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

### M. Oyekunle<sup>a,b,\*</sup>, B. Badu-Apraku<sup>a,\*</sup>, S. Hearne<sup>a,1</sup>, J. Franco<sup>a</sup>

<sup>a</sup> International Institute of Tropical Agriculture (IITA), IITA (UK) Ltd, Carolyn House, 26 Dingwall Road, Croydon CR9 3EE, UK
<sup>b</sup> Department of Plant Science, Ahmadu Bello University, Zaria, Kaduna State, Nigeria

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#### ABSTRACT

Hybrid development is enhanced by the assessment and understanding of genetic diversity and distance within inbreds. One hundred and fifty hybrids derived from 30 early-maturing inbreds plus six checks were evaluated at three locations in Nigeria for 2 years to assess their performance under drought and well-watered conditions. In addition, twenty three microsatellite markers were employed to assess genetic diversity of selected 42 inbreds. Significant differences were observed among inbreds and hybrids for most traits under both research conditions. A total of 130 alleles were detected ranging from two for nc133 to nine for *phi299852* with an average of 5.7 alleles per locus. Polymorphic information content ranged from 0.17 for *phi308707* to 0.77 for *phi084* with an average of 0.54. Thirty-one unique alleles were detected in 21 inbreds. Microsatellite markers classified the inbred lines into five groups. Genetic 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  $F_1$  hybrids were not significant for grain yield and other traits under drought and well-watered conditions. However, significant correlations existed between  $F_1$  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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#### 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

\* Corresponding authors at: Ahmadu Bello University, Department of Plant Science, Zaria, Nigeria. Tel.: +234 8059884805.

*E-mail addresses:* moyekunle@abu.edu.ng, oyemuhyideen@yahoo.com

(M. Oyekunle), b.badu-apraku@cgiar.org (B. Badu-Apraku).

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

Drought is a major abiotic stress contributing to severe maize yield loss in the lowland savanna belt of WCA (Fajemisin et al., 1985). Edmeades et al. (1995) reported 15% annual maize yield loss from drought in the savannas of WCA and indicated that localized losses may be much higher in the marginal areas with annual rainfall below 500 mm and on sandy soils. The risk of drought stress is severe particularly in the Sudan savanna zone due to unreliable and uneven distribution of rainfall (Eckebil, 1991). Even in those





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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 × 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.

<sup>&</sup>lt;sup>1</sup> Current address: International Maize and Wheat Improvement Centre (CIMMYT), KM 45 Carretera Mexico-Veracruz, El Batan, Texcoco, Mexico.

lowlands with adequate precipitation for maize production, periodic drought may occur at the most sensitive stages of the crop such as flowering and grain filling. While drought will impact the growth and ultimate performance of a crop at any stage, it is of most detriment at flowering and grain-filling resulting in yield penalties of between 40 and 90% (Menkir and Akintunde, 2001; Badu-Apraku et al., 2011a; Badu-Apraku and Oyekunle, 2012). Therefore, improved tolerance to drought is an important breeding objective to stabilize maize production in the sub region.

Until recently, adoption of maize hybrids was insignificant in WCA due to limited seed production and marketing by existent and emerging few seed companies. However, during the last couple of years, seed companies have emerged in Ghana, Mali, and Nigeria setting the stage for large scale hybrid production in these countries (Badu-Apraku et al., 2011a). Hybrid development and promotion in maize is a promising strategy to appreciably increase maize production and productivity in WCA. Presently, within the portfolio of hybrids in WCA there are no commercially available early maturing hybrids. An important requirement for a commercially successful hybrid program is the availability of information on the heterotic patterns of inbred lines in the program. Accurate assessment of inbred lines in hybrid combinations is crucial to the success of a hybrid program and the selection of parents for the development of synthetic varieties or introgression of favorable alleles into elite source populations. Knowledge and understanding of the genetic diversity and distance of early-maturing inbred lines would be very useful in planning crosses, assigning inbred lines to specific heterotic groups and designing breeding strategies.

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

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

Assessment of genetic diversity among early-maturing inbred lines with contrasting response to drought using molecular markers and determining any association with  $F_1$  hybrid performance under drought would be invaluable in selecting parental lines for development of productive hybrids with tolerance to drought. Molecular markers have been used to assess the genetic diversity of parental lines and to determine the relationship between marker-based genetic distance among pairs of parental lines and grain yield of their hybrids in maize (Menkir et al., 2010; Makumbi et al., 2011; Akaogu et al., 2012).

Information on the performance of inbreds in hybrid combinations is crucial for hybrid development programs. It is of great importance for maize breeders to identify potential inbred lines that would produce hybrids exhibiting high levels of heterosis without making all possible crosses among inbreds available in a breeding program. Inbred line information indicative of hybrid performance is desirable to reduce costs and time associated with hybrid production and evaluation. Moll et al. (1965) and Paterniani and Lonnquist (1963) reported positive relationship between midparent heterosis (MPH) of  $F_1$  hybrid grain yield and parental genetic divergence. Makumbi et al. (2011) also reported positive association between midparent heterosis, specific combining ability, genetic distance (GD), and grain yield. Betrán et al. (2003b) observed that the differences in grain yield between hybrid and inbreds increases with the intensity of drought stress.

Information on the effect of genetic diversity of parental lines with contrasting response to drought on the performance of their hybrids is limited in the IITA maize improvement program. The objectives of the present study were to (i) assess the genetic diversity among selected early-maturing maize inbreds using SSR markers (ii) assess the relationship between  $F_1$  hybrid performance, heterosis, and SSR-based genetic distance, and (iii) assess genotype × environment interaction and identify high-yielding and stable hybrids across research environments.

#### 2. Materials and methods

#### 2.1. Genetic materials and field procedures

One hundred and fifty six early-maturing inbred lines extracted from six diverse germplasm sources (TZE-W Pop DT STR C<sub>0</sub>, WEC STR, TZE-Y Pop DT STR C<sub>0</sub>, TZE Comp 5-YC<sub>6</sub>, TZE-W Pop  $\times$  LD and TZE-W Pop  $\times$  1368 STR C<sub>0</sub>) with tolerance/resistance to Striga and maize streak virus, and/or tolerance to drought were used in this study. The procedure adopted for the development of the inbred lines has been described in detail by (Badu-Apraku et al., 2007). Two field experiments were conducted between 2007 and 2010 in Nigeria to evaluate the early-maturing inbreds and their hybrids for tolerance to drought. In addition, the genetic diversity among 42 of the 156 inbreds used for the field studies was examined using simple sequence repeat (SSR) markers. In the first experiment, the 156 early-maturing maize inbreds were evaluated under managed drought stress at Ikenne (forest-savanna transitional zone, (6°53'N, 30°42'E, 60 m asl, 1500 mm annual rainfall) in Nigeria during 2007/2008 and 2008/2009 dry seasons and in well-watered environments at Ikenne and Bagauda (Sudan savanna, 12°00'N, 8°22′E, 580 m asl, 800 mm annual rainfall) during the growing seasons of 2008 and 2009. A  $12 \times 13$  randomized incomplete block design with two replications was used for the experiment. Row length was 4 m long with 22 plants per row. Row and hill spacing were 0.75 m and 0.4 m, respectively. Three seeds were planted per hill and seedlings were thinned to two per stand about 2 weeks after emergence, giving a population density of 66 666 plants  $ha^{-1}$ . A compound fertilizer (NPK 15:15:15) was applied at the rate of  $60 \text{ kg N} \text{ ha}^{-1}$ ,  $60 \text{ kg P} \text{ ha}^{-1}$  and  $60 \text{ kg K} \text{ ha}^{-1}$  at the time of sowing at each test site. An additional  $60 \text{ kg N} \text{ ha}^{-1}$  urea was top-dressed 3 weeks later. The experiment was kept weed-free by the application of a mixture of gramaxone (Shandong Dongtai Agricultural Chemistry Co., Ltd.), a contact herbicide, and atrazine, a pre-emergence herbicide, at 51ha<sup>-1</sup> each of gramaxone and primextra (Syngenta Crop Protection Canada, Inc.). Subsequently, manual weeding was done periodically to keep the trials weed-free.

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