1 Genetic diversity of tropical early-maturing maize inbreds and their performance
2 in hybrid combinations under drought and optimum growing conditions

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13 Abstract

14 Hybrid development is enhanced by the assessment and understanding of genetic
15 diversity and distance within inbreds. One hundred and fifty hybrids derived from 30
16 early-maturing inbreds plus six checks were evaluated at three locations in Nigeria for
17 2 years to assess their performance under drought and well-watered conditions. In
18 addition, twenty three microsatellite markers were employed to assess genetic
19 diversity of selected 42 inbreds. Significant differences were observed among inbreds
20 and hybrids for most traits under both research conditions. A total of 130 alleles were
21 detected ranging from two for *nc133* to nine for *phi299852* with an average of 5.7
22 alleles per locus. Polymorphic information content ranged from 0.17 for *phi308707* to
23 0.77 for *phi084* with an average of 0.54. Thirty-one unique alleles were detected in 21
24 inbreds. Microsatellite markers classified the inbred lines into five groups. Genetic
25 distance estimates among pairs of inbreds ranged from 0.42 (TZEI 26 vs TZEI 108) to

0.85 (TZEI 24 vs TZEI 4) with an average of 0.67. Correlation between microsatellite-based GD estimates of the parental lines and their F₁ hybrids were not significant for grain yield and other traits under drought and well-watered conditions. However, significant correlations existed between F₁ hybrid grain yield and heterosis under drought and well-watered conditions. TZEI 31 × TZEI 18 was identified as the highest-yielding and stable hybrid across environments and should be promoted for adoption by farmers in West and Central Africa.

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Keywords: Drought stress, Heterosis, Drought tolerance, Inbred lines, Simple sequence repeat markers.

36

Abbreviations: **ASI**, anthesis silking interval; **AMMI**, additive main effects and multiplicative interaction; **BPH**, Better parent heterosis; **DAP**, Days after planting; **EPP**, ear number per plant; **GD**, Genetic distance; **GEI**, genotype-by-environment interaction; **GGE**, genotype main effect plus genotype x environment interaction; **IITA**, International Institute of Tropical Agriculture; **LD**, Stay green characteristic; **MPH**, Midparent heterosis; **MRD**, Modified Roger's distance; **PC**, Principal component; **PIC**, Polymorphic information content; **SCA**, Specific combining ability; **SSR**, Simple sequence repeat; **WCA**, West and Central Africa;

45

1. Introduction

Maize (*Zea mays* L.) is a major staple crop for millions of people in West and Central Africa (WCA) and accounts for 0.15 of the total caloric intake of the rural and urban consumers. Its importance in the food basket in WCA has been increasing steadily over the last few decades (Fakorede et al, 2003). The availability of early-maturing maize has contributed significantly to the expansion of maize into new frontiers in the savanna agro-ecologies of WCA which have the greatest potential for

53 increased maize production because of the high solar radiation, and low incidence of
54 pests and diseases (Badu-Apraku et al, 2008). As a result, maize has been adopted
55 extensively in the zone leading to the replacement of the traditional crops, sorghum
56 and millet by maize (Fakorede et al, 2001). However, maize productivity in these
57 regions is greatly constrained by several biotic and abiotic factors, including drought,
58 low soil fertility, and the parasitic weed, *Striga hermontica* (Del.) Benth (Badu-
59 Apraku et al, 2003).

60 Drought is a major abiotic stress contributing to severe maize yield loss in the
61 lowland savanna belt of WCA (Fajemisin et al, 1985). Edmeades et al (1995) reported
62 15% annual maize yield loss from drought in the savannas of WCA and indicated that
63 localized losses may be much higher in the marginal areas with annual rainfall below
64 500 mm and on sandy soils. The risk of drought stress is severe particularly in the
65 Sudan savanna zone due to unreliable and uneven distribution of rainfall (Eckebil,
66 1991). Even in those lowlands with adequate precipitation for maize production,
67 periodic drought may occur at the most sensitive stages of the crop such as flowering
68 and grain filling. While drought will impact the growth and ultimate performance of a
69 crop at any stage, it is of most detriment at flowering and grain-filling resulting in
70 yield penalties of between 40 to 90% (Menkir and Akintude, 2001; Badu-Apraku et al,
71 2011a; Badu-Apraku and Oyekunle, 2012). Therefore, improved tolerance to drought
72 is an important breeding objective to stabilize maize production in the sub region.

73 Until recently, adoption of maize hybrids was insignificant in WCA due to
74 limited seed production and marketing by existent and emerging few seed companies.
75 However, during the last couple of years, seed companies have emerged in Ghana,
76 Mali, and Nigeria setting the stage for large scale hybrid production in these countries
77 (Badu-Apraku et al, 2011a). Hybrid development and promotion in maize is a

78 promising strategy to appreciably increase maize production and productivity in WCA.
79 Presently, within the portfolio of hybrids in WCA there are no commercially available
80 early maturing hybrids. An important requirement for a commercially successful
81 hybrid program is the availability of information on the heterotic patterns of inbred
82 lines in the program. Accurate assessment of inbred lines in hybrid combinations is
83 crucial to the success of a hybrid program and the selection of parents for the
84 development of synthetic varieties or introgression of favourable alleles into elite
85 source populations. Knowledge and understanding of the genetic diversity and
86 distance of early-maturing inbred lines would be very useful in planning crosses,
87 assigning inbred lines to specific heterotic groups and designing breeding strategies.

88 Understanding of heterotic grouping of inbred lines on the basis of phenotypic
89 values of genetic traits is highly desirable in maize breeding in designing guidelines
90 for use in developing heterotic populations and synthetic varieties (Hallauer and
91 Miranda, 1988; Badu-Apraku et al, 2006). Badu-Apraku and Lum (2007) used
92 multivariate analysis of morphological and agronomic traits to assess the genetic
93 diversity among early-maturing inbred lines. Menkir et al (2004) reported the
94 difficulty of classifying tropical inbred lines into distinct heterotic groups based only
95 on the results of combining ability studies. Molecular markers are a powerful tool for
96 defining heterotic groups and examining the relationships among inbred lines at the
97 DNA level (Smith et al, 1997; Senior et al, 1998; Melchinger, 1999) and may
98 complement existing approaches. Therefore, the combined use of molecular markers
99 that allow direct comparison of the similarity of inbreds at the DNA level with
100 testcross evaluation in the field should facilitate the separation of inbred lines into
101 well-defined heterotic groups (Menkir et al, 2004).

102 Several DNA marker technologies have been developed and are available to
103 study genetic diversity. Favorable characteristics sought in DNA based markers
104 include high levels of polymorphism, co-dominance, abundance and even distribution
105 in the genome. On the technical side, the ease, speed and reproducibility of the assays
106 are also very important characteristics (Weising et al, 1998). Of the markers available
107 to this study, microsatellite or simple sequence repeat (SSR) fulfills most of the
108 desired requirements. SSR markers offer advantages in reliability, reproducibility,
109 discrimination, standardization, and cost effectiveness over other marker types (Smith
110 et al, 1997) and have been demonstrated in maize to be a valuable tool for diversity
111 measurements (Warburton et al, 2002; Betrán et al, 2003a).

112 Assessment of genetic diversity among early-maturing inbred lines with
113 contrasting response to drought using molecular markers and determining any
114 association with F₁ hybrid performance under drought would be invaluable in selecting
115 parental lines for development of productive hybrids with tolerance to drought.
116 Molecular markers have been used to assess the genetic diversity of parental lines and
117 to determine the relationship between marker-based genetic distance among pairs of
118 parental lines and grain yield of their hybrids in maize (Menkir et al, 2010; Makumbi
119 et al, 2011; Akaogu et al, 2012).

120 Information on the performance of inbreds in hybrid combinations is crucial
121 for hybrid development programs. It is of great importance for maize breeders to
122 identify potential inbred lines that would produce hybrids exhibiting high levels of
123 heterosis without making all possible crosses among inbreds available in a breeding
124 program. Inbred line information indicative of hybrid performance is desirable to
125 reduce costs and time associated with hybrid production and evaluation. Moll et al
126 (1965) and Paterniani and Lonnquist (1963) reported positive relationship between

127 midparent heterosis (MPH) of F₁ hybrid grain yield and parental genetic divergence.
128 Makumbi et al (2011) also reported positive association between midparent heterosis,
129 specific combining ability, genetic distance (GD), and grain yield. Betrán et al (2003b)
130 observed that the differences in grain yield between hybrid and inbreds increases with
131 the intensity of drought stress.

132 Information on the effect of genetic diversity of parental lines with
133 contrasting response to drought on the performance of their hybrids is limited in the
134 IITA maize improvement program. The objectives of the present study were to (i)
135 assess the genetic diversity among selected early-maturing maize inbreds using SSR
136 markers (ii) assess the relationship between F₁ hybrid performance, heterosis, and
137 SSR-based genetic distance, and (iii) assess genotype × environment interaction and
138 identify high-yielding and stable hybrids across research environments.

139

140 2. Materials and Methods

141 2.1 Genetic materials and field procedures

142 One hundred and fifty six early-maturing inbred lines extracted from six
143 diverse germplasm sources (TZE-W Pop DT STR C₀, WEC STR, TZE-Y Pop DT
144 STR C₀, TZE Comp 5-YC₆, TZE-W Pop x LD and TZE-W Pop x 1368 STR C₀) with
145 tolerance/resistance to *Striga* and maize streak virus, and/or tolerance to drought were
146 used in this study. The procedure adopted for the development of the inbred lines has
147 been described in detail by (Badu-Apraku et al, 2007). Two field experiments were
148 conducted between 2007 and 2010 in Nigeria to evaluate the early-maturing inbreds
149 and their hybrids for tolerance to drought. In addition, the genetic diversity among 42
150 of the 156 inbreds used for the field studies was examined using simple sequence
151 repeat (SSR) markers. In the first experiment, the 156 early-maturing maize inbreds

152 were evaluated under managed drought stress at Ikenne (forest–savanna transitional
153 zone, (6°53'N, 30°42'E, 60 m asl, 1500 mm annual rainfall) in Nigeria during 2007/08
154 and 2008/09 dry seasons and in well-watered environments at Ikenne and Bagauda
155 (Sudan savanna, 12°00' N, 8°22' E, 580 m asl, 800 mm annual rainfall) during the
156 growing seasons of 2008 and 2009. A 12 x 13 randomized incomplete block design
157 with two replications was used for the experiment. Row length was 4 m long with 22
158 plants per row. Row and hill spacing were 0.75 m and 0.4 m, respectively. Three seeds
159 were planted per hill and seedlings were thinned to two per stand about 2 weeks after
160 emergence, giving a population density of 66 666 plants ha⁻¹. A compound fertilizer
161 (NPK 15:15:15) was applied at the rate of 60 kg N ha⁻¹, 60 kg P ha⁻¹ and 60 kg K ha⁻¹
162 at the time of sowing at each test site. An additional 60 kg N ha⁻¹ urea was top-dressed
163 3 weeks later. The experiment was kept weed-free by the application of a mixture of
164 gramaxone (Shandong Dongtai Agricultural Chemistry Co., Ltd.), a contact herbicide,
165 and atrazine, a pre-emergence herbicide, at 5 litres ha⁻¹ each of gramaxone and
166 primextra (Syngenta Crop Protection Canada, Inc.). Subsequently, manual weeding
167 was done periodically to keep the trials weed-free.

168 In the second experiment, 30 early-maturing maize inbred lines selected for
169 contrasting responses to drought in experiment 1 (Table 1) were divided into six sets
170 each of five inbreds. The five inbreds in one set were used as females and crossed with
171 five other inbreds in another set (males) using the North Carolina Design II (NCII)
172 mating scheme of Comstock and Robinson (1948). A total of 150 hybrids along with
173 six checks (three each of drought tolerant open-pollinated varieties and hybrids) were
174 evaluated using 12 x 13 incomplete block design with two replications during the dry
175 seasons of 2008/2009 and 2009/2010 at Ikenne, and during the growing seasons (well-
176 watered or rainfed) of 2008 and 2009 at Ikenne and Bagauda, and Zaria in 2009. The

177 managed drought stress at Ikenne was achieved through an irrigation system that
178 provided 17 mm of water per week up to 28 days after planting. Thereafter, the
179 irrigation water was withdrawn until maturity, so that the maize plants relied on stored
180 water in the soil for growth and development. The hybrids were evaluated using
181 single-row plots. Plant spacing, population density, fertilizer application and other
182 crop management practices were as described earlier for the first experiment.

183

184 *2.2 Microsatellite Assay*

185 The 30 inbred lines used in the NCII mating scheme along with 12 inbred
186 checks from diverse source populations were used for the genetic diversity study. The
187 DNA was extracted from leaves of 10 plants of each inbred line employing CTAB
188 (cetyltrimethyl ammonium bromide) procedure (Saghai-Maroo et al, 1984) modified
189 based on Standard Operating Procedure (Hearne, 2009). Thirty-four labeled SSR
190 markers were selected from the Maize Genetics Database (MaizeGDB)
191 (www.maizegdb.org). At least three markers were selected per chromosome for each
192 of the 10 maize chromosomes. Twenty-three of the thirty-four primer pairs screened
193 on five selected inbreds were found to be polymorphic. The twenty-three polymorphic
194 markers were used for the genotyping of the 42 inbreds. The polymerase chain
195 reaction (PCR) mastermix contained, 10 ng template DNA, 0.25 pmol each primer, 1.6
196 mM MgCl₂, 1X PCR buffer (New England Biolabs (NEB); 1 unit Taq polymerase
197 (NEB), 0.2 mM of each dNTP. Forward primers were labeled at the 5' end with one
198 of four phosphormidite fluorescent dyes, 6FAM™, VIC™, NED™, PET™ (ABI).
199 The thermal cycler (Applied Biosystems (ABI) GeneAmp 9700) reaction profile
200 consisted of 94°C for 2 min, 29 cycles of 94°C for 30 s, 54-64°C for 2 min and 72°C
201 for 5 min. For fragment analysis, 0.5µl of PCR product was added to 9.5 ul of HiDi

formamide (ABI) containing Liz 500 size standard (ABI) (prepared at ¼ manufacturers recommended concentration). Fragment analysis was performed using an ABI 3130xl Genetic Analyser (PE Applied Biosystems, Foster City, Calif.). Data were assembled and allele calls made using GeneMapper V.3.9 software (ABI) with custom designed panels and bin sets.

2.3 Data collection

Days to anthesis and silking were recorded for each plot as the number of days from planting to when 50% of the plants in a row had shed pollen and had emerged silks, respectively. Anthesis-silking interval (ASI) was computed as the interval in days between silking and anthesis. Plant and ear heights were calculated as the average of measurements on 10 competitive plants (excluding plants at the edges) per plot and were measured after anthesis as the distance from the base of the plant to the height of the first tassel branch and the node bearing the upper ear, respectively. Plant aspect was rated on a scale of 1 to 5, where 1 = excellent overall phenotypic appeal and 5 = poor overall phenotypic appeal. Ear aspect was scored on a 1 to 5 scale, where 1 = clean, uniform, large, and well-filled ears and 5 = rotten, variable, small, and partially or poorly filled ears. In addition, stay green characteristic (LD) was recorded on a scale of 1 to 10, where 1 = 10% dead leaf area; 2 = 20% dead leaf area; 3 = 30% dead leaf area; 4 = 40% dead leaf area; 5 = 50% dead leaf area; 6 = 60% dead leaf area; 7 = 70% dead leaf area; 8 = 80% dead leaf area; 9 = 90% dead leaf area; 10 = 100% dead leaf area, for the drought stressed plots at 70 days after planting (DAP). The number of ears per plant (EPP) was computed as the total number of ears at harvest divided by the number of plants at harvest. Under drought environments, all ears harvested from each plot were shelled to determine percentage grain moisture and grain weight. Grain

yield, adjusted to 15% moisture, was computed from the shelled grain weight. On the other hand, under well-watered environments, harvested ears from each plot were weighed and representative samples of ears were shelled to determine percent grain moisture. Grain yield adjusted to 150 g/kg moisture, was computed from ear weight and grain moisture assuming a shelling percentage of 80% (800 g grain/kg ear weight).

232

233 *2.4 Statistical analyses*

234 Separate analysis of variance (ANOVA) were conducted for inbred and
235 hybrid experiments on data collected across years and locations for each research
236 condition (drought stress and well-watered conditions) with General Linear model
237 (GLM) procedure of Statistical Analysis System (SAS) software, version 9.2
238 (SAS, Institute 2001). The location-year combinations under each research
239 condition were referred to as environments in the combined analysis for each field
240 experiment. Environments, replications, and incomplete blocks nested within
241 environments were considered random effects, while research conditions and
242 hybrids were considered as fixed effects. Subsequently, ANOVA was performed
243 for each and across test environments for grain yield to determine the performance
244 and yield stability of the single-cross hybrids across test environments.

245 In order to characterize inbred lines in experiment 1 for tolerance to drought, a
246 base index that combined increased grain yield under drought stress with good plant
247 and ear aspects and delayed leaf senescence, short ASI and increased number of ears
248 per plant and grain yield under well-watered conditions was developed according to
249 Mesaka et al (2006) and Badu-Apraku et al (2011b). Since each parameter was
250 standardized with a mean of zero and standard deviation of 1 to avoid the effects of
251 different scales, a positive value indicated tolerance of lines to drought while a

negative value indicated susceptibility of lines to drought. The base index value was calculated using the following equation:

Base index = $[(2 \times \text{Yield}_{\text{DS}}) + \text{Yield}_{\text{WW}} + \text{EPP} - \text{ASI} - \text{Plant aspect} - \text{Ear aspect} - \text{LD}]$

where, DS = drought stress conditions,

WW = well-watered conditions

Average heterosis for grain yield was computed as the difference between F_1 value and the mid-parent value (Hallauer and Miranda, 1988). Mid-parent value for a cross was computed as the mean of the two parental lines for each research condition.

Mid-parent heterosis (MPH) for individual crosses was calculated as:

$$\text{MPH} = \frac{(F_1 - \text{MP}) \times 100}{\text{MP}}$$

where, F_1 is the mean of the hybrid performance and $\text{MP} = (P_1 + P_2)/2$ in which P_1 and P_2 were the means of the inbred parents, respectively.

Also, Better-parent heterosis for individual crosses was calculated as:

$$\text{BPH} = \frac{(F_1 - \text{BP}) \times 100}{\text{BP}}$$

where, BP was the better-parent

Twenty five hybrids, including the top 20 and worst five along with the best two checks were selected based on combined ANOVA across research environments.

The grain yield data of the hybrids were subjected to an additive main effects and multiplicative interaction (AMMI) analysis to assess relationships among hybrids, environments and interactions between test environments and hybrids (Zobel et al., 1988). This analysis was done using GGE biplot, a Windows application that fully automates biplot analysis, version 5.4 (Yan, 2001). The beta version of the program is

276 available at www.ggebiplot.com (verified 12 July 2012). Furthermore, the outstanding
277 hybrid was identified using the rank summation index (Ajala, 2010).

278 For the molecular data, the electropherograms obtained from the ABI 3130xl
279 Genetic Analyser were analysed with GeneMapper 4.0 software (Applied Biosystems).
280 Allele calls were exported as an excel file and data analyzed using PowerMarker
281 V3.25 (Lu, 2004). The software generated number of alleles and polymorphic
282 information content (PIC) for each of the SSR loci. A Modified Roger's distance
283 (MRD) matrix was generated from the data using the R open source software
284 (<http://www.r-project.org/>) and subsequently grouped by Ward minimum variance
285 within group method (1963) using the DARwin 5 software (Perrier and Jacquemoud-
286 Collet, 2006).

287 Correlations were computed between grain yield and the trait means of F₁
288 hybrids under drought and optimal growing conditions. Similarly, correlation analysis
289 was conducted between the GD estimates of pairs of parental lines and the trait means
290 of the corresponding F₁ hybrids, MPH, BPH observed under drought and optimal
291 growing environments. The relationship between *per se* performance of the parental
292 lines and their F₁ hybrids was determined under drought and optimal growing
293 conditions using regression analysis. Both correlation and regression analyses were
294 carried out using the SAS package, version 9.2 (SAS Institute, 2001).

295

296 3. Results

297 3.1 Performance of inbred lines under drought and well-watered conditions

298 Results of the combined analysis of variance of inbreds across environments
299 showed significant ($P < 0.01$) genotype (G), environment (E), and G $\times$ E interaction
300 (GEI) mean squares for grain yield and all other measured traits under drought and

301 well-watered conditions (data not shown). Grain yield of the inbreds ranged from 0.06
 302 t ha⁻¹ for TZEI 123 to 1.92 t ha⁻¹ for TZEI 17 under drought and 0.58 t ha⁻¹ for TZEI
 303 123 and 2.31 t ha⁻¹ for TZEI 17 under well-watered conditions (Table 1). TZEI 17 had
 304 the highest drought tolerance index (15.31) while TZEI 30 had the lowest (-10.45).
 305 Under drought, the grain yield of TZEI 17 represented 83% of what it would have
 306 produced in the same environment under optimal conditions. Under drought stress, the
 307 least reduction in grain yield was observed for TZEI 112 (4%) while the greatest
 308 reduction was recorded for TZEI 24 (84%) (Table1). Twenty-four of the 30 inbreds in
 309 the present study had positive index values for grain yield. The improved grain yield
 310 of inbreds was associated with shorter ASI, better ear and plant aspects, higher EPP,
 311 and lower rate of leaf senescence under drought conditions (data not shown).

312

313 *3.2 Performance of single-cross hybrids under drought and well-watered conditions*

314 Combined analysis of variance showed significant ($P < 0.01$) mean squares for
 315 genotypes, water treatment, and environments (water treatment) for grain yield and all
 316 other measured traits except husk cover and plant aspect for water treatment (data not
 317 shown). The interactions of hybrids with water treatment were significant ($P < 0.01$)
 318 for all traits except grain yield, ear aspect and the stay green characteristic. However,
 319 the interaction of hybrids with environment (water treatment) had significant effects
 320 on grain yield and all other traits except plant aspect. Significant ($P < 0.01$)
 321 correlations were observed between the rankings of hybrids for all traits under drought
 322 stress and well-watered conditions. Grain yield of hybrids ranged from 0.31 to 2.86 t
 323 ha⁻¹ under drought and 2.24 to 8.77 t ha⁻¹ under well-watered conditions. The mean
 324 grain yield under drought stress represented 39% of what the average yield would have
 325 been in the same environments under well-watered conditions. On the average,

induced moisture stress reduced grain yield by 61%. Similarly, reductions were observed in days to anthesis (9%), days silking (7%), plant height (7%), ear height (4%), and number of ears per plant (22%) under induced drought stress. On the other hand, significant increases were observed in ASI (36%) and ear aspect (10%). Out of the 12 top-yielding single-cross hybrids selected using the base index, 10 hybrids involved $T \times T$ while the other two involved $T \times S$ crosses (Table 2). In contrast, out of the worst nine single-cross hybrids, four involved $T \times T$, four $T \times S$ while the remaining involved $S \times S$ crosses. The best single-cross hybrid TZEI 31 $\times$ TZEI 18 out-yielded the best open-pollinated cultivar, TZE Comp3 C₃ by 19% under drought and 34% under well-watered conditions.

3.3 Genetic diversity of inbred lines

The 23 SSR markers used for the current study were dispersed on all 10 chromosomes, with at least two markers per chromosome except chromosome nine, which had one SSR marker. All the markers were polymorphic. A total of 130 alleles were distributed among the 42 early-maturing maize lines using 23 SSR primers (Table 3). The number of amplified alleles per marker varied from two for marker *nc133* to nine for *phi299852* with an average of 5.7, indicating a high level of genetic diversity among the lines used. Sizes of alleles amplified by the markers ranged from 62 to 435 bp. Primers *phi014*, *phi085* and *phi227562* amplified larger size alleles as compared with other primers. The Polymorphic information content (PIC) per marker ranged from 0.17 for marker *phi308707* to 0.77 for *phi084* with a mean value of 0.54 (Table 3). SSR markers *phi084*, *phi029*, *phi299852*, *phi085*, *phi031* and *phi085* showed PIC values equal to or greater than 0.70. All of these markers, except *phi029*, amplified a higher number of alleles (range 7-9). Significant positive correlation ($r =$

0.80, $p < 0.001$) was observed between PIC values and number of alleles amplified per primer. A total of 31 unique/rare alleles (alleles found in one inbred out of 42 or less than 0.03 allele frequency) were detected in 14 SSR markers across 21 of the 42 lines analyzed (data not shown). Only two inbreds had unique alleles ranging from three for TZEI 2 to four for TZEI 4. Heterozygosity ranged from 0 in *phi062* to 0.55 in *phi115* (Table 3), indicating that all the 42 inbred lines were homozygous at *phi062* locus but that residual heterozygosity was present in some genomic regions despite inbreeding.

Genetic distance estimates based on SSR markers expressed as Euclidean distance for all 861 pairwise comparisons among the 42 maize inbred lines ranged from 0.42 (TZEI 26 vs TZEI 108) to 0.85 (TZEI 24 vs TZEI 4) (data not shown). The mean genetic distance for all pairwise comparisons was 0.67. Low genetic distance estimates were observed between inbred lines TZEI 26 and TZEI 108 (0.42), TZEI 157 and TZEI 149 (0.49), and TZEI 161 and TZEI 149 (0.43). High genetic distance estimates were observed between TZEI 24 and TZEI 4 (0.85), TZEI 22 and TZEI 63 (0.80), TZEI 129 and TZEI 51 (0.83), TZEI 63 and TZEI 51 (0.80), TZEI 31 and TZEI 24 (0.83), and TZEI 31 and TZEI 4 (0.80). Results of the cluster analysis of the 42 lines on the basis of SSR marker based genetic distance estimates resulted in distinct separation of the lines into five groups (Fig. 1). The number of inbred lines in a cluster ranged from five for cluster 4 to 15 for cluster 1.

3.4 Correlation of genetic distance of inbreds with performance of F₁ hybrids and inbred-hybrid relationship

Grain yield of F₁ hybrids was positively correlated with HP, MP, MPH, and BPH ($p < 0.01$) under both drought and well-watered environments (Table 4). Grain yield of inbreds was significantly correlated with F₁ hybrid yield under both research

conditions. Similarly, MPH and BPH had significant positive ($P < 0.01$) correlations with grain yield, plant height, and EPP but negative correlations with days to anthesis and silking, plant and ear aspects, husk cover, and LD of F₁ hybrids under drought. Similarly, MPH and BPH had significant positive ($P < 0.01$) correlations with grain yield, plant and ear heights but negative correlations with days to anthesis and silking, ASI, and plant and ear aspects under well-watered conditions. Grain yield had significant and positive correlations with plant and ear heights and EPP but negative correlation with days to anthesis and silking, ASI, plant and ear aspects, husk cover, and LD under both research conditions except LD under well-watered conditions. Correlations between GD estimates of the pairs of parental lines and hybrid means were not significant for grain yield and other measured traits under drought and well-watered conditions (Table 4).

The mid-parental values for grain yield under drought stress allowed only modest prediction of hybrid grain yield; that is, one t ha⁻¹ increase in mid-parent grain yield resulted in only 0.45 t ha⁻¹ increases in hybrid grain yield under drought and 0.60 t ha⁻¹ under well-watered conditions (Fig. 2). Regression analysis also revealed that the variation in grain yield of the parental lines explained 0.08 of the total variation in grain yield of F₁ hybrid under drought and 0.05 under well-watered conditions (Fig. 2).

395

396 *3.5 AMMI biplot analysis of grain yield and stability of early-maturing hybrids*

The highly significant GEI for grain yield and most measured traits under drought and well-watered conditions justified the use of the AMMI biplot to determine the yield performance and stability of the hybrids in the seven research environments. The AMMI biplot with genotype and environment main effects for grain yield on the abscissa (x) and the first interaction principal component axis (IPC1) on the ordinate

(y) is presented in Fig. 3. The biplot revealed that E accounted for 65.53% of the total variation in the sum of squares for grain yield, G accounted for 23.42% and IPC1 accounted for 3.8%. Thus, E, G, and IPC1 together accounted for 92.8% of the total variation in the sum of squares for grain yield (Fig. 3). This result indicated that the biplot is effective in explaining both the main effects as well as the G x E interaction. The vertical line is the grand mean for grain yield while the horizontal line represents the IPCA1 value of zero. Hybrids close to the horizontal line have small interactions and therefore considered to be more stable than those further from it. The biplot showed large variability among the hybrids and environments. Hybrids, 9 and 11 had similar grain yield response (above the grand mean) and strong positive interaction with IPCA1 score. The large interaction of 9 and 11 hybrids with IPCA1 indicated that the hybrids may probably adapted to high-yielding environment (IK08WW). In contrast, 5, 6, and 8 had similar grain yield response (above the grand mean) and strong negative interaction with IPCA1 score and thus, probably adapted to ZA09WW. Hybrids, 12, 13, 27, and 3 were characterized by similar grain yield response (above the grand mean) and had IPCA1 scores near zero and therefore had small interaction with the environments. The closeness of 12, 13, 27, and 3 hybrids to near zero IPCA1 indicated that they have comparable mean grain yields and are similar in their interaction with the different environments, and therefore are the most stable hybrids. Hybrid, 2 (TZEI 31 x TZEI 18) had similar grain yield response (above the grand mean) with 11 and 9 and had relatively low positive interaction with IPCA1 score and thus, identified as the highest-yielding and stable hybrid across environments. The rank summation index computed for the best 11 hybrids based on the results of AMMI biplot analysis revealed that hybrid 2 (TZEI 31 x TZEI 18) had an outstanding yield performance and low rank summation index across the seven environments (Table 5).

427 **4. Discussion**

428 The significant variation among inbreds, hybrids, and environments for grain
429 yield and other measured traits demonstrated that the test environments were unique
430 and that adequate genetic variation existed among the early-maturing maize inbreds
431 and hybrids. The implication is that significant progress could be made from selection
432 of inbreds for improvements in most of the measured traits for the development of
433 productive early-maturing maize hybrids for drought prone environments in WCA.
434 This result corroborates the findings of Badu-Apraku et al (2011b) and Badu-Apraku
435 and Oyekunle (2012). The significant interaction between hybrid and environment for
436 grain yield and other measured traits across environments indicated that the expression
437 of these traits would not be consistent in varying test environments. Similar findings
438 have been reported by several other workers (Badu-Apraku et al, 2003, 2011a, b) and
439 suggest the need to evaluate inbreds and hybrids in varying environments to identify
440 drought tolerant genotypes with consistently favorable response to unpredictable
441 growing environments. The reduction in the days to anthesis and silking, EPP, grain
442 yield, deterioration in plant and ear aspects and the increased ASI under drought is
443 consistent with the results of several earlier workers (Bolanos and Edmeades, 1993;
444 Edmeades et al, 1993). The severe grain yield reduction of 61% observed under
445 managed drought in the present study falls within the range reported by earlier workers
446 (Badu-Apraku et al, 2005, Campos et al, 2006; Derera et al, 2008, Badu-Apraku et al,
447 2011b). Under drought stress the hybrids flowered about 5 days earlier and hence had
448 shorter growing cycles and reduced leaf area duration and this might have accounted
449 for the lower grain yield compared to that under well-watered conditions. This result is
450 in agreement with the findings of Derera et al (2008).

451 The superior grain yield of hybrids involving two tolerant inbreds compared to
452 those involving tolerant x susceptible or two susceptible inbreds indicated the
453 importance of dosage effect of drought tolerance genes in parental lines. This result is
454 in agreement with the findings of (Kirkham et al, 1984; Betrán et al, 2003b; Meseka et
455 al, 2007, Badu-Apraku et al, 2011b) who reported that hybrids with at least one
456 drought tolerant parental lines would give higher yields than those formed from two
457 susceptible lines.

458 Prediction of hybrid performance based on the per se performance of the
459 parental lines is of crucial importance for genetic gain from selection. The significant
460 positive correlation observed between mid-parent values and hybrid means for grain
461 yield and other traits indicated that preliminary screening of parental lines for drought
462 tolerance would be useful for the development of superior hybrids for the drought
463 prone environments of WCA. This finding is in agreement with those of Meseka et al
464 (2006) and Betrán et al (2003b) who reported significant relationship between mean
465 grain yields of parental lines and their hybrids under low-N than under high-N
466 conditions. The positive and significant correlation observed between MPH and grain
467 yield in this study is consistent with the findings of Makumbi et al (2011) and Betran
468 et al (2003b). Furthermore, the presence of strong associations between grain yield and
469 number of ears per plant, plant and ear heights, ASI, days to anthesis and silking, plant
470 and ear aspects and rate of leaf senescence of F₁ hybrids under drought and well-
471 watered conditions justified the inclusion of most of these traits in the base index for
472 selection of genotypes for tolerance to drought. This result is in agreement with the
473 findings of Badu-Apraku et al (2011b) and Badu-Apraku and Oyekunle (2012).

474 The mean number of alleles (5.7 per primer) observed in the present study was
475 similar to those reported by several other workers (Senior et al, 1998; Lu and

476 Bernardo, 2001; Warburton et al, 2002). However, the mean number of alleles
477 observed was higher than those reported by earlier workers (Bantte and Prasanna,
478 2003; Makumbi et al, 2011; Legesse et al, 2007; Akaogu et al, 2012) but lower than
479 those reported by Pejic et al, (1998) and Adetimirin et al (2008). Differences in
480 numbers of alleles between studies could be attributed to the size of the samples
481 studied, expected diversity or uniformity based on pedigrees, high throughput
482 genotype used by various workers and most importantly, repeats type of microsatellite
483 used. For example, dinucleotide SSR primers show higher numbers of alleles (Enoki et
484 al, 2002; Legesse et al, 2007; Adetimirin et al, 2008), but were not used in the present
485 study except marker *phi112*. The average PIC value of 0.54 observed in this study was
486 lower than the values reported by other researchers for maize inbred lines using SSR
487 markers (Choukan et al, 2006; Adetimirin et al, 2008; Legesse et al, 2007). However,
488 some markers with high PIC values and hence high discriminatory power were
489 identified; *phi084* (0.77), *phi029* (0.70), *phi031* (0.75), *phi076* (0.76), *phi085* (0.76)
490 and *phi299852* (0.70). According to Bantte and Prasanna (2003), the overall PIC value
491 could be influenced by the type of germplasm used, number and distribution of SSR
492 loci assayed, type of marker used and methodology employed for allele detection.

493 The detection of unique alleles in some of the inbred lines analyzed indicated
494 the usefulness of SSR markers in germplasm identification. Presence of unique alleles
495 that are specific to single accessions were reported by Bantte and Prasanna (2003) and
496 Choukan et al (2006). The average genetic distance among all the inbred lines was
497 relatively high (0.67), indicating that there was considerable genetic diversity among
498 the early-maturing inbreds. This result corroborates the findings of Menkir et al
499 (2005), Legesse et al (2007), and Akaogu et al (2012). The low genetic distance of
500 0.42 obtained for TZEI 26 vs TZEI 108 is not surprising because these inbreds are

501 related by pedigree and belong to the same group. In contrast, the highest genetic
502 distance of 0.85 observed for TZEI 24 vs TZEI 4 confirmed the unrelatedness of these
503 inbreds by pedigree.

504 The dendrogram obtained from the Ward's minimum variance clustering based
505 on SSR markers data identified five distinct groups. Cluster IV contained mostly,
506 closely related inbred lines. All inbred lines in cluster III and IV were yellow-grained
507 maize except TZEI 187 in cluster III. These results indicated that the grouping of the
508 inbreds was based largely on the pedigree of the inbreds and to a lesser extent on the
509 reaction of the inbreds to the stress environments. This result is consistent with the
510 findings of Menkir et al (2010), Makumbi et al (2011), and Akaogu et al (2012).

511 The detection of more than one allele at some of the SSR loci for some inbreds
512 may be due to residual heterozygosity. Duplication in maize and the amplification of
513 similar sequences in two separate genomic regions is another plausible explanation for
514 the observed "heterozygosity" (Helentjaris, 1995). These results are similar to the
515 findings of Senior et al (1998) and Adetimirin et al (2008). It is striking, however, that
516 *phi062* detected homozygosity for all the 42 inbred lines used in this study.

517 The lack of association between GD and F₁, MPH, and BPH under both
518 research conditions indicated that GD was not a good predictor of hybrid performance
519 or heterosis in maize under drought and well-watered conditions. This result is
520 contrary to the findings of Betrán et al (2003a) who reported that the correlation of GD
521 with F₁, SCA, MPH, and HPH increased when the drought stress levels decreased.
522 However, this result corroborates the findings of several other workers (Shieh and
523 Thseng, 2006; Benchimol et al, 2008; Menkir et al, 2010). The lack of association
524 between GD and F₁ hybrid performance could be attributed to the absence of linkage
525 between genes controlling the trait and markers used to estimate GD, inadequate

526 genome coverage, different levels of dominance among hybrids (Bernardo, 1992) and
527 presence of epistasis (Moll et al, 1965).

528

529 5. Conclusions

530 Results of this study showed that significant differences existed among inbreds and
531 hybrids for most traits under both research conditions and that drought tolerance in at
532 least one of the parental lines would be desirable to achieve acceptable hybrid
533 performance under drought. The broad range of genetic diversity obtained with SSR
534 markers indicated the presence of a significant reservoir of diversity among tropical
535 early-maturing maize inbred lines that could be utilized in breeding programs in WCA
536 to develop productive drought tolerant hybrids and synthetics. The lack of association
537 between the microsatellite-based GD estimates of the parental inbreds and the mean
538 grain yield and other traits of their F₁ hybrids in contrasting environments implied that
539 the microsatellite-based GD between pairs of inbreds could not be used to predict the
540 performance of hybrids under drought and well-watered conditions. The high-yielding
541 and stable hybrid identified in this study, TZEI 31 × TZEI 18 should be tested
542 extensively in multilocation trials and promoted for adoption and commercialization in
543 WCA to contribute to increased food security.

544

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Table 1. Grain yield of parental maize inbred lines used in Design II crosses evaluated under drought at Ikenne in 2007/08 and 2008/09 dry seasons and well-watered conditions at Ikenne and Bagauda in 2008 and 2009.

Name	Pedigree	Response†	Grain yield		Yield reduction (%)	Base index	Grain colour
			(t/ha)				
			DS	WW			
TZEI 17	TZE COMP5-Y C6 S6 Inb 35	T	1.92	2.31	17	15.31	Yellow
TZEI 129	TZE-Y Pop STR C0 S6 Inb 16-1-3	T	1.67	2.27	26	13.28	Yellow
TZEI 31	TZE-W Pop x LD S6 Inb 4	T	1.75	2.58	32	12.70	White
TZEI 18	TZE-W Pop STR C0 S6 Inb 136-3-3	T	1.83	3.13	42	11.61	White
TZEI 16	TZE COMP5-Y C6 S6 Inb 31	T	1.42	2.15	34	11.57	Yellow
TZEI 158	TZE-Y Pop STR C0 S6 Inb 102-2-2	T	1.67	2.28	27	10.73	Yellow
TZEI 149	TZE-Y Pop STR C0 S6 Inb 66-2-2	T	1.54	1.86	17	9.36	Yellow
TZEI 178	TZE COMP5-Y C6 S6 Inb 62-3-3	T	1.53	2.07	26	8.36	Yellow
TZEI 63	TZE-W Pop STR C0 S6 Inb 136-2-3	T	1.65	1.99	17	7.68	White
TZEI 107	WEC STR S7 Inb 19B	T	1.23	2.34	48	6.70	White
TZEI 161	TZE-Y Pop STR C0 S6 Inb 103-2-3	T	1.27	1.54	18	6.67	Yellow
TZEI 87	TZE-W Pop x 1368 STR S7 Inb 11	T	1.63	2.23	27	6.34	White
TZEI 26	WEC STR S7 Inb 4	T	1.50	1.86	19	6.27	White
TZEI 157	TZE-Y Pop STR C0 S6 Inb 102-1-2	T	1.27	2.07	39	6.19	Yellow
TZEI 135	TZE-Y Pop STR C0 S6 Inb 17-2-3	T	1.27	2.29	44	5.89	Yellow
TZEI 2	TZE-W Pop x 1368 STR S7 Inb 2	T	1.49	1.92	22	5.66	White
TZEI 8	TZE-Y Pop STR C0 S7 Inb 62-3-3	T	1.35	1.59	15	5.41	Yellow
TZEI 177	TZE COMP5-Y C6 S6 Inb 62-1-2	T	1.01	2.83	64	5.12	Yellow
TZEI 59	TZE-W Pop STR C0 S6 Inb 80	T	1.42	1.94	27	4.63	White
TZEI 108	WEC STR S7 Inb 7	T	1.27	1.61	21	4.53	White
TZEI 112	WEC STR S7 Inb 16-1-2	T	1.34	1.39	4	2.50	White
TZEI 165	TZE COMP5-Y C6 S6 Inb 8A	T	1.11	1.72	36	1.91	Yellow
TZEI 51	TZE-W Pop STR C0 S6 Inb 57-4-4	T	1.13	1.66	32	0.20	White
TZEI 21	TZE-W Pop x 1368 STR S7 Inb 1	T	1.14	1.64	31	0.17	White
TZEI 27	TZE-Y Pop STR C0 S7 Inb 1-4-5	S	0.66	2.04	68	-0.05	Yellow
TZEI 122	TZE-Y Pop STR C0 S6 Inb 2-2-3	S	0.53	1.69	69	-0.09	Yellow
TZEI 89	TZE-W Pop x 1368 STR S6 Inb 14	S	0.65	1.57	59	-4.74	White
TZEI 24	TZE-Y Pop STR C0 S6 Inb 142-2-2	S	0.33	2.07	84	-6.18	Yellow
TZEI 187	TZE-W Pop STR C0 S6 Inb 14-2-3	S	0.38	1.84	80	-8.07	White
TZEI 30	TZE-W Pop STR C0 S6 Inb 14-3-3	S	0.33	1.56	79	-10.45	White
Mean			0.84	1.62	48		
S.E			0.30	0.35			

†T = Tolerant and S = Susceptible.

Table 2. Grain yield and other agronomic traits of 26 of 150 maize single-cross hybrids (the best 12 and the worst nine based on grain yield) evaluated under induced drought stress at Ikenne during 2007/08 and 2008/09 dry seasons and well-watered conditions at Ikenne and Bagauda in 2008 and 2009; Zaria in 2009.

Hybrid	Category [†]	Grain yield,		Days to silk		ASI		Plant height, m		Stalk lodging,		Husk cover		Plant aspect		Ear aspect		LD	Ears per plant	
		t/ha								%										
		DS [‡]	W	DS	W	DS	W	DS	W	DS	W	DS	W	DS	W	DS	W	DS	DS	W
TZEI 31 × TZEI 18	T x T	2.86	5.06	52	57	2.0	1.1	1.50	1.67	13.8	7.2	2.0	2.5	2.3	2.5	2.2	2.1	3.6	0.9	0.9
TZEI 31 × TZEI 107	T x T	2.84	4.66	49	54	1.4	1.0	1.54	1.61	19.6	7.0	2.5	2.3	1.8	2.7	2.1	2.4	2.8	0.8	1.0
TZEI 63 × TZEI 108	T x T	2.68	4.46	50	55	1.7	0.8	1.46	1.59	25.6	6.7	2.0	3.6	2.3	2.3	2.1	2.3	3.6	0.8	1.0
TZEI 27 × TZEI 161	S x T	2.68	4.74	50	55	1.6	0.9	1.60	1.64	25.0	6.6	2.0	2.2	2.6	2.6	2.3	2.3	3.6	0.9	1.0
TZEI 51 × TZEI 31	T x T	2.65	4.47	50	55	1.6	1.4	1.48	1.53	11.8	2.9	1.8	2.4	2.3	2.6	2.2	2.6	4.3	0.9	1.0
TZEI 63 × TZEI 112	T x T	2.58	4.38	50	55	1.4	1.1	1.57	1.66	23.4	8.3	1.9	2.1	2.2	2.4	2.2	2.4	4.2	0.9	1.0
TZEI 178 × TZEI 17	T x T	2.55	4.56	48	55	1.4	1.3	1.41	1.48	15.6	3.4	2.3	2.4	2.5	2.7	2.0	2.2	3.4	0.9	1.0
TZEI 18 × TZEI 108	T x T	2.47	4.63	49	57	0.8	1.6	1.56	1.60	26.9	3.0	1.8	1.9	2.0	2.7	2.2	2.0	4.2	0.9	1.0
TZEI 31 × TZEI 63	T x T	2.44	5.36	51	57	1.6	1.5	1.61	1.73	14.6	3.4	3.0	2.6	2.2	2.3	2.3	2.0	3.3	0.8	1.0
TZEI 177 × TZEI 165	T x T	2.44	5.10	51	55	2.3	1.1	1.52	1.53	15.4	3.6	1.8	2.1	2.6	2.3	2.4	1.9	3.3	0.9	1.0
TZEI 26 × TZEI 63	T x T	2.40	4.93	51	56	2.4	1.1	1.46	1.80	13.7	10.3	2.3	2.7	2.5	2.3	2.5	2.2	4.2	0.8	1.0
TZEI 89 × TZEI 31	S x T	2.31	4.83	51	56	2.0	1.6	1.61	1.66	12.4	8.8	2.1	2.4	2.2	2.7	2.5	2.1	3.2	0.8	0.9
TZEI 27 × TZEI 8	S x T	1.00	4.13	52	53	1.3	1.0	1.38	1.50	20.8	5.1	2.6	3.0	3.4	2.9	3.0	2.6	4.1	0.6	1.0
TZEI 187 × TZEI 21	S x T	0.90	3.65	52	56	2.5	1.0	1.52	1.78	19.3	8.2	2.9	2.9	2.7	3.0	3.2	2.7	5.0	0.7	0.9
TZEI 8 × TZEI 158	T x T	0.89	2.76	51	57	0.3	1.1	1.43	1.32	37.2	4.7	2.1	2.4	3.1	3.1	2.8	2.8	4.2	0.6	1.0
TZEI 89 × TZEI 187	S x S	0.84	4.45	55	55	4.0	1.2	1.59	1.78	14.6	3.0	2.4	2.6	3.2	2.7	3.4	2.4	5.1	0.5	0.9
TZEI 122 × TZEI 177	S x T	0.79	2.24	55	56	1.8	1.6	1.21	1.26	40.4	5.6	2.9	2.0	3.1	3.3	3.3	3.2	4.9	0.5	0.8
TZEI 21 × TZEI 2	T x T	0.78	2.33	54	58	3.5	1.5	1.47	1.58	20.8	7.1	2.7	2.1	3.4	5.2	3.2	3.0	6.0	0.4	0.8
TZEI 157 × TZEI 161	T x T	0.76	2.58	54	57	1.1	1.3	1.34	1.43	36.8	6.1	3.3	2.4	3.2	3.1	3.3	3.2	4.3	0.6	1.0
TZEI 161 × TZEI 158	T x T	0.72	2.31	53	58	1.3	0.9	1.23	1.37	34.8	7.2	2.6	2.1	3.3	3.4	3.2	3.2	4.4	0.5	0.9
TZEI 108 × TZEI 187	T x S	0.31	3.42	56	54	4.8	0.7	1.51	1.79	9.9	10.8	3.1	2.8	3.3	3.0	3.5	3.0	5.6	0.4	0.9
TZE COMP3 C3		2.32	3.36	53	59	1.7	2.0	1.60	1.62	11.1	4.5	2.3	1.9	2.2	2.6	2.1	2.6	4.2	0.8	1.0
Mean		1.70	4.41	51	56	1.8	1.2	1.50	1.62	22.0	5.3	2.3	2.3	2.7	2.7	2.7	2.4	4.3	0.8	1.0
SE		0.340	0.042	1.0	0.1	0.81	0.02	0.100	0.011	9.60	4.82	0.50	0.02	0.33	0.04	0.33	0.02	0.51	0.10	0.03

[†] T = Tolerant, S = Susceptible; [‡]DS, Drought stress; W, well-watered condition.

Table 3. SSR marker information, number of alleles detected in maize inbreds, polymorphic information content (PIC) and heterozygosity at SSR loci.

SSR Marker	Bin	Repeat motif	Size (bp)	No. of allele	PIC	Heterozygosity
<i>phi084</i>	10.04	GAA	148-157	7	0.77	0.50
<i>phi062</i>	10.04	ACG	159-164	4	0.21	0.00
<i>phi059</i>	10.02	ACC	147-165	6	0.57	0.19
<i>phi072</i>	4.01	AAAC	142-162	8	0.62	0.24
<i>umc1196</i>	10.07	CACACG	140-160	7	0.55	0.19
<i>phi046</i>	3.08	ACGC	62-66	4	0.42	0.52
<i>phi115</i>	8.03	TA-ATAC	242-258	8	0.66	0.55
<i>phi308707</i>	1.1	AGC	118-125	4	0.17	0.02
<i>phi079</i>	4.05	AGATG	180-220	3	0.31	0.17
<i>phi112</i>	7.01	AG	148-158	4	0.35	0.21
<i>phi029</i>	3.04	AG/AGCG	150-161	5	0.70	0.07
<i>phi102228</i>	3.06	AAGC	120-131	4	0.53	0.21
<i>phi331888</i>	5.04	AAGC	129-136	4	0.45	0.29
<i>nc133</i>	2.05	GTGTC	110-120	2	0.29	0.40
<i>phi031</i>	6.04	GTAC	186-225	8	0.75	0.36
<i>phi069</i>	7.05	GAC	187-207	5	0.60	0.24
<i>phi299852</i>	6.07	AGC	99-132	9	0.70	0.52
<i>phi085</i>	5.06	AACGC	243-267	7	0.76	0.02
<i>phi014</i>	8.04	GGC	431-435	6	0.63	0.20
<i>phi083</i>	2.04	AGCT	125-139	6	0.61	0.37
<i>phi227562</i>	1.11	ACC	309-325	8	0.66	0.26
<i>phi108411</i>	9.05	AGCT	116-123	4	0.44	0.12
<i>phi076</i>	4.11	AGCGGG	161-172	7	0.76	0.36
Mean				5.7	0.54	0.28

Table 4. Correlation among GD, mid-parent heterosis (MPH), better-parent heterosis (BPH), grain yield, and other measured traits of 150 single-cross hybrids under drought stress (above diagonal) and well-watered conditions (below diagonal).

	Grain yield	Days to anthesis	Days to anthesis	ASI	Plant height	Husk cover	Plant aspect	Ear aspect	EPP	GD	MP	HP	MPH	BPH	LD
Grain yield		-0.45**	-0.50**	-0.22**	0.31**	-0.37**	-0.76**	-0.86**	0.78**	0.04	0.29**	0.29**	0.48**	0.54**	-0.51**
Days to anthesis	-0.28**		0.78**	-0.10	-0.23**	0.13	0.32**	0.37**	-0.50**	-0.08	-0.09	-0.17	-0.21**	-0.19	0.08
Days to silking	-0.26**	0.96**		0.54**	-0.07	0.25**	0.33**	0.47**	-0.61**	-0.09	-0.18**	-0.18**	-0.22**	-0.24**	0.23**
ASI	-0.17**	0.35**	0.53**		0.18**	0.22**	0.12	0.26**	-0.31**	-0.03	-0.17**	-0.08	-0.06	-0.12	0.26**
Plant height	0.40**	0.01	0.05	-0.04		-0.09	-0.35**	-0.25**	0.21**	-0.08	-0.05	0.03	0.23**	0.19**	0.05
Husk cover	-0.05	-0.10	-0.11	-0.07	0.24**		0.34**	0.30**	-0.45**	-0.11	-0.08	-0.06	-0.25**	-0.28**	0.21**
Plant aspect	-0.58**	0.19*	0.17*	0.12	-0.38**	0.15		0.70**	-0.58**	-0.01	-0.17*	-0.16	-0.42**	-0.46**	0.46**
Ear aspect	-0.64**	0.07	0.04	0.03	-0.30**	0.18**	0.56**		-0.64**	0.05	-0.29**	-0.32**	-0.38**	-0.43**	0.50**
EPP	0.12	-0.09	-0.11	-0.16**	-0.15	-0.07	-0.12	-0.11		0.10	0.26**	0.26**	0.36**	0.39**	-0.41**
GD	-0.04	0.02	0.00	-0.03	-0.01	-0.04	-0.09	0.03	-0.07		0.03	0.04	0.01	0.01	0.10
MP	0.22**	-0.08	-0.05	0.06	-0.15	-0.19**	-0.16**	-0.24**	0.22**	-0.02		0.81**	-0.62**	-0.39**	-0.14
HP	0.28**	0.09	0.12	0.11	-0.01	-0.19**	-0.19**	-0.26**	0.05	0.02	0.76**		-0.55**	-0.56**	-0.14
MPH	0.77**	-0.20**	-0.20**	-0.20**	0.47**	0.07	-0.44**	-0.44**	-0.01	-0.03	-0.45**	-0.25**		0.91**	-0.28**
BPH	0.56**	-0.28**	-0.29**	-0.25**	0.36**	0.12	-0.33**	-0.30**	0.04	-0.06	-0.48**	-0.62**	0.84**		-0.31**

MP: mid-parent; BP: high-parent; MPH: mid-parent heterosis; BPH: better-parent heterosis; GD: genetic distance

Table 5. Grain yield of the best 11 single-cross maize hybrids based on the AMMI analysis evaluated at seven environments in Nigeria, 2008 and 2009.

PEDIGREE	Code	IK08DS	IK09DS	BG08TD	BG09TD	IK08WW	IK09WW	ZA09WW	Rank summation index
----- t ha ⁻¹ -----									
TZEI 31 × TZEI 18	2	3.14	2.76	6.62	2.77	5.45	4.68	7.00	27
TZEI 24 × TZEI 16	3	1.87	1.79	7.00	4.11	5.66	3.69	6.30	42
TZEI 129 × TZEI 161	4	2.32	2.28	7.27	4.32	5.03	3.99	5.92	40
TZEI 16 × TZEI 122	5	2.34	2.41	6.72	3.94	4.10	3.12	7.60	43
TZEI 177 × TZEI 165	6	2.37	2.50	6.32	3.92	5.17	2.94	6.65	47
TZEI 26 × TZEI 63	7	1.96	2.95	6.60	2.26	6.04	4.56	5.24	41
TZEI 129 × TZEI 178	8	1.59	2.60	7.08	4.49	4.84	3.53	5.81	45
TZEI 16 × TZEI 135	9	2.06	2.67	6.33	4.06	5.20	6.00	5.56	39
TZEI 18 × TZEI 89	10	1.76	2.42	6.22	3.80	5.93	4.16	5.72	49
TZEI 89 × TZEI 31	11	2.66	1.93	6.39	2.61	5.23	5.32	6.79	41
CHECK 4 - TZEI 26 × TZEI 5	27	1.62	2.42	5.77	3.68	5.29	4.30	7.02	48

IK08DS and IK09DS = Ikenne under drought stress in 2008 and 2009; IK08WW and IK09WW = Ikenne under well-watered in 2008 and 2009; BA08TD and BG09TD = Bagauda terminal drought stress in 2008 and 2009; ZA09WW = Zaria under rainfed condition in 2009.

Fig. 1. Dendrogram of 42 maize inbred lines revealed by Ward's minimum cluster analysis based on Modified Roger's distance (MRD) using SSR markers.

Fig. 2. Mean grain yields of single-cross hybrids regressed on mean grain yields of parental lines under drought and well-watered conditions.

PEDIGREE	Code
TZEI 31 × TZEI 63	1
TZEI 31 × TZEI 18	2
TZEI 24 × TZEI 16	3
TZEI 129 × TZEI 161	4
TZEI 16 × TZEI 122	5
TZEI 177 × TZEI 165	6
TZEI 26 × TZEI 63	7
TZEI 129 × TZEI 178	8
TZEI 16 × TZEI 135	9
TZEI 18 × TZEI 89	10
TZEI 89 × TZEI 31	11
TZEI 18 × TZEI 51	12
TZEI 165 × TZEI 135	13
TZEI 108 × TZEI 59	14
TZEI 26 × TZEI 18	15
TZEI 27 × TZEI 135	16
TZEI 51 × TZEI 87	17
TZEI 21 × TZEI 108	18
TZEI 112 × TZEI 87	19
TZEI 107 × TZEI 108	20
TZEI 157 × TZEI 161	21
TZEI 122 × TZEI 158	22
TZEI 161 × TZEI 158	23
TZEI 122 × TZEI 177	24
TZEI 21 × TZEI 2	25
CHECK 1 - TZE-W DT Pop STR C4	26
CHECK 4 - TZEI 26 × TZEI 5	27

Fig. 3. AMMI biplot of grain yield for 25 early maturing maize hybrids and two checks evaluated across drought stress and optimal growing conditions in Nigeria, 2008 - 2009. The vertical line is the grand mean, while the horizontal line represents IPCA1 values of zero. IK08DS and IK09DS = Ikenne under drought stress in 2008 and 2009; IK08WW and IK09WW = Ikenne under well-watered in 2008 and 2009; BA08TD and BG09TD = Bagauda terminal drought stress in 2008 and 2009; ZA09WW = Zaria under rainfed condition in 2009.