and 10.90n days compared with FS67 at Ambo,  
418 respectively. Similarly, at Arsi-Negele, the mean difference indicates that the mean of QPM  
419 lines was late for DT by 8.38 and DS by 10.50 compared with the FS67 CM check (Table  
420 8).In line with this, Dufera et al.(2018) reported the lateness of the lines for DT and DS  
421 compared with hybrid testers.

422 The positive orthogonal mean difference for CLR (0.96) between QPM lines vs FS67  
423 confirms that QPM lines were more attacked by this disease than FS67. Based on the value  
424 the higher value (0.18) for PAS cored by FS67, the new QPM lines were good for this trait  
425 (Table 8). Regarding LANG, the mean of QPM lines had a narrow-leaf angle than FS67 by  
426  $11.06^\circ$  and  $30.36^\circ$  at Ambo and Arsi-Negele, respectively. For LW the mean of QPM lines  
427 was thinner by 0.76 cm than FS67 at Ambo which is more advantageous for crop production  
428 in terms of improving photosynthesis. Similarly, Li et al (2015) suggested and confirmed the  
429 importance of erect leaf angle and optimum leaf orientation value to allow for more efficient  
430 light capture during photosynthesis and better wind circulation under dense planting  
431 conditions. Other linked traits like leaf length, leaf width, and leaf area have a role in

432 photosynthesis. For LFAE, QPM lines had greater by 1.06 and 1.33 than FS67 at Ambo and  
433 Arsi-Negele, respectively (Table 8).

434

435 **Table 8.** MS contrast between lines, testers, and CM line check (FS67) and the estimated value of mean difference for traits at Ambo and Arsi-  
 436 Negele which are not included in the combined analysis, 2017.

MS of Contrast											
Ambo											
Source of Variation	DF	GY	DT	DS	NKR	TSW	LANG	LW	LFAE		
QPM Line vs QPM check	1	0.002	26.671**	70.86***	0.13	17406.84***	26.51	2.34*	0.45		
QPM Lines vs FS67	1	9.02***	189.10***	227.017***	10.18*	30516.9***	233.34***	1.10***	2.15**		
Estimate of Mean Difference											
QPM Line vs QPM check	1	0.03	2.70**	4.40***	-0.19	69.04***	2.69	-0.80	-0.35		
QPM Line vs FS67	1	-2.17***	9.95***	10.90***	2.31*	-126.43***	-11.06***	-0.76	1.06		
MS of Contrast											
Arsi-Negele											
Source of Variation	DF	GY	DT	DS	MD	CLR	PAS	NKR	TSW	LANG	LFAE
QPM Line vs QPM check	1	1.36	0.53	0.23	30.06***	0.30	1.12*	1.00	181.42	502.19**	0.94*
QPM Line vs FS67	1	3.02*	134.10***	210.48***	6.75	1.78*	0.06	23.10**	33399.98***	1759.33***	3.35***
Estimate of Mean Difference											
QPM Line vs QPM check	1	-0.61	0.38	0.25	2.87***	-0.29	0.55*	-0.52	7.05	11.73**	-0.51*
QPM Line vs FS67	1	-1.26*	8.38***	10.50***	-1.88	0.96*	0.18	3.48**	-132.27***	-30.36**	1.33***

437

438

439

440

## 441 **Conclusion**

442 The analysis variance for grain yield shows a significant difference between 24 tested lines in each  
443 location and the two locations combined. The significant difference in G\*L for grain yield indicates  
444 that the performance of the genotypes was not consistent across the two locations. Based on the mean  
445 performance L8 and L15 had higher GY compared with both the QPM checks and CM check (FS67) at  
446 Arsi-Negele. L15 showed good tolerance to GLS, and CLR and it had also good EAS recorded  
447 whereas L8 even if it showed good performance for GY, EAS, and GLS, was affected by CLR. Arsi-  
448 Negele is a hot spot for CLR, at this location, some lines showed relative tolerance to CLR which is  
449 manifested by low scored value. Thus, L14 had the lowest score value compared with the other tested  
450 lines for CLR. In addition, L4, L5, L16, L18, and L19 had relatively lower score values for this trait.  
451 The orthogonal analysis for DT and DS highlighted that the new lines were relatively late for flowering  
452 compared with both QPM and CM checks. In general, the study confirmed the existence of promising  
453 new QPM parental lines that can be used in the breeding program for further improvement.

## 454 **Acknowledgments**

455 The financial support of CIMMYT and the Ethiopian Institute of Agricultural Research  
456 (EIAR) for funding this study is greatly appreciated. The staff of the Maize breeding section  
457 Ambo and Kulumsa Research Centers are greatly acknowledged for their support in field trial  
458 management and data collection, especially for the former for their facilitation and  
459 contribution to the hybrid formation and seed preparation. Thanks to all EIAR- Hawassa  
460 maize research staff for their support in field trial management and data collection.

## 461 **Authors' contributions**

462 This work was carried out in collaboration among all authors. Author GMM collected the data,  
463 performed the analysis, and developed the manuscript. Author SAS, DNW, and BTG reviewed and  
464 made editorial comments on the draft manuscript.

## 465 **Authors' information**

466 Author GMM is a plant breeder with more than 10 years of experience who has been involved in maize  
467 germplasm development with the national maize breeding program.

468 Author DNW is Research Officer currently working for CIMMYT -Ethiopia who was responsible for  
469 planning and executing highland maize breeding activities for East and Central Africa.

470 Author BTG is a researcher at the Ethiopian Institute of Agricultural Research with more than 10-year  
471 experience in cereal breeding including maize. Currently, he is a Ph.D. student at the University of  
472 Cape Town.

## 473 **Funding**

474 Funding – Funding was provided by CIMMYT and the Ethiopian Institute of Agriculture Research  
475 (EIAR).

#### 476 **Availability of data and materials**

477 Availability of data and materials – data can be requested from the author.

#### 478 **Declarations**

#### 479 **Ethics approval**

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

#### 483 **Competing interests**

484 The authors declare that they have no competing interests.

#### 485 **Reference**

- 486 Abebe M, Melake-Berhan A, Engelbrecht ACI, Adepoju A. Grouping of tropical mid-altitude maize inbred lines  
487 based on yield data and molecular markers. *Theoretical and Applied Genetics*, 2004, 108: 1582-1590.
- 488 Adefris T, Dagne W, Abraham T, Birhanu T, Kassahun B, Friesen DK, Prasanna BM. Quality Protein Maize  
489 (QPM): A Guide to the Technology and Its Promotion in Ethiopia. CIMMYT, 2015, Addis Ababa,  
490 Ethiopia.
- 491 Atlin GN, N. Palacios N, Badu R, Das B, Twumasi-Afriye S, Friesen DK, De Groot A, Vivek B, Pixley KV.  
492 Quality Protein Maize: Progress and prospects. *Plant Breeding Reviews*, 2011. Volume 34.
- 493 Badu-Apraku B, Oyekunle M, Akinwale RO, Fontem LA. Combining Ability of Early-maturing White Maize  
494 Inbreds under Stress and Nonstress Environments. *Agronomy Journal*. 2011, DOI:  
495 10.2134/agronj2010.0345.
- 496 Bain L, Awah P, Geraldine N, Kindong N, Sigal, Y, Bernard N, Tanjeko A. Malnutrition in Sub-Saharan Africa:  
497 burden, causes, and prospects. *Pan African Medical Journal*, 2013, 15:120.
- 498 Bekele S, Prasanna BM, Hellin J, Bänziger M. Crops that feed the world 6. Past successes and future challenges  
499 to the role played by maize in global food security. *Food Security*, 2011, 3:307-327.
- 500 Berhanu T. Heterosis and combining ability for yield, yield-related parameters, and stover quality traits for  
501 food-feed in maize (*Zea Mays L.*) adapted to the mid-altitude agroecology of Ethiopia. MSc.Thesis.  
502 Haramaya University, Haramaya, Ethiopia, 2009, 199 p.
- 503 Beyene A. Heterosis and combining ability of mid-altitude quality protein maize (*Zea Mays L.*) inbred lines at  
504 Bako, Western Ethiopia. MSc.Thesis. Haramaya University, Haramaya, 2016, 158 p.
- 505 Bolaños J and Edmeades GO. The importance of the anthesis-silking interval in breeding for drought tolerance  
506 in tropical maize. *Field Crops Research*, 1996, 48: 65-80.
- 507 Bressani R, Elias LG, Gomez-Brenes RA. Protein quality of *opaque-2* maize. Evaluation in rats. *Journal of*  
508 *Nutrition*, 1969, 97: 173-180.
- 509 Bressani, R. Nutritional value of high-Lysine maize in humans. In: Mertz E.T. (Eds.), 1992.
- 510 Bressani, R. *Opaque-2* in human nutrition and utilization. In: B.A. Larkins and E.T. 1995.
- 511 CIMMYT (International Maize and Wheat Improvement Center). CIMMYT's Work for Maize Systems and  
512 Farmers in Sub-Saharan Africa: Center Commissioned External Review (CCER), CIMMYT, Mexico,  
513 D.F., Mexico. 2004b.
- 514 CSA (Central Statistical Agency). Agricultural sample survey volume I: Report on area and production of  
515 major crops for private peasant holdings (Meher season). Statistical bulletin. CSA Addis Ababa, Ethiopia,  
516 2018.
- 517 Dagne WG. Genotypic variability and combining ability of quality protein maize inbred lines under stress and  
518 optimal conditions. Ph.D. Thesis. University of the Free State, South Africa. 2008, 221-243 p.

519 Demissew A. Genetic diversity and combining ability of selected quality protein maize (QPM) inbred lines  
520 adapted to the highland agroecology of Ethiopia. 2014.

521 Dufera T, Girum A, and Bulti T. Perse Performance and Heterosis of Quality Protein Maize (*Zea mays* L.) Inbred  
522 Lines Adapted to Mid-Altitude SubHumid Agro-Ecology of Ethiopia. International Journal of Research  
523 Studies in Science, Engineering, and Technology, 2018. ISSN: 2349-476X, Vol.5, Issue 8, 14-26.

524 Etagenehu G/M and Amare F. Effect of Different Time and Frequency of Weeding on Castor (*Ricinus*  
525 *communis* L.) in Central Rift Valley of Ethiopia. International Journal of Research in Agriculture and  
526 Forestry, 2016, Vol 3, Issue 7, 31-37 pp.

527 FAO (Food and Agriculture Organization of the United Nations) Statistical. FAO Statistical Pocketbook world  
528 food and agriculture. 2015, <http://Faostat.fao.org>.

529 FAO (Food and Agriculture Organization of the United Nations). FAO Agricultural Outlook 2017-2026, OECD  
530 Publishing, Paris. 2017.

531 Gomez KA and Gomez AA. Statistical Procedures for Agricultural Research, 2nd edit. John Wiley and Sons,  
532 New York. 1984.

533 Gudeta N, Dagne W, and Habtamu Z. Heterosis and combining ability of highland quality protein maize inbred  
534 lines. Maydica, 2015, 60-2015.

535 Gupta SC, Asnani UL, Khare BP. Effect of the *opaque-2* gene in maize (*Zea mays* L.) on the extent of  
536 infestation by *Sitophilus oryzae* L. Journal of Stored Products Research, 1970, 6: 191-194.

537 Krivanek AF, De Groote A, Gunaratna NS, Diallo AO, Friesen DK. Breeding and disseminating quality protein  
538 maize (QPM) for Africa. African Journal of Biotechnology, 2007, 6: 312-324.

539 Li C, Li Y, Shi Y, Song Y, Zhang D, Buckler ES, Zhang Z, Tianyu Wang T, Li Y. Genetic Control of the Leaf  
540 Angle and Leaf Orientation Value as Revealed by ultra-high-density Maps in Three Connected Maize  
541 Populations. PLoS ONE, 2015, 10(3): e0121624. doi:10.1371/journal.pone.0121624.

542 Lobell DB, Schlenker W, Costa-Roberts J. Climate Trends and Global Crop Production Since 1980. Science  
543 333, 2011, 616. (2011); DOI: 10.1126/science.1204531.

544 Lopez-Pereira MA. The economics of quality protein maize as an animal feed. Case studies of Brazil and El  
545 Salvador. CIMMYT Economics working paper 92-06. 1992, CIMMYT, Mexico, D.F., Mexico.

546 Mbuya K, Nkongolo KK, Kalonji-Mbuyi A. Nutritional Analysis of Quality Protein Maize Varieties selected  
547 for agronomic characteristics in a breeding program. Int. J. Plant Breed. Genet, 2011, 5 (4), 317-327.

548 Mertz ET, Bates LS, Nelson OE. Mutant gene that changes protein composition and increases lysine content of  
549 maize endosperm. Science, 1964, 145: 279 -280.

550 Mertz ET. Discovery of high-lysine, high Tryptophan cereals. In: E.T. Mertz (Eds.). Quality Protein Maize.  
551 American Association of Cereal Chemists, St. Poul, Minnesota, USA. 1992, 1-8 pp.

552 Morley EJ, Protein-Energy Undernutrition (PEU). nutritional disorders/undernutrition/protein-energy-  
553 undernutrition-peu. 2016, <http://www.merckmanuals.com/professional>.

554 Mosisa W, Bänziger M, Erley GS, Friesen DK, Diallo AO, Horst WJ. Nitrogen Uptake and Utilization in  
555 Contrasting Nitrogen Efficient Tropical Maize Hybrids. Crop Sci, 2007, 47:519-528, DOI:  
556 10.2135/cropsci2005.05.0070.

557 Norman MJT, Pearson CJ, Searle PGE. The ecology of Tropical food crops, 2<sup>nd</sup> ed. Cambridge University  
558 Press, New York. NY USA. 1995, pp 127-140.

559 Osei SA, Okai DB, Ahenkora K, Dzah BD, Haag W, Twumasi-Afriyie S, Tua AK. Quality protein maize is the  
560 main source of energy and amino acids in the diets of starter pigs. Proceedings of Ghanaian Animal Science  
561 Symposium, 1994a, 22: 31-36.

562 Pavan R, Gangashetty P, Mallikarjuna NM. General and specific combining ability studies in single cross hybrids  
563 of maize (*Zea mays* L.). Current Biotica, 2011, 5(2): 196-208.

564 Rouf Shah T, Kamlesh P, Pradyuman K. Maize-Potential source of human nutrition and health: A review.  
565 SAS Institute, 2002. The SAS system for windows. Version 9.0. SAS Inst., Cary, NC. 2016.

566 Tekeba E. Review of quality protein maize as food and feed: In alleviating protein deficiency in developing  
567 country. American of food and nutrition, 2017, Vol.5, 99-105.

568 Weber VS, Melchinger AE, Magorokosho C, Makumbi D, Bänziger M, Atlin GN. The efficiency of Managed-  
569 Stress Screening of Elite Maize Hybrids under Drought and Low Nitrogen for Yield under Rainfed  
570 Conditions in Southern Africa. Crop Sci., 2012, 52:1011–1020, DOI: 10.2135/cropsci2011.09.0486.