

Multivariate Analysis of Agronomic Traits and Disease Response of Inbred Lines of Maize (*Zea mays* L.)

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ABSTRACT

Pattern of genetic diversity and traits inter-relatedness is of great importance to germplasm collectors and plant breeders for creating variability. Thirty-two maize inbred lines were evaluated in three locations in 2014 and 2015 to establish the distinction among them based on their agronomic traits and diseases infection, as well as relatedness of the maize traits. The trial was laid out in a Randomized Complete Block Design with three replicates for two years. Data were collected on agronomic traits of the maize. Incidence of streak (STK), blight (BLT), Curvularia (CURV) and rust (RST) were scored. The grain yield (GRY) and anthesis-silking-interval (ASI) were estimated. Data collected across the environments were pooled for analysis using analysis of variance, Pearson's correlation, principal component analysis (PCA) and single linkage cluster analysis (SLCA). Effects of genotype (G), environment (E) and G × E were highly significant for all the traits. Mean GRY was 1639.7 kg ha⁻¹ while mean days to anthesis, days to silking (DTS) and ASI were 58.7, 62.6 and 3.9 days, respectively. Scores for lodging, stay green and husk cover were low. GRY associated with ASI (0.45), plant height (0.44*), ear height (0.42*), BLT (0.49*), CURV (0.38*), plant aspect (r=0.57***) and ear aspect (r=0.66***). The GRY, DTS, ASI and SG contributed mostly to variation among the lines. The high yielding lines were tolerant to major maize diseases, but BD74-31, TZEI 1, TZEI 4, BD74-55, TZEI 7, BD74-399 and BD74-170 were most suitable for hybrids development.*

Key words: Maize disease, cluster analysis, inbred lines, PCA

INTRODUCTION

Maize (*Zea mays* L.) is a popular and multipurpose cereal crop grown to provide food for man, feed for livestock and raw materials for the industries in West and Central Africa (Adetimirin *et al.*, 2008). It is a major income earner for numerous farmers, processors and other important stakeholders in maize value chain. Guinea savanna regions are the most important maize growing agro-ecologies; though it is also well adapted to the rainforest and the derived savanna agro-ecology (Iken and Amusa, 2004; Ajala *et al.*, 2007). Despite the suitability of savanna and rainforest agro-ecological zones in WCA for production of maize, yield of the crop is still low due to the effects of biotic and abiotic constraints, such as pests, diseases and stress conditions (Badu-Aparaku *et al.*, 2010). Consequently, there is the need for maize improvement for high productivity achievable through introduction of germplasms as donors of various novel alleles. However, adaptability of the introduced germplasm needs to be tested with the local ones in order to make outstanding selections among them.

Numerous multivariate statistical and numerical taxonomy techniques have been proposed to estimate genetic similarity and divergence in crop accessions (Ariyo, 1993; Flores *et al.*, 1998; Cardi, 1998). Some of such proposed techniques are coefficient of racial likeness (Pearson, 1926), principal component analysis (PCA) by Gower (1967), numerical taxonomic (Sneath and Sokal, 1973), factor and cluster analyses (Mungomery *et al.*, 1974), Metroglyph analysis and index scoring (Anderson, 1957) and Mahalanobis D₂ (Mahalanobis, 1928). Correlation analysis was proposed by Karl Pearson from a related idea introduced by Francis Galton in the 1880s (Stigler, 1989). Suitability and comparative advantages of each of the methods are properly weighed before plant breeders make a choice of which one to use.

Maize lines have been grouped with metroglyph (Saikia *et al.*, 1996; Khumkar and Singh, 2002; Showemimo and Yeye, 2005). Similarly, factor analysis has been used to identify seven factors which explained 85.1% of the variance as important for classifying the variation among

hybrid maize (Khavari *et al.*, 2011). Dutta *et al.* (2017) complemented PCA with cluster analysis to assess the genetic diversity of 84 maize genotypes and clustered the maize genotypes into two almost equal clusters based on agro-morphological traits. Cluster analysis usually complement the results of multivariate analysis. Shalini *et al.* (2003) reported that PCA alone is not adequate to represent character in terms of their relative importance when numerous characters are considered concurrently. The PCA with single linkage cluster analysis (SLCA) has been successfully used to distinguish and classify crop genotypes. Thus, there is the need to complement PCA with a compatible technique in this type of study.

Principal component analysis is a descriptive method which shows the pattern of co-variation of characters among the individual data by removing inter-correlation among variables and allows a multi-dimensional relationship to be plotted on two or three principal axes. It relies upon the eigenvector decomposition of the covariance or correlation matrix (Granati *et al.*, 2003). The technique is useful for identifying patterns in data and expressing the data by highlighting their similarities and differences. On the other hand, single linkage cluster analysis (SLCA) summarizes the position of accessions by sorting them into distinct groups.

A condition to achieve the goal of improved cultivars for release for farmers' use is to create adequate variability in which desired genes are drawn for the goal. However, information on the genetic variability in genes pools from which options are drawn for hybridization or other crop improvement programmes is also vital. Dissimilarity between pairs of a lines based on a multivariate criterion is useful to determine effects of specific traits and the contributions of each of the traits to the total variability in the germplasm. Similarly, understanding traits association aids simultaneous selections which reduces costs and save time in breeding programmes. Morphological characters have been widely utilized to distinguish and classify maize but very few information exist on the use of response of crop to diseases. Therefore, this study was conducted to establish the distinction among 32 maize inbred lines based on their agronomic traits and diseases infection, as well as relatedness of the maize traits using principal component and single linkage cluster analyses.

MATERIALS AND METHODS

The trial was conducted in three locations in different agro-ecological zones. The three locations were Kisi (03.51° E and 9.05° N 364 m asl), Ibadan (3.56° E and 7.33° N 168 m asl) and Iwo (04.18° E and 07.63° N 231 m asl). Kisi lies in Southern Guinea Savanna; Ibadan in Rainforest-Savanna-Transition and Iwo in Derived Savanna agro-ecology. Mean amount of rainfall of the locations during

the trial were 77.5 cm, 122.3 cm and 102.2 cm per annum for Kisi, Ibadan and Iwo, respectively. Various agro-ecological zones were used for this trial in two years to increase precisions in the results. Thirty-two maize inbred lines which comprises of 20 white and 12 yellow kernels (Table 1) were planted in three locations representing three agro-ecologies in 2014 and 2015. Ten of the 20 white and nine of the yellow kernel inbred lines were obtained from IITA, Ibadan while the remaining lines were obtained from International Wheat and Maize Improvement Centre (CIMMYT), Kenya.

Table 1: Description and sources of 40 maize inbred lines obtained from International Institute of Tropical Agriculture and International Maize and Wheat Improvement Centre

Entry	Name of inbred line	Source	Attribute (tolerance)	Kernel colour
1	TZEI1	IITA	Low N	White
2	TZEI2	IITA	Low N	White
3	TZEI3	IITA	Low N	White
4	TZEI4	IITA	Low N	White
5	TZEI7	IITA	Low N	White
6	TZEI22	IITA	Low N	White
7	TZEI98	IITA	Low N	White
8	TZEI106	IITA	Low N	White
9	TZEI188	IITA	Low N	White
10	TZEI136	IITA	Low N	White
11	TZEI8	IITA	Low N	Yellow
12	TZEI10	IITA	Low N	Yellow
13	TZEI11	IITA	Low N	Yellow
14	TZEI12	IITA	Low N	Yellow
15	TZEI13	IITA	Low N	Yellow
16	TZEI16	IITA	Low N	Yellow
17	TZEI124	IITA	Low N	Yellow
18	TZEI128	IITA	Low N	Yellow
19	TZEI146	IITA	Low N	Yellow
20	BD74-152	CIMMYT	Low N	White
21	BD74-147	CIMMYT	Low N	White
22	BD74-31	CIMMYT	Low N	White
23	BD74-170	CIMMYT	Low N	White
24	BD74-128	CIMMYT	Low N	White
25	BD74-171	CIMMYT	Low N	White
26	BD74-179	CIMMYT	Low N	White
27	BD74-175	CIMMYT	Low N	White
28	BD74-399	CIMMYT	Low N	White
29	BD74-55	CIMMYT	Low N	White
30	BD74-165	CIMMYT	Low N	Yellow
31	BD74-161	CIMMYT	Drought	Yellow
32	BD74-222	CIMMYT	Drought	Yellow

IITA : International Institute of Tropical Agriculture, Ibadan
CIMMYT: International Maize and Wheat Improvement Centre

Seeds of the maize were planted in two-row plots of 5 m long with a spacing of 0.75 m between rows and 0.5 m between plants in the row. Three seeds were planted and later thinned to two plants per stand at two weeks after planting (WAP) resulting to plant population density of

53,333 plants ha⁻¹. The trial was laid out in a Randomized Complete Block Design (Gomez and Gomez, 1984) with three replicates. Cultural practices were carried out according to IAR&T (2010). Data were collected on the plants as follows:

- Days to anthesis (DTA) and days to silking (DTS) as number of days from planting to 50% pollen shed and 50% silking emergence, respectively.
- Anthesis-silking-interval (ASI) was calculated as DTA minus DTS.
- Plant and ear heights were measured on five plants randomly selected per plot using metre rule from soil surface to the base of tassel and base of uppermost ear, respectively.
- Stay green (SG) was scored, three times at eight days interval during the latter part of grain filling on scale from 0 to 10, according to Bänziger *et al.* (2000), by dividing the percentage of estimated total leaf area that were green by 10. Scale 1 = 10% of leaves were green, 2 = 20 %, 3 = 30 %, 4 = 40 %, 5 = 50 %, 6 = 60 %, 7 = 70 %, 8 = 80 %, 9 = 90 % and 10 = 100 % of the leaves were green.
- Plant aspect (PASP) was the visual assessment of the plant quality. It was scored before harvesting, when the

plants were still green and ears fully developed on scale 1 to 5 where 1 = excellent and 5 = very poor before harvest, General appeal of the whole row plants, based on the relative plant and ear heights, uniformity of the plant stand, reaction of plants to diseases and insects as well as lodging was considered in the plant aspect scoring.

- Ear aspect (EASP) was also the visual assessment scoring of the ear quality. The scored was on a scale of 1 to 5 where 1=excellent; 5=very poor. The score was taken on the pile of harvested ears of each plot when spread out and the general look of the ears is taken into account. Ear size, uniformity of colour and texture, grain fill, disease and insect damage were considered for this score.
- Blight (BLT) was scored on scale 1 to 5 at three weeks after silking (WAS), where 1 = slight infection; very few lesions on leaves, usually only on the lower leaves of the plant; 5 = very heavy infection; all leaves with large number of lesions leading to premature death of the plant and light ears.

Table 2: Mean squares of agronomic and scores of disease traits from the combined analyses of variance for the maize inbred lines evaluated across three locations in two years.

Trait	Source of variation				
	Genotype (df=32)	Environment (df=5)	Genotype × environment (df=155)	Rep.(year) (df=12)	Error (df=372)
Grain yield	2166044.1***	11811808.2***	977678.2***	519133.9 ^{ns}	448404.1
DTA	51.82***	79.18***	5.85***	0.96 ^{ns}	2.36
DTS	75.79***	70.89***	6.82***	1.54 ^{ns}	2.51
ASI	9.47***	11.13***	1.37***	0.58 ^{ns}	0.56
Plant height	3320.24***	11428.62***	699.04***	298.54*	173.3
Ear height	611.84***	2193.94**	150.04***	159.42**	58.5
Root lodging	0.10**	8.05***	0.05 ^{ns}	0.09*	0.04
Stem lodging	0.11***	9.10***	0.07***	0.03 ^{ns}	0.4
Stay green	6.40***	3.68***	0.95***	0.38 ^{ns}	0.3
Husk cover	0.43***	1.14***	0.25***	0.13 ^{ns}	0.14
Plant aspect	1.00***	2.94***	0.28***	0.40**	0.16
Ear aspect	1.07***	2.15***	0.55***	0.21 ^{ns}	0.18
Streak	1.56***	23.60***	0.32**	0.36 ^{ns}	0.23
Blight	0.55***	4.72***	0.50***	0.33**	0.16
<i>Curvularia</i>	0.78***	3.07***	0.28***	0.27 ^{ns}	0.18
Rust	0.33***	2.89***	0.15***	0.07 ^{ns}	0.06

DTA, DTS and ASI mean days to anthesis, days to silking and anthesis-silking-interval, respectively
^{ns}, ***, df and Rep are non-significant, significant ($p < 0.001$), degree of freedom and replicate, respectively

- Rust (RUST) was scored on scale 1 to 5 at 3 WAS, where 1 = no rust; 5 = severe rust.
- *Curvularia* (CURV) and streak (STK) were scored on scale 1 to 5 based on the proportion of the ear leaf that is covered with lesions. 1 = slight infection; less than 10% of the ear-leaf covered by lesions while 5 = very heavy infection; 76 to 100% of the ear-leaf covered by lesions, leading to premature death of the plant and light cobs. The diseases infections were scored according to Badu-Apraku *et al.* (2012).
- Husk cover (HSK) was scored when ears are fully developed and husk leaves are drying out (3 weeks before harvest) on scale 1 to 5, where 1 = husks tightly arranged and extend beyond the ear tip; 5 = ear tips exposed.
- Root lodging (RLG) was scored on the percentage of plants that are leaning more than 45° from the upright position at two weeks to harvest, where 1 = none; 5 = all lodged.
- Stem lodging (SLG) was scored on the percentage of plant stalks broken below the ear at two weeks to harvest, where 1 = none; 5 = all lodged.
- Grain yield (GRY) adjusted to 15% moisture content was estimated as:

$$\text{GRY (kg ha}^{-1}\text{)} = \frac{\text{GWT (kg)}}{7.5 \text{ m}^2} \times \frac{(100-\text{MC})}{(100-15\%)} \times 10,000 \text{ m}^2$$

where GWT = Grain weight, MC = grain moisture content at harvest, seed moisture content at weighing = 15%, plot area = 7.5 m² and 1 ha = 10,000 m².

Analysis of variance (ANOVA) was performed on the pooled data collected for the maize inbred lines combined across six environments (3 locations × 2 years) using SAS (2009). Relationships among the traits were determined for the maize using Pearson's correlation analysis. Principal component analysis (PCA) and single linkage cluster analysis (SLCA) were utilized to determine the extent of genetic variation among the lines and the extent of contribution of factors to the variation using Paleontological Statistics software (PAST) ver. 2.03 (Hammer *et al.*, 2001).

RESULTS

Variability and means of traits of the maize inbred lines

Combined ANOVA for the data on the maize across the three locations over the two years showed that the effects of genotype, environment and G × E were highly significant for all the traits considered except G × E for RLG (Table 2). Means and coefficients of variation (CVs) for traits of the 32 maize inbred lines were shown in Table 3. The CV was highest (21.15%) for GRY and lowest

(2.9%) for DTA. The PH and EH also had high CV (greater than 10%). Six lines namely TZEI 1, TZEI 4, BD74-55, TZEI 7, BD74-399 and BD74-170 had mean yield greater than 2000 kg ha⁻¹ while BD74-31, BD74-165, TZEI 13 and TZEI 11 were prominent among the least grain yielding inbred lines (each had approximately 1000 kg ha⁻¹). Mean GRY was 1639.7 kg ha⁻¹ while the range was 1029.2 kg ha⁻¹ to 2188.6 kg ha⁻¹. The DTA ranged from 55.4 days in TZEI 8 to 61.4 days in BD74-171 while the mean DTA was 58.7 days. Inbred lines BD74-399, TZEI 1, BD74-222, BD74-31 and BD74-171 had the highest DTA. The TZEI 8, TZEI 136 and TZEI 7 were among those with least DTA. Mean DTS of the inbred lines was 62.6 days. Inbred lines TZEI 8 had the least DTS (57.6 days) while BD74-128, TZEI 4, BD74-399, BD74-55, BD74-171 and BD74-31 (approximately 65 days) ranked highest among the inbred lines with highest DTS. The ASI ranged from 2.2 days in TZEI 8 to 5.0 days in BD74-55. Inbred lines BD74-147, TZEI 22, BD74-31, TZEI 4 and TZEI 1 were similar to BD74-55 which had the highest ASI.

Top seven inbred lines in terms of PH were BD74-161, BD74-55, TZEI 128, TZEI 1, TZEI 124, BD74-170, BD74-152 and TZEI 10 (Table 3). Each of them had PH greater than 110.00 cm while a total of 16 inbred lines had PH less than 100.00 cm. Prominent among the inbred lines with least PH were TZEI 12, TZEI 11, BD74-31 and TZEI 13. The listed inbred lines with least PH also had the least EH while TZEI 98, BD74-55, BD74-222, TZEI 124, BD74-161 and TZEI 10 among other had highest EH. Table 3 also shows scores for RLG, SLG, SG, HKC, PASP and EASP of the inbred lines. Mean values for the traits were 1.0, 1.0, 3.6, 2.1, 3.9 and 3.8, respectively. Scores for diseases incidence in the maize lines shows that BD74-55 (2.7) was most infested by streak while BD74-175, BD74-179, BD74-152 and TZEI 136 were least infested, (score 1.4 each). Inbred lines BD74-175, BD74-399, TZEI 106, BD74-170, TZEI 4 and TZEI 188 had blight scores greater than 3.0 while each of BD74-179, BD74-31, TZEI 22, TZEI 8 and TZEI 128 had 2.6 blight score. However, TZEI 98, TZEI 136 and BD74-171 were conspicuous among the lines that had high scores (> 3.0) for *Curvularia* while TZEI 10, TZEI 11, TZEI 146 and TZEI 13 among other inbred lines were least infested with *Curvularia*. Line TZEI 1 top the list of the inbred lines that were mostly infested by rust while BD74-31, TZEI 3, BD74-175, BD74-128 and TZEI 136.

Multivariate analysis of agronomic traits

Table 3: Mean values of agronomic and scores of disease traits for the maize inbred lines evaluated across three locations in two years.

Inbred line	GRY (kg ha ⁻¹)	DTA (days)	DTS (days)	ASI (days)	PH (cm)	EH (cm)	RLG (1-5)	SLG (1-5)	SG (1-10)	HKC (1-5)	PASP (1-5)	EASP (1-5)	STK (1-5)	BLT (1-5)	CURV (1-5)	RST (1-5)
TZEI 106	1534.8	58.6	62.9	4.3	95.1	36.5	1.0	0.9	4.1	2.0	3.8	4.2	1.5	3.1	2.9	1.6
TZEI 2	1717.4	59.1	63.3	4.2	90.4	28.2	1.0	1.0	4.3	2.2	4.0	3.8	1.8	2.8	2.6	1.4
BD74-128	1843.9	59.8	64.6	4.8	96.0	38.9	1.1	1.0	3.4	1.9	3.9	3.5	1.7	2.7	2.9	1.3
TZEI 136	1687.9	55.8	59.9	4.1	98.3	38.6	1.0	1.1	4.7	2.2	3.7	3.8	1.4	3.0	3.1	1.3
TZEI 1	2121.8	60.8	65.6	4.7	116.2	38.4	0.9	0.9	3.3	2.0	3.7	3.6	1.5	2.9	2.8	1.8
BD74-152	1750.6	59.7	64.1	4.4	111.0	38.0	1.1	1.0	3.8	2.0	3.8	3.8	1.4	2.8	2.8	1.4
BD74-171	1431.7	61.4	65.4	4.0	100.3	36.9	1.0	1.0	3.3	2.0	3.7	4.1	1.7	2.7	3.3	1.4
TZEI 98	1950.9	57.4	61.8	4.3	101.1	40.6	0.9	0.9	4.2	2.1	3.8	4.0	1.8	2.8	3.1	1.5
TZEI 188	1997.1	57.4	61.8	4.3	96.5	39.9	1.0	1.1	3.7	2.1	4.1	3.8	1.6	3.2	3.0	1.7
TZEI 4	2188.6	59.7	64.6	4.9	97.4	35.6	0.9	0.9	3.2	2.0	3.8	3.6	2.1	3.1	2.9	1.6
BD74-55	2039.7	60.0	65.0	5.0	113.5	41.9	0.9	0.9	2.8	2.2	3.8	3.8	2.7	3.0	2.7	1.6
BD74-147	1587.7	59.9	64.4	4.5	106.8	38.3	1.0	1.1	3.6	2.1	3.7	3.8	2.1	2.8	2.9	1.4
TZEI 7	2022.0	55.9	59.9	4.0	107.3	38.0	0.9	1.0	3.4	2.1	3.5	3.8	1.9	2.8	2.9	1.6
BD74-31	1029.2	61.1	65.7	4.6	74.6	21.6	0.8	0.8	2.8	2.1	3.5	3.6	1.8	2.6	2.5	1.3
BD74-179	1589.4	59.7	63.3	3.6	91.2	31.2	0.9	0.9	2.6	2.0	4.1	3.8	1.4	2.6	2.8	1.4
TZEI 3	1775.0	57.2	61.2	3.9	105.4	38.1	1.0	1.1	3.6	2.0	4.0	3.6	1.8	2.8	2.6	1.3
TZEI 22	1629.4	57.1	61.6	4.6	101.2	36.9	0.9	0.9	4.1	2.0	3.8	3.9	1.8	2.6	2.8	1.6
BD74-170	2009.8	59.0	63.3	4.3	117.8	39.3	1.0	1.0	3.3	2.0	3.4	3.6	1.7	3.1	2.7	1.4
BD74-175	1472.1	59.7	64.0	4.3	87.1	31.1	0.9	1.0	2.8	2.0	3.9	3.7	1.4	3.1	2.7	1.3
BD744-399	2096.5	60.6	64.6	4.0	89.0	35.7	1.1	1.1	3.2	2.0	4.0	3.4	1.5	3.1	2.9	1.4
TZEI 13	1080.2	58.8	62.1	3.3	75.4	29.8	1.0	1.0	4.1	2.1	4.1	4.2	2.2	2.7	2.4	1.5
TZEI 146	1993.1	56.6	60.1	3.4	94.3	38.2	1.0	1.0	3.4	2.3	3.9	3.8	2.1	2.7	2.4	1.6
TZEI 10	1995.1	57.7	60.2	2.6	122.1	50.0	1.0	1.0	3.2	2.1	3.6	3.3	1.9	2.7	2.4	1.6
TZEI 12	1245.2	59.0	62.3	3.3	73.6	29.1	1.0	1.0	4.0	2.4	4.1	4.1	2.1	2.8	2.6	1.5
TZEI 128	1228.4	56.2	58.9	2.8	114.0	37.0	1.1	1.1	3.8	2.1	4.0	3.9	2.0	2.6	2.8	1.5
TZEI 11	1096.8	59.7	62.6	2.8	74.4	28.7	1.0	1.0	4.0	2.2	4.2	4.3	2.0	2.7	2.4	1.6
TZEI 8	1430.0	55.4	57.6	2.2	82.7	32.9	1.1	1.0	5.0	2.3	4.3	4.0	1.9	2.6	2.7	1.7
TZEI 16	1496.5	59.9	62.7	2.8	94.0	37.9	1.0	1.0	3.4	2.2	4.1	4.1	1.9	2.8	2.5	1.6
TZEI 124	1327.7	56.7	60.3	3.6	117.1	45.8	1.1	1.1	5.1	2.3	4.0	4.1	2.0	2.7	2.6	1.5
BD74-165	1034.5	59.2	62.7	3.5	106.4	39.9	1.1	1.1	3.6	2.3	4.1	3.9	1.9	2.9	2.6	1.7
BD74-161	1383.8	59.9	63.2	3.3	111.2	46.7	1.0	1.0	3.6	2.5	3.6	3.6	2.1	2.7	2.7	1.5
BD74-222	1683.9	60.9	64.3	3.4	110.3	43.7	1.0	1.0	3.3	2.5	3.4	3.4	2.2	2.8	2.6	1.6
Mean	1639.7	58.7	62.6	3.9	99.1	37.0	1.0	1.0	3.6	2.1	3.9	3.8	1.8	2.8	2.7	1.5
LSD	438.9	1.0	1.0	0.5	8.6	5.0	0.1	0.1	0.4	0.2	0.3	0.3	0.3	0.3	0.3	0.2
CV (%)	21.2	2.9	3.3	8.8	13.7	15.8	7.7	7.8	6.4	7.3	6.1	6.0	6.2	7.6	9.0	6.4

GRY, DTA, DTS, ASI, PH, EH, RLG, SLG, SG, HKC, PASP, EASP, STK, BLT, CURV and RST are grain yield, days to anthesis, days to silking, anthesis-silking-interval, plant height, ear height, root lodging, stem lodging, stay green, husk cover, plant aspect, ear aspect, streak, blight, *Curvularia* and rust, respectively.

Table 4: Correlation coefficients of agronomic and scores of disease traits of maize inbred lines evaluated across three locations in two years.

	GRY	DTA	DTS	ASI	PH	EH	RLG	SLG	SG	PASP	EASP	STK	BLT	CURV
DTA	-0.01													
DTS	0.11	0.94***												
ASI	0.45*	0.33	0.63***											
PH	0.44*	-0.11	-0.05	0.14										
EH	0.42*	-0.19	-0.19	-0.07	0.84***									
RLG	-0.18	-0.26	-0.41*	-0.55*	0.05	0.20								
SLG	-0.15	-0.34	-0.40*	-0.33	0.11	0.22	0.69***							
SG	-0.27	-0.61***	-0.62***	-0.33	-0.08	0.05	0.46**	0.35						
PASP	0.57***	-0.01	-0.09	-0.25	-0.72***	-0.66***	0.24	0.05	0.18					
EASP	0.66***	-0.13	-0.16	-0.16	-0.51**	-0.49**	-0.08	-0.11	0.42*	0.53**				
STK	-0.10	-0.03	-0.09	-0.20	0.09	0.19	-0.01	-0.09	0.00	-0.04	0.10			
BLT	-0.49*	0.09	0.25	0.48**	0.11	0.14	-0.15	0.10	-0.14	-0.27	-0.27	-0.23		
CURV	-0.38*	-0.01	0.17	0.50**	0.20	0.15	-0.26	0.03	0.03	-0.38*	-0.10	-0.43*	0.41*	
RST	0.11	-0.15	-0.24	-0.33	0.15	0.28	0.12	-0.18	0.14	0.00	0.01	0.36*	0.01	-0.21

GRY, DTA, DTS, ASI, PH, EH, RLG, SLG, SG, PASP, EASP, STK, BLT, CURV and RST are grain yield, days to anthesis, days to silking, anthesis-silking-interval, plant height, ear height, root lodging, stem lodging, stay green, plant aspect, ear aspect, streak, blight, *Curvularia* and rust, respectively.

*, ** and *** mean significant at 5%, 1% and 0.1%, respectively.

Inter-relatedness of the traits of the inbred lines

Positive and negative relationships existed among pairs of traits of the inbred lines (Table 4). The GRY had positive and significant association ($p < 0.05$) with ASI, PH and EH while negative and significant correlations existed between GY with PASP ($r = 0.57^{***}$), EASP ($r = 0.66^{***}$), BLT ($r = -0.49^*$) and CURV ($r = -0.38^*$). The DTA had significant correlation with DTS ($r = 0.94^{***}$) while DTS had significant correlations with ASI ($r = 0.63^{***}$) but negative significant correlations with SLG and SG. The DTS and ASI had negative and significant correlation with RLG. The ASI also correlated with BLT ($r = 0.48^{**}$) and CURV ($r = 0.50^{**}$). The PH had significant correlation with EH ($r = 0.84^{***}$) while PH and EH had negative correlations with PASP ($r = -0.66^{***}$) and EASP ($r = -0.49^{**}$). The RLG had positive correlations with SLG ($r = 0.69^{***}$) and SG ($r = 0.46^{**}$) while the SG had positive and significant correlation with EASP only. The PASP negatively correlated with EASP ($r = 0.53^{**}$) and CURV ($r = -0.38^*$). It was also shown that the CURV had correlation with STK ($r = -0.43^*$) and BLT ($r = 0.41^*$), while STK correlated with BLT ($r = 0.36^*$).

Classification and cluster analysis of the maize inbred lines

Principal components analysis: A total of 15 axes accounted for 100% variance for the inbred lines. The PCs I and II captured about 27.5% and 21.4%, respectively (Table 5). The first four PCs had eigenvalues greater than one and the axes accounted for about 72% of the total variance. The PCs I, II and III had eigenvalues greater than 2.00. The PC II was 0.97 different from PC I while PC III differed from PC II with eigenvalue of 1.25. Important eigenvectors loadings for the traits ranged from 0.30 to 0.55 across the first four axes. The GRY, DTS, ASI and SG loaded the PC I; PH, EH and PASP loaded PC II while SLG, HSK, EASP, BLT and CURV loaded PC III and DTA, RLG, SLG and RST loaded PC IV.

Cluster analysis: The dendrogram achieved through SLCA on scale of 144 units (Figure 1) illustrates the distinction and relatedness of the 32 maize inbred lines. Up to unit 8, all the inbred lines were distinct from one another. Only lines TZEI 188 and TZEI 146 were most related below 16 distant level while all the lines clustered into two at the highest distance level (136). The midpoint (72) had four clusters while at lesser distant level (16), the maize lines clustered into 29. It was found that inbred lines BD74-31 and TZEI 4 were most distant and distinct. Four of the 32 maize inbred lines clustered into one (cluster I) while 16, 2 and 10 inbred lines formed cluster II, III and IV, respectively.

Mean GRY of the maize lines was about 1060 kg ha⁻¹ for Group I; 1584 kg ha⁻¹ for Group II; 1237 kg ha⁻¹ for Group III and 2042 kg ha⁻¹ for Group IV (Table 6). The DTA

ranged from 57.6 days for Group III to 59.7 days for Group I while the DTS ranged from 60.6 days for Group III to 63.2 days for Group I. Moreover, the range for the ASI was from 3.1 days for Group III to 4.1 days for Group IV. Lowest mean PH and EH were observed for Group I and highest for Group IV. Mean scores for RLG were similar to those of SLG for Group I, III and IV than for groups II where RLG score was higher than SLG. Group II also had highest incidence of lodging. The scores for PASP and EASP were highest for Group I and least for Group IV.

Highest STK score was observed for Group III (2.06) and least for Group III (1.74). Mean scores of BLT were similar for all the groups while mean scores for CURV was least in Group I, moderate in Group III but similar in Groups II and IV. For RST, mean scores were similar for Group I, III and IV but was lower for Group II than other groups.

DISCUSSION

Information of the genetics and relatedness of traits helps to plan and effectively execute breeding programme for developing improved genotypes of crops. The low CVs for all the traits considered in this study suggest genotypic uniformity within the inbred lines and high level of precision in the experiment. But, the relatively high CV for the GRY may be connected with the polygenic nature of the traits. Grain yield is governed by additive gene actions which results from contributions of several minute genes. Hence, couples of analyses may be required to discriminate and distinguish the inbred lines based on the GY. Since GY is a major crop trait, it is necessary to discriminate and classify the maize inbred lines for successful selection for further improvement. Significant variation in genotypes for all the traits of the maize inbred lines suggests that adequate variability is present among the inbred lines to form efficient gene pool from which germplasm can be drawn for hybridization. The variability may be expected because the two sets of germplasms were from different climatic regions; IITA materials were of tropical adaptation while CIMMYT inbred lines were of temperate region. Adebayo (2012) and Aminu *et al.* (2014) also observed wide variability among wide based genetic maize materials such as those from IITA and CIMMYT.

Inbred lines TZEI 1, TZEI 4, BD74-55, TZEI 7, BD74-399 and BD74-170 had yield greater than grand mean GRY. Selection can be made among these promising inbred lines for high grain yielding hybrids development in Nigeria, especially the rainforest and savanna agro-ecologies where the evaluation was conducted. The BD74-399, TZEI 1, BD74-222, BD74-31 and BD74-171 that had either high or average grain yield were included among those had the highest DTA, DTS and ASI. Therefore, flowering may not necessarily be a determinant for high yield of the maize inbred lines. In the same vein, high

Multivariate analysis of agronomic traits

Table 5: Eigen vectors loading, eigenvalues and percent variance for traits of maize inbred lines evaluated across three locations in two years as decomposed into first four principal component axes

Trait	Eigenvectors			
	PC I	PC II	PC III	PC IV
Grain yield	0.3166	0.2621	-0.0411	-0.1337
Days to anthesis	0.2327	-0.2642	0.2908	0.3786
Days to silking	0.3284	-0.2603	0.2005	0.2554
Anthesis-silking-interval	0.3827	-0.1159	-0.1043	-0.1521
Plant height	0.1678	0.419	0.1076	0.026
Ear height	0.0831	0.4745	0.1263	0.0407
Root lodging	-0.2711	0.1995	-0.1566	0.4548
Stem lodging	-0.1756	0.2185	-0.3208	0.4497
Stay green	-0.3013	0.1354	-0.2732	-0.2453
Husk cover	-0.2535	0.1499	0.3548	0.0402
Plant aspect	-0.2664	-0.3419	-0.0935	0.0769
Ear aspect	-0.1341	0.0862	0.4776	-0.1351
Streak	0.257	0.0863	-0.1951	-0.0216
Blight	0.2608	0.0718	-0.3645	-0.2041
<i>Curvularia</i>	-0.1159	0.1803	0.3052	-0.298
Rust	-0.2457	-0.2863	-0.0791	-0.3497
Eigenvalue	4.4	3.43	2.18	1.42
Difference	0.97	1.25	0.75	0.44
Percent variance (%)	27.51	21.44	13.64	8.87
Cumulative percent variance (%)	27.51	48.95	62.59	71.47

PC is principal component

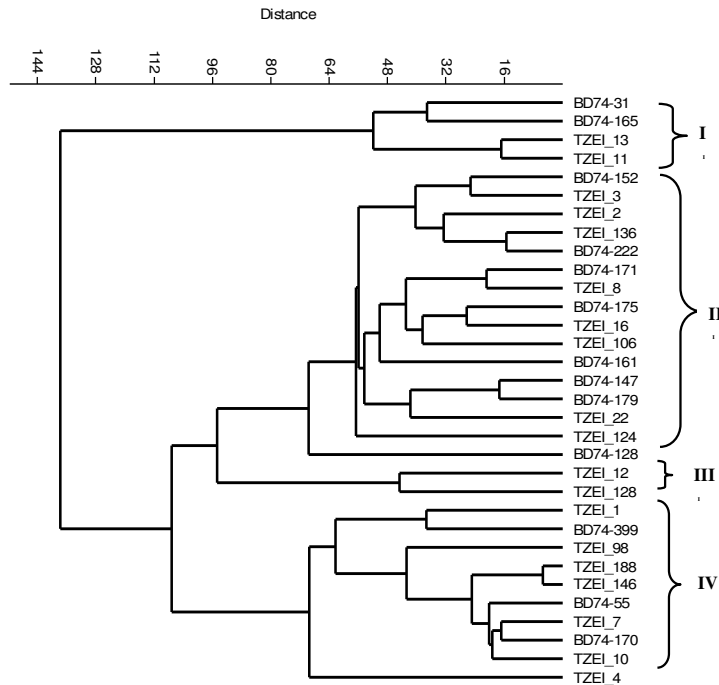


Figure 1: Dendrogram showing genetic relatedness and classification of 32 maize inbred lines based on evaluation of the lines across three locations in two years.

Multivariate analysis of agronomic traits

Table 6: Means and ranges of agronomic and scores of disease parameters of the maize inbred lines according to groups lines based on evaluation of the lines across three locations in two years

Parameter	GRY (kg ha ⁻¹)	DTA (days)	DTS (days)	ASI (days)	PH (cm)	EH (cm)	RLG (1-5)	SLG (1-5)	SG (1-5)	HKC (1-5)	PASP (1-5)	EASP (1-5)	STK (1-5)	BLT (1-5)	CURV (1-5)	RST (1-5)
Group I (n = 4)																
Mean	1060.18	59.68	63.24	3.56	82.72	30.01	0.98	0.97	3.61	2.16	4.22	4.12	1.95	2.72	2.48	1.52
Maximum	1096.80	61.06	65.67	4.61	106.39	39.86	1.05	1.05	4.07	2.31	4.39	4.25	2.19	2.89	2.61	1.69
Minimum	1029.22	58.78	62.06	2.83	74.43	21.64	0.82	0.82	2.77	2.06	4.11	3.86	1.79	2.58	2.36	1.27
Group II (n = 16)																
Mean	1583.84	58.82	62.69	3.88	99.87	37.47	0.99	1.02	3.78	2.13	3.87	3.83	1.74	2.78	2.77	1.44
Maximum	1843.85	61.44	65.44	4.78	117.07	46.68	1.11	1.14	5.05	2.53	4.28	4.17	2.19	3.11	3.25	1.67
Minimum	1327.65	55.44	57.61	2.17	82.73	28.18	0.87	0.88	2.59	1.86	3.44	3.42	1.36	2.56	2.47	1.27
Group III (n = 2)																
Mean	1236.78	57.61	60.64	3.06	93.83	33.06	1.06	1.05	3.93	2.25	4.06	3.97	2.06	2.72	2.65	1.53
Maximum	1245.18	59.00	62.33	3.33	114.02	37.03	1.10	1.10	4.03	2.36	4.14	4.06	2.11	2.81	2.75	1.53
Minimum	1228.37	56.22	58.94	2.78	73.64	29.08	1.02	1.00	3.83	2.14	3.97	3.89	2.00	2.64	2.56	1.53
Group IV (n = 10)																
Mean	2041.45	58.52	62.68	4.14	105.51	39.80	0.96	0.96	3.38	2.08	3.76	3.68	1.88	2.94	2.78	1.57
Maximum	2041.45	58.52	62.68	4.14	105.51	39.80	0.96	0.96	3.38	2.08	3.76	3.68	1.88	2.94	2.78	1.57
Minimum	1950.87	55.94	59.94	2.56	89.01	35.61	0.85	0.85	2.78	1.97	3.44	3.33	1.50	2.69	2.44	1.37

GRY, DTA, DTS, ASI, PH, EH, RLG, SLG, SG, HKC, PASP, EASP, STK, BLT, CURV and RST are grain yield, days to anthesis, days to silking, anthesis-silking-interval, plant height, ear height, root lodging, stem lodging, stay green, husk cover, plant aspect, ear aspect, streak, blight, *Curvularia* and rust, respectively.

yielding BD74-55, TZEI 1, and BD74-170 had high PH and EH. This height attribute with high yield further proves the suitability of the inbred lines for hybridization. This also buttressed the significant correlation among GY, PH and EH. Mean scores for RLG and SLG were low, suggesting the inbred lines were relatively sturdy and could withstand effect of wind during growth. However, the lines with lowest lodging incidence could be good candidates for maize improvement.

The HKC scores that was generally low shows that hybrids that will result from the parent lines are likely to have ears which husks would be tightly arranged and extend beyond the ear tip. This is an attribute which protects maize ears from pests' infestation (Fayenuwo *et al.*, 2007). Mean stay green of the lines was high because the inbred lines were bred for low nitrogen tolerance. This suggests that the lines belong to same quality class. However, none of the inbred lines had low scores for plant aspect and ear aspect. This may be because the maize evaluated was inbred lines. Inbred lines may not be appealing to sight as the improved varieties. The implication of the result is that lodging, HKC or SG cannot be a reliable selection of trait for improvement of the crop. Only 10, representing 27.8% of the total inbred lines had streak infection greater than the grand mean. It can be deduced that the maize lines were relatively tolerant to the disease. The high yielding inbred lines had relatively moderate blight, *Curvularia* and rust scores.

Positive and negative relationships existed among pairs of traits of the inbred lines. This implies that traits can be selected for or against to improve grain yield of maize. For instance, the ASI, PH, EH can be considered directly in place of GRY while the PASP, EASP, blight and *Curvularia* can be selected against when improving the crop for GRY. By implication high ASI, PH, EH are expected to result in high GRY while blight and *Curvularia* infection diminished GRY. Similarly, high aspects are expected to support GY. It has been shown earlier in this study that positive associations may exist between pairs of GRY, DTA, DTS or ASI because some high yielding maize share certain similarity with the respect to these traits. Ojo *et al.* (2006) and Ogunniyan and Olakojo (2014) had reported that pairs of days to anthesis and silking, ear height and weights can be improved simultaneously.

In this study, though the discriminating power of PC I was strongest, but could not adequately distinguish the maize inbred lines. The PC I accounted for less than one third (27.5%) variation. Even the PC I with the PC II explained less than 50% of the variation. It is therefore, required that other PCs be employed to distinguish the maize inbred lines. The first three PCs are more important in discriminating the inbred lines for having eigenvalue greater than 2.0. However, the first four PCs accounted for about 71.5% of the total variance among the traits that contributed to the variations among the lines. The percent

variance (71.5%) explained is adequate to infer that genetic variability exist among the maize inbred lines for the traits. It had been found that PC I alone may not adequately explain the variability among traits of crops (Ariyo, 1993; Ogunniyan, 2016 and Dutta *et al.*, 2017).

Traits associated with the axes could effectively distinguish the lines because of various high loadings of the traits. The GRY, DTS, ASI and SG loaded the PC I, thereby facilitating the assumption that the traits contributed most significantly to the variation among the inbred lines. The PC I accounted for GRY, two of the flowering traits and SG. These traits have been reported as effective N stress tolerance indices (Bänziger *et al.*, 2000). It has also been found through PC analysis that yield traits and flowering traits are mostly responsible for variation among genotypes of maize (Khavari *et al.*, 2011; Faruq *et al.*, 2013; Halidu *et al.*, 2015). Therefore, any of the traits can be used more efficiently in discriminating the inbred lines than other. Only SLG loaded two PCs (III and IV), while the remaining traits loaded a PC each. It also signifies rich genetic variability regarding the contributing ability of the traits of the inbred lines.

Plant height, EH and PASP loaded PC II which was equally important as PC I suggesting the traits can be used effectively to compliment GRY, DTS, ASI and SG that are most effective selection indices in this study. Though variation existed among the inbred lines for HSK, EASP, BLT, CURV, DTA, RLG, SLG and RST, there are limitation to their utilization due to their eigenvectors. The PCs I, II and III accounted for GY, DTS, ASI, PH, EH, SG, HSK, PASP and EASP as well as BLT and CURV infection. Hence, the traits are very important in selection of the inbred lines. This, however, supported the relationship of GRY with ASI, PH, EH, PASP, EASP, BLT and CURV. Contributions of traits of the inbred lines based on the eigenvectors have been used to estimate genetic variability among the accessions (Granati *et al.*, 2003; Nwangburuka *et al.*, 2011; Denton and Nwangburuka, 2012; Ogunniyan, 2016). All the authors in their various study also found that yield and flowering traits contributed most to the variations among accessions of the respective test crops.

Based on the totality of the contributions of each attribute by PCA and discrimination of the maize inbred lines by SLCA on dendrogram, inbred lines BD74-31 and TZEI 4 which are most distinct and distant are expected to possess variant attributes useful in hybrids development if further examined. Tolerance of the two inbred lines to diseases was also contrasting connoting variability in the resistance or tolerance of each inbred line to diseases infection. However, they performed similarly with respect to flowering, heights, lodging, stay green, aspects and husk cover. This confirmed that many traits should be used for selection of BD74-31 and TZEI 4 and other inbred lines that

associated with each of the two inbred lines can be harnessed for hybridization to improve the crop. For instance, BD74-31 fell in cluster I and TZEI 4 fell in cluster IV. Similarly, those in Group II are more related to those in Group I while those in Group IV are more related to those in Group III, hence all the inbred lines in same group with two inbred lines should possess distinct attributes which can be deployed for the crop improvement. Crosses can be generated among the inbred lines based on this classification. Mean values for GRY, BLT, CURV and RST did not correspond with the grouping of the two most distant inbred lines. The remaining 12 traits might have influenced expression and subsumed the contributions of the four traits. This also supports the earlier report that the cluster analysis should complemented. The PCA showed that GRY, DTS, ASI and SG were most important traits to discriminate the inbred lines. Mean GRY which was also found to be at the extremes for the Group I and Group IV confirms that the GRY is vital in maize accessions discrimination.

CONCLUSION

Inbred lines BD74-31, TZEI 1, TZEI 4, BD74-55, TZEI 7, BD74-399 and BD74-170 are most suitable for hybrid development in savannah and forest agro-ecological zones. The inbred lines are high grain yielding and had average to high tolerance to the diseases infection. The grain yield, days to silking, anthesis-silking-interval and stay green contributed most significantly to the variation among the inbred lines. The grain yield positively correlated with anthesis-silking-interval, plant height, ear height, plant aspect and ear aspect but negatively correlated with blight and *Curvularia*. Based on the contributions of each attribute by PCA and discrimination of the maize inbred lines by SLCA on dendrogram, inbred lines BD74-31 and TZEI 4 are most distinct and distant especially in grain yield and diseases infection.

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