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Combining ability and heterotic grouping of early maturing provitamin A maize inbreds across *Striga* infested and optimal growing environments

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Abstract: The development, deployment and production of stress tolerant provitamin A maize is crucial to the fight against vitamin A deficiency in sub-Saharan Africa (SSA) where maize is a major staple food crop. 17 early maturing provitamin A and normal yellow endosperm maize inbreds were crossed using the diallel mating design to generate 136 single-cross provitamin A hybrids. The hybrids were evaluated with 4 hybrids checks during 2015 growing season at three locations under two *Striga* infested and three optimal growing environments in Nigeria. The objectives were to (i) determine the combining ability and heterotic groups of early maturing provitamin A inbreds; (ii) examine the performance of the inbreds in hybrid combinations across environments and (iii) identify early maturing provitamin A inbred testers for use in tropical maize breeding programs. The general combining ability (GCA) and specific combining ability (SCA) effects were significant for grain yield and most other traits indicating that additive and non-additive genetic effects were important in the inheritance of these characters. Inbreds TZEI 10, TZEIOR 108, and TZEI 17 had significant positive GCA for grain yield. TZEIOR 108, TZEIOR 42, and TZEI 10 had significant negative GCA for *Striga* damage while TZEIOR 122, TZEIOR 127, TZEIOR 108, and TZEI 10 had significant negative GCA for number of emerged *Striga* plants. The inbreds were classified based on heterotic groups' SCA and GCA of grain yield (HSGCA), SCA and GCA of multiple traits (HGCAMT) methods into four, three and five heterotic groups, respectively. The inbreds TZEIOR 108, TZEI 10 and TZEI 17 were identified as testers. Hybrids TZEIOR 57 x TZEIOR 108, TZEIOR 57 x TZEIOR 127, TZEIOR 13 x TZEIOR 59, TZEIOR 57 x TZEI 10 and TZEIOR 127 x TZEI 10 were identified as high yielding and most stable across environments.

Keywords: Combining ability, Striga *, maize*

Introduction

Food security and malnutrition are major challenges facing rural populations in sub-Saharan Africa (SSA). Vitamin A deficiency is an important health problem affecting more than 43 million children under 5 years of age in the sub-region (Menkir *et al.,* 2013; Pillay *et al.,* 2013). Besides the provitamin A activity, carotenoids are involved in essential biological functions in human health (Mishra and Singh, 2010). According to Menkir *et al.* (2013), maize plant naturally accumulates carotenoids with provitamin A activity, making the crop a suitable dietary source of this nutrient. The strategy of breeding maize with high carotenoid concentrations offers a sustainable alternative to direct vitamin supplementation. However, maize production which has the greatest potential in the SSA is seriously constrained by *Striga hermonthica* Del. Benth (Badu-Apraku *et al.,* 2013). *Striga* infestation is often so severe that there could be total crop yield loss and farmers may be compelled to abandon their fields. Badu-Apraku *et al.* (2011) and Akinwale *et al.* (2014) showed that under severe *Striga* infestation, grain yield may be reduced by more than 40%. Breeding maize for *Striga* resistant hybrids and open-pollinated varieties is therefore the most economic and sustainable strategy for controlling the yield losses caused by *Striga*.

Many countries in sub-Saharan Africa have adopted hybrid maize varieties and several high-yielding early and extra-early maturing hybrids are available to Companies for commercial seed production (Badu-Apraku *et al.* 2013). However, there is no commercial early maturing provitamin A hybrids available in the sub-region. In an effort to enhance the diet quality, a program was initiated in 2007 by IITA Maize Improvement Program to develop early and extra-early maturing stress tolerant (drought tolerant and *Striga* resistant), high provitamin A and/or quality protein maize (QPM) varieties for West and Central Africa. Presently many early inbred lines with high level of provitamin A are available in the IITA maize program. The usefulness of the inbred lines in breeding program depends on the information of their combining ability and the heterotic groups (Hallauer and Miranda, 1988). However there is no early maturing provitamin A inbred tester available in IITA Maize Improvement Program. The lack of knowledge about these informations on early maturing provitamin A maize inbred lines may limit their use in hybrid development (Menkir *et al.*, 2013). Combining ability has been used in plant breeding to compare the performances of the lines in hybrid combinations (Griffing, 1956). Diallel mating models developed by Griffing (1956) are the major models used in combining ability analysis (Fan *et al.,* 2008) to study the inheritance of important traits and to identify appropriate testers for use in a breeding program (Badu-Apraku *et al.*, 2011). The value of hybrid is largely dependent on its yield performance and stability across test environments. Menkir *et al.* (2013) reported maize hybrid that combines high provitamin A levels with high yield potential across environments may likely be adopted in SSA, because it has the advantage of requiring no change in the current consumer behavior. The objectives of this study were to (i) determine the combining ability and heterotic groups of early maturing provitamin A inbreds, (ii) examine the performance of the provitamin A inbred lines in hybrid combinations across environments and (iii) identify best testers for use in the breeding program.

Materials and Methods

Genetic materials

One hundred and thirty six single cross hybrids derived from a diallel cross of seventeen early maturing provitamin A and yellow inbred lines were used in this study. The seventeen inbred lines involved in the single cross hybrids are listed in Table 1. The provitamin A inbred lines are designated TZEIOR while yellow lines are designated TZEI.

Field layout and experimental evaluation

In the first study, the single cross hybrids with four hybrids checks were evaluated for *Striga* resistance under *Striga* infested environment at Mokwa (9°18’ N, 5°4’ E, 457 m above the sea level (asl) and 1100 mm annual rainfall) and Abuja (9°16’ N, 7°20’ E, 300 m asl and 1500 mm annual rainfall) during the 2015 growing season. Before planting, ethylene gas was injected into the soil to stimulate suicidal germination of existing *Striga* seeds. *Striga* seeds collected from previous season were mixed with finely sieved sand and used in each hill for infestation in the ratio of 1:99. In the second study, the single cross hybrids with the checks were evaluated at Ikenne (6°54’ N, 3°7’ E, 60 m asl and 1200 mm annual rainfall), Abuja and Mokwa under optimal growing conditions during the 2015 growing season. The optimal trial is a non-stress condition and allows assessing the real potential of the genotypes.

In all the studies, 14 x 10 randomized incomplete block design with two replications, single row plot, and 3 m long and spaced of 0.75 m between rows and 0.40 m between plants within the row were used. Three seeds were planted per hill and thinned to two per stand about 2 weeks after emergence to give a final population density of 66 000 plants/ha.

Fertilizer was applied 30 days after planting, when 30 kg ha^{-1} each of N, P, and K was applied as 15-15-15 NPK on *Striga* infested trials at Abuja and Mokwa. Additional 10-20 kg N ha^{-1} fertilizer was applied depending on the growth condition of the crops. Weeds other than *Striga* were controlled manually. The optimal growing conditions were *Striga*-free environments and compound fertilizer (NPK 15-15-15) was applied on the trials at Ikenne, Mokwa and Abuja at the rate of 60 kg N, P and K per ha with additional 30 kg N ha^{1} as top dressing four weeks after planting.

Table 1 - Description of the 17 inbred lines involved in the diallel crosses and hybrids checks used in this study.

Inbred name	Pedigree information	Grain color	Source
TZEIOR 9	2009 TZE OR1 DT STR S_6 inb 10-1/1-1/2-1/1-1/3-1/1	Orange	IITA
TZEIOR 11	2009 TZE OR1 DT STR S_6 inb 10-1/1-2/2-1/3-3/4-1/1	Orange	IITA
TZEIOR 12	2009 TZE OR1 DT STR S_6 inb 10-1/1-2/2-2/3-1/3-1/1	Orange	IITA
TZEIOR 13	2009 TZE OR1 DT STR S6 inb 10-1/1-2/2-3/3-1/2-1/1	Orange	IITA
TZEIOR 57	2009 TZE OR1 DT STR S_6 inb 38-2/3-1/3-1/3-2/2-1/1	Orange	IITA
TZEIOR 60	2009 TZE OR1 DT STR S_6 inb 38-2/3-1/3-2/3-3/3-1/1	Orange	IITA
TZEIOR 122	2009 TZE OR1 DT STR S_6 inb 77-2/3-2/2-2/2-1/2-1/1	Orange	IITA
TZEIOR 127	2009 TZE OR1 DT STR S_6 inb 77-3/3-2/2-2/2-1/3-1/1	Orange	IITA
TZEIOR 56	2009 TZE OR1 DT STR S_6 inb 38-2/3-1/3-1/3-1/2-1/1	Orange	IITA
TZEIOR 59	2009 TZE OR1 DT STR S_6 inb 38-2/3-1/3-2/3-2/3-1/1	Orange	IITA
TZEIOR 108	2009 TZE OR1 DT STR S_6 inb 66-1/1-1/2-1/2-1/2-1/1	Orange	IITA
TZEIOR 118	2009 TZE OR1 DT STR S_6 inb 77-2/3-1/2-1/3-1/2-1/1	Orange	IITA
TZEIOR 42	2009 TZE OR1 DT STR S_6 inb 22-2/2-2/2-2/3-1/3-1/1	Orange	IITA
TZEIOR 58	2009 TZE OR1 DT STR S_6 inb 38-2/3-1/3-2/3-1/3-1/1	Orange	IITA
TZEIOR 24	2009 TZE OR1 DT STR S_6 inb 12-2/2-2/2-3/3-1/3-1/1	Orange	IITA
TZEI 17	TZE Comp5-Y C6 S6 inbred 35	Yellow	IITA
TZEI 10	TZE-Y Pop STRC0 S6inbred 152	Yellow	IITA
Check1	TZEI 124 x TZEI 25	Yellow	IITA
Check2	TZEI 24 x TZEI 17	Yellow	IITA
Check3	TZEI 11 x TZEI 23	Yellow	IITA
Check4	TZE-Y Pop DTSTR x TZEI 13	Yellow	IITA

Data collection

Observations for the two studies were made for days to 50% anthesis and silking, as the number of days from planting to when 50% of the plants had shed pollen and had emerged silks. The anthesis-silking interval (ASI) was calculated as the difference between days to 50% silking and days to 50% anthesis. The plant height was measured as the distance from the base of the plant to the height of the first tassel branch and ear height as the distance to the node bearing the upper ear. The percent root lodging was determined as the percentage of plants leaning more than 30° from the vertical.

Plant aspect, ear aspect and husk cover were recorded respectively on a scale of 1 to 9, where 1= excellent plant type, clean ear type, husk tightly arranged and extended beyond the ear tip, 9=undesirable plant, ear tips exposed.

Ears harvested per plot were weighted (field weight). Grain yield estimated based on 80% shelling percentage and adjusted to 15% moisture was calculated as follow:

$$Grain\ yield = field\ weight\ x\, \frac{(100 - m)}{85}\ x\, \frac{10000}{\phi}\ x\ 0.8$$

Where m= grain moisture at harvest,
10000= land area per hectare (m^2)
ϕ= land area per plot (m^2)

In *Striga* experiments, the host plant damage syndrome at 8 and 10 weeks after planting (WAP) were scored per plot on a scale of 1 to 9, where 1=no damage and 9= complete collapse or death of the plant while emerged *Striga* plants count were taken at 8 and 10 weeks after planting (WAP) and the data was later transformed using LOG_{10} (X+1). Ear per plant was calculated as number of ear harvested over number of plant harvested.

Statistical analysis

Separate analyses of variance (ANOVA) were performed on data collected across locations for each research condition (*Striga* infested and *Striga* free) with General Linear Model procedure (PROC GLM) of Statistical Analysis System (SAS) using RANDOM statement with the TEST option. Afterwards, combined ANOVA across all research conditions were performed across five environments for grain yield, days to silking (DYSK), anthesis-silking interval (ASI), ear per plant (EPP), plant height (PLHT), ear aspect (EASP); three environments for plant aspect (PASP) and two environments for *Striga* damage rating and number of emerged *Striga* plants at 8 and 10 weeks after planting (WAP). In the combined ANOVA, environments, replicates and block were considered as random factors while entries were considered as fixed effects. Means were separated using the LSD. The mixed model procedure of SAS was used (SAS Institute, 2002). The statistical model used for combined analysis is as follows:

$$Y_{ijkg}= u+E_i+R_{j(i)}+B_{k(ij)}+G_g+EG_{ig}+ \varepsilon_{ijkg}$$

Where Y_{ijkg} is the observed measurement for the g^{th} genotype grown in the environment i, in the block k, in replicate j; u is the grand mean; E_i is the main effect of environment; $R_{j(i)}$ is the effect of replicate nested within environment; $B_{k(ij)}$ is the effect of block nested within replicate j by environment i; G_g is the effect of genotypes (hybrids and checks); EG_{ig} is the interaction effect between genotype and environment, and ε_{ijkg} the error term.

Effects of the GCA of the parents and SCA of the crosses, as well as their mean squares in each environment and across environments, were estimated on 17 x 17 diallel crosses excluding the checks, following the procedure described by Griffing (1956) for diallel analyses method 4 model 1 (fixed model) using DIALLEL SAS program developed by Zhang *et al.,* (2005) adapted to SAS software version 9.3. The statistical model used for the combined diallel analysis across environments is as follows:

$$Y_{ijk} = u + E_e + g_i + g_j + s_{ij} + gE_{eg} + sE_{es} + \varepsilon_{ijk}$$

Where Y_{ijk} is the observed measurement for the ijth cross grown in the kth environment; u is the grand mean; g_i and g_j are the GCA effects; s_{ij} is the SCA effects; gE_{eg} is the interaction effect between GCA and the environment; sE_{es} is the interaction effect between SCA and the environment, and ε_{ijk} is the error term associated with the ijth cross evaluated in the kth replication and E_e environment (Hallauer and Miranda, 1988; Badu-Apraku *et al.,* 2011). The inbreds lines were assigned into heterotic groups using heterotic group's specific and general combining ability (HSGCA) and heterotic grouping based on general combining ability of multiple traits (HGCAMT) methods. In heterotic grouping by HGCAMT method, GCA of grain yield and traits, that had significant mean square across test environments were considered whereas in HSGCA method, GCA for grain yield of the lines and SCA for grain yield of their crosses were used. The two grouping methods were subjected to Ward's minimum variance cluster analysis based on the Euclidean distances generated from the different method of GCA estimation using SAS software version 9.3.

The genotype effects and genotype by environment interaction (GGE) biplot analysis was performed to obtain information on the performance and yield stability of the hybrids across the environments. The program is available at www.ggebiplot.com. The GGE biplot model equation was:

$$Y_{ij} - \beta_j = \alpha_1 \acute{E}_{i1} n_{j1} + \alpha_2 \acute{E}_{i2} n_{j2} + E_{ij}$$

Where Y_{ij} is the genetic value of the combination (pure line or hybrid) between entry i and tester j for the trait of interest; β_j is the mean of all combinations involving tester j; α_1 et α_2 are the singular values for principal component (PC) 1 and PC2; $\acute{E}_{i1}$ and $\acute{E}_{i2}$ are the PC1 and PC2 eigenvectors for entry i; n_{j1} and n_{j2} are the PC1 and PC2 eigenvectors for tester j; E_{ij} is the residual of the model associated with the combinations of entry i and tester j.

To identify productive hybrids under *Striga* infested environments for commercial use, a selection index was computed using standardized data for grain yield, EPP, *Striga* damage rating and *Striga* emergence counts at 8 and 10 WAP. In order to select superior genotype under *Striga*-infested conditions, specifications were made for independent selection rate, independent culling rate, and overall cutting rate on the traits (YIELD, EPP, STRAT8, STRAT10, STRC8, STRC10) used by IITA maize improvement program in the base index for selecting *Striga* tolerant genotype. Weight +2 was assigned to YIELD, +1 was assigned respectively to EPP, STRAT8, and STRAT10; while +0.5 was assigned to STRC8 and STRC10. The means of selected traits were expressed in standard deviation units and the base index scores computed as I= [(2 xYIELD) + EPP - (STRAT8 + STRAT10) – 0, 5 (STRC8 + STRC10)], where YIELD was the yield of *Striga* infested plots, EPP as number of ears per plant, STRAT8 and STRAT10 were *Striga* damage ratings at 8 and 10 WAP, STRC8 and STRC10 were number of emerged *Striga* plants at 8 and 10 WAP.

Results

Combining ability estimated of provitamin A early maturing inbreds across test environments

The combined analysis of variance across test environments of the diallel crosses revealed significant mean squares for environments (E), entries (G) and entry x environment interaction (GEI) for all the measured traits, except for entry for *Striga* emergence count 8 and 10 weeks after planting (WAP), and for GEI for *Striga* damage rating 8 and 10 WAP and *Striga* emergence count 8 WAP (Table 2). Moreover the general combining ability (GCA) and specific combining ability (SCA) effects were significant for grain yield and most others agronomic traits. In diallel experiment with fixed model, the ratio of 2GCA mean square over 2GCA mean square plus SCA mean square was used to express the relative importance of additive and non-additive gene effects for controlling characters (Fan *et al.*, 2008). In this study, the GCA effects were greater than SCA effects for all traits under *Striga* infested, optimal and across test environments (Fig.1). Significant means squares of GCA x Environment were detected for all the traits except *Striga* damage at 8 and 10 WAP whereas SCA x Environment mean squares were significant for grain yield, DYSK, ASI, EPP, plant height and EASP.

Significant positive GCA effects for grain yield and ear per plant were observed for the inbreds TZEI 10, TZEI 17, and TZEIOR 108 whereas inbreds TZEIOR 42, TZEIOR 108, and TZEI 10 had significant negative GCA for *Striga* damage (Table2). Similarly, inbreds TZEIOR 122, TZEIOR 127, TZEIOR 108, and TZEI 10 had significant negative GCA for number of emerged *Striga* plants while TZEIOR 60, TZEIOR 56, TZEIOR 58, and TZEIOR 59 had significant positive GCA effects respectively for *Striga* damage, EASP, PASP, husk cover (HUSK) and stem lodging (SL). It should be noted that high positive GCA effects of any genotype for these traits is undesirable.

Table 2 - Mean squares from the combined analysis of variance of grain yield and other agronomic traits (days to silking, anthesis-silking interval, ear aspect, ear per plant and plant height) across five environments, Striga rating at 8 and 10 weeks after planting (WAP) and Striga count at 8 and 10 WAP across two environments of early maturing provitamin A hybrids.

Source of variation	DF	Grain Yield (Kg/ha)	Days to silking	Anthesis-silking-interval	Plant height (cm)	Ears per plant	Ear aspect	DF	*Striga* rate at 8WAP	*Striga* rate at 10WAP	*Striga* count at 8WAP	*Striga* count at 10WAP
Environnent (ENV)	4	920847262**	2927,42**	180,71**	54231**	0,91**	38,21**	1	1,39**	0,56**	4,46**	5,45**
Entry	135	10445396**	29,18**	1,58**	600*	0,04**	3,64**	135	0,04**	0,04**	0,69	0,54
GCA	16	16814399**	55,04**	3,91**	1077*	0,08**	13,06**	16	0,14**	0,16**	1,15**	0,96**
SCA	119	9589060**	25,71**	1,27	536	0,03**	2,38**	119	0,03**	0,03	0,63	0,48**
ENV x Entry	540	1312974**	3,31**	0,98**	481**	0,02**	0,57**	135	0,02	0,02	0,61	0,44**
GCA x ENV	64	1672329**	5,92**	2,34**	600**	0,02*	0,89**	16	0,02	0,02	1,02*	0,88**
SCA x ENV	476	1264658**	2,96**	1,17*	465**	0,02**	0,52*	119	0,02	0,02	0,56	0,38
ERROR	675	831979	2,30	0,74	338,81	0,02	0,45	270	0,64	0,85	0,52	0,31

*Significantly different at 0.05 level of probability, ** significantly different at 0.01 level of probability, GCA=General combining ability, SCA=specific combining ability

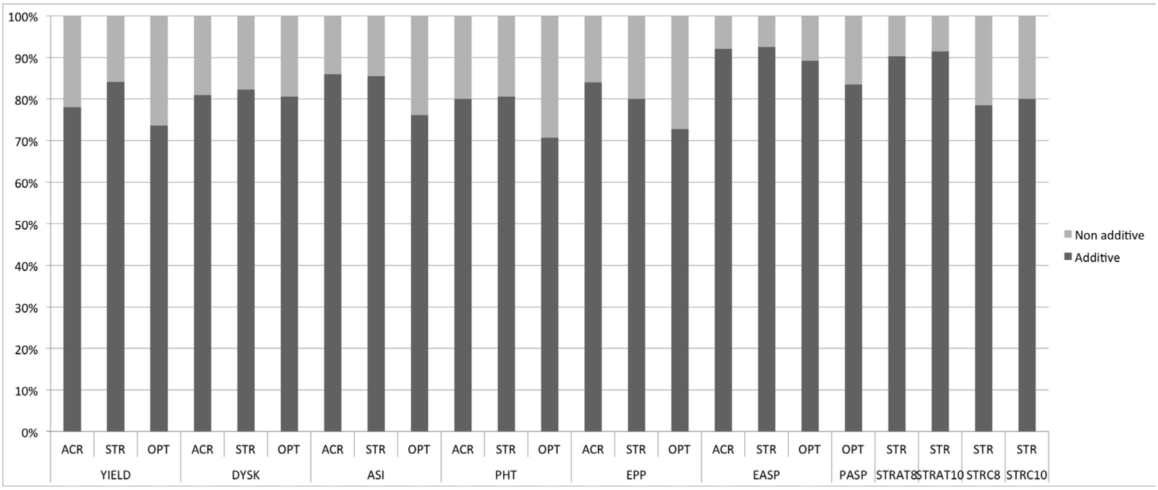

Fig. 1 - Proportion of additive (lower bar) and non-additive (upper bar) genetic variance for grain yield, day to silking (DYSK), anthesis-silking interval (ASI), plant height (PHT), ear per plant (EPP), ear aspect (EASP), plant aspect (PASP), Striga *damage rating at 8 and 10 weeks (STRAT8, STRAT10) and* Striga *emerged count at 8 and 10 weeks after planting (STRC8 and STRC10) under* Striga *infested, optimal growing conditions and across environments. ACR= across environments, STR=* Striga *environments and OPT= optimal growing conditions.*

However significant negative GCA effects were found for TZEIOR 127, TZEIOR 108, TZEIOR 24, TZEI 10, and TZEI 17 and for TZEIOR 9, TZEIOR 127, TZEIOR 108, and TZEI 10 for EASP, PASP respectively whereas inbreds TZEIOR 108, TZEIOR 42, TZEI 10 and TZEI 17 had significant positive GCA effects for EPP. Five crosses, TZEIOR 57 x TZEIOR 11, TZEIOR 12 x TZEIOR 60, TZEIOR 127 x TZEIOR 57, TZEIOR 127 x TZEIOR 60 and TZEIOR 13 x TZEIOR 59 had significant positive SCA effects for grain yield (table not shown).

Heterotic grouping of provitamin A early maturing inbreds across test environments

The grouping of the 17 inbreds using dendrogram (Fig. 2) constructed based on the HSGCA classified them into four heterotic groups across test environments. The group 1 consists of TZEIOR 9, TZEIOR 12, TZEI 17, TZEIOR 11, and TZEIOR 13; group 2 includes TZEIOR 122, TZEIOR 118, TZEIOR 108, and TZEIOR 127; the inbreds TZEIOR 42, TZEIOR 24 and TZEI 10 belong to the group 3 while the group 4 comprises TZEIOR 57, TZEIOR 56, TZEIOR 58, TZEIOR 59 and TZEIOR 60. The heterotic grouping based on GCA of multiple traits (HGCAMT) classified the inbreds into five groups (Fig. 3), while SCA method classified into three groups. The latter method could not classify TZEIOR 42, TZEIOR 24, TZEI 10 and TZEI 17 in any of the groups. The summary of the heterotic groups identified by different heterotic grouping methods is shown in Table 3.

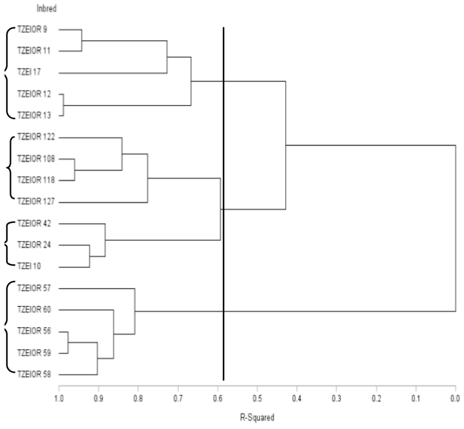

Fig. 2 - Cluster analysis (ward's minimum variance) of 17 early maturing provitamin A and yellow inbred lines based on the grain yield of hybrids in diallel study using HSGCA across environments. The thick line demarcates the groups.

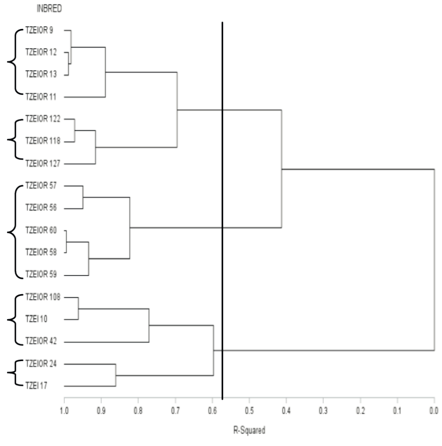

Fig. 3 - Cluster analysis (ward's minimum variance) of 17 early maturing provitamin A and yellow inbred lines based on the grain yield of hybrids in diallel study using HGCAMT across environments. The thick line demarcates the groups.

Table 3 - General combining ability (GCA) effects of grain yield and other traits of early maturing inbred lines evaluated across five environments in Nigeria, 2015.

Inbred	YIELD	DYTS	DYSK	ASI	EASP	PASP	EPP	PLHT	EHT	HUSK	STRAT 8	STRAT 10	STRC8	STRC10
TZEIOR 9	151,17	0,04	0,17	0,08	0,04	-0,30**	-0,02	-0,82	-1,1	-0,11	-0,12	-0,13	2,48	2,44
TZEIOR 11	-174,17	0,1	0,36	0,26**	0,25**	0,12	-0,05**	-2,72	-1,18	0,17	0,13	0,1	0,67	0,76
TZEIOR 12	51	-0,12	0,04	0,17	0,07	-0,05	-0,02	-1,16	-0,49	0,07	0,15	0,07	4,70**	2,96
TZEIOR 13	56,67	-0,34	-0,25	0,1	0,13*	-0,02	-0,02	-0,16	-0,63	0,08	-0,05	-0,17	-1,42	-1,58
TZEIOR 57	-144,94	1,01**	0,88**	-0,13	0,15*	0,25**	0,01	0,57	0,69	0,14	-0,07	-0,03	-0,7	0,16
TZEIOR 60	-313,73	0,54*	0,47	-0,07	0,25**	0,40**	-0,01	0,02	-0,79	0,24*	0,31**	0,45**	3,78*	3,09
TZEIOR 122	-152,52	-0,84**	-0,70*	0,16	0,07	-0,04	0	1,65	0,68	-0,08	0,46**	0,40**	-2,75	-5,26**
TZEIOR 127	-170,42	-0,67**	-0,58*	0,06	-0,15*	-0,14*	0,01	-0,46	-0,69	-0,12	0,05	0,12	-4,87**	-5,04**
TZEIOR 56	-286,78	1,10**	1,11**	-0,02	0,23**	0,29**	-0,02	1,54	1,23	0,25*	0,18	0,30*	-2,08	0,77
TZEIOR 58	-385,44*	0,58*	0,38	-0,13	0,33**	0,32**	-0,02	-2,68	-0,55	0,25*	0,28*	0,35**	-0,52	1,06
TZEIOR 59	-323,51*	0,39	0,39	0,05	0,36**	0,44**	0	-4,82*	-2,85*	0,23*	0,18	0,25*	1,35	2,91
TZEIOR 108	496,09**	-0,4	-0,59*	-0,17	-0,43**	-0,57**	0,03*	4,38*	3,45**	-0,51**	-0,55**	-0,52**	-3,03	-3,68*
TZEIOR 118	-294,67	-1,06**	-0,86**	0,16	0,16*	0,07	-0,01	2,27	-2,07	0,02	0,1	0,25*	2,68	0,84
TZEIOR 42	15,98	-0,45	-0,83**	-0,39**	-0,09	-0,08	0,025*	2,14	1,36	-0,14	-0,47**	-0,53**	2,68	4,71*
TZEIOR 24	156,71	0,60**	0,53	-0,05	-0,29**	0,05	0,02	-0,87	-2,23	0,22*	-0,25*	-0,1	-0,68	2,22
TZEI 10	671,19**	-0,57*	-0,49	0,06	-0,45**	-0,57**	0,04**	4,97*	4,31**	-0,49**	-0,30**	-0,47**	-2,75	-4,61*
TZEI 17	647,37**	0,08	-0,06	-0,12	-0,62**	-0,17	0,031**	-3,87*	0,86	-0,22*	0,01	-0,35	0,45	-1,73
SE	159,95	0,23	0,28	0,09	0,06	0,06	0,01	1,95	1,23	0,1	0,11	0,12	1,7	1,86

*Significant at 0.05 level of probability. **significant at 0.01 level of probability. SE=standard error; DYTS=days to 50% pollen shed; DYSK=days to 50% silking; ASI= anthesis-silking interval; PASP= plant aspect; EASP=ear aspect; EPP= ear per plant; PLHT= plant height; EHT=ear height; HUSK=husk cover; STRAT8= *Striga* damage at 8 weeks after planting (WAP); STRAT10=*Striga* damage at 10WAP; STRC8=*Striga* emergence count at 8 WAP; STRC10=*Striga* emergence count at 10WAP.

Table 4 - Summary of heterotic groups of 17 early maturing provitamin A and yellow inbreds identified by different heterotic grouping methods across Striga *infestation and optimal conditions*

Methods	Group				
	1	2	3	4	5
HSGCA	TZEIOR 9, TZEIOR 11, TZEIOR 12, TZEIOR 13, TZEI 17	TZEIOR 122, TZEIOR 127, TZEIOR 108, TZEIOR 118	TZEIOR 42, TZEIOR 24, TZEI 10	TZEIOR 60, TZEIOR 57, TZEIOR 56, TZEIOR 58, TZEIOR 59	
SCA	TZEIOR 9, TZEIOR 11, TZEIOR 12, TZEIOR 13	TZEIOR 60, TZEIOR 57, TZEIOR 56, TZEIOR 58, TZEIOR 59	TZEIOR 122, TZEIOR 127, TZEIOR 108, TZEIOR 118		
HGCAMT	TZEIOR 9, TZEIOR 12, TZEIOR 11, TZEIOR 13	TZEIOR 122, TZEIOR 118, TZEIOR 127	TZEIOR 60, TZEIOR 57, TZEIOR 56, TZEIOR 58, TZEIOR 59	TZEIOR 108, TZEIOR 42, TZEI 10	TZEIOR 24, TZEI 17

Performance of the single-cross hybrids and yield stability across test environments

Under *Striga* infestation the yield range of the top 10 single-cross provitamin A hybrids varied from 3132 kg ha^{-1} to 4302 kg ha^{-1} per hectare while the yield of the commercial checks varied from 2195 kg ha^{-1} to 2844 kg ha^{-1} (table4). About all of the top 10 single-cross provitamin A hybrids out-yielded the high-yielding commercial hybrid TZEI 11 x TZEI 23. However under optimal condition, the yield range per hectare of the single-cross early provitamin A hybrids was 5078 kg ha^{-1} to 6060 kg ha^{-1} whereas it was 4137 kg ha^{-1} to 6147 kg ha^{-1} for the commercial checks. In addition the top 10 single-cross hybrids achieved 50% silking between 53 to 57 days while the checks reached 50% days silking between 57 to 58 days under *Striga* infested conditions. In contrast, under optimal conditions the day to 50% was between 51 to 53 days for the single-cross whereas it has been between 51 to 55 days for the checks.

The best *Striga* tolerant single-cross provitamin A hybrid TZEIOR 127 x TZEI 10 showed a grain yield reduction of 18.6% under *Striga* infestation while the yield reduction was 31% for the best *Striga* tolerant commercial check TZEI 11 x TZEI 23.

The significant Genotype x Environment Interaction for grain yield prompted the use of GGE biplot analysis to identify high yielding and most stable hybrids among the top fifteen best *Striga* tolerant provitamin A hybrids plus the top 10 susceptible and the four commercial checks across multiple environments. In the GGE biplot (Fig. 4), the single arrowed line is the average environment coordination (AEC) axis and points in the direction of higher mean grain yield while the double arrowed line represents the stability of the genotypes; either direction away from the biplot origin on this axis indicates less stability.

The ideal genotypes are those with longest vector from the biplot origin and closer to the AEC axis. Based on these, the hybrids TZEIOR 57 x TZEIOR 108, TZEIOR 57 x TZEIOR 127, TZEIOR 13 x TZEIOR 59, TZEIOR 57 x TZEI 10 and TZEIOR 127 x TZEI 10 were identified as the highest-yielding and most stable genotypes in this order across environments.

Table 5 - Grain yield and other traits of hybrids (the best 10 and the worst 10 based on base index and grain yield) and checks evaluated under Striga and optimal growing conditions.

Hybrid	Grain yield (kg/ha)		Days to silk		Anthesis-silking interval		Ear aspect (1-9)		Ear per plant		Plant aspect (1-9)	Striga damage rate (1-9)		Striga emergence count		Base index
	STR	OPT	STR	OPT	STR	OPT	STR	OPT	STR	OPT	OPT	8WAP	10WAP	8WAP	10WAP	
TZEIOR 127 x TZEI 10	4302	5286	53	52	1.0	0.3	4.3	5	1	1	5	3.5	4.3	10	12	12.83
TZEIOR 57 x TZEI 10	3816	5732	53	51	0.5	0.3	4.8	4.8	0.7	1	4.8	4	4.3	14	20	6.82
TZEIOR 12 x TZEI 10	3535	5427	56	53	2.0	1.2	5.3	4.7	0.9	1	4.7	4.5	4.8	23	29	5.18
TZEIOR 13 x TZEIOR 59	3476	5824	57	51	1.8	0.0	5.5	5.5	1	1	5.5	3.8	4.5	30	40	6.62
TZEIOR 59 x TZEI 17	3452	5439	56	52	1.5	1.0	5	5	0.8	0.9	5.5	4.3	4.8	16	20	5.63
TZEIOR 118 x TZEI 17	3379	5498	55	52	1.0	0.5	4.8	4.5	1	0.9	5	4.3	4.5	22	26	6.89
TZEIOR 57 x TZEIOR 127	3277	6060	55	51	0.8	0.2	5.8	4.8	0.9	1	4.7	4	4.8	7	16	7.73
TZEIOR 13 x TZEIOR 58	3224	5078	54	53	0.5	0.3	5.8	5.3	0.9	0.9	5.3	4.3	4.8	14	14	6.3
TZEIOR 42 x TZEI 10	3197	5481	56	52	1.3	0.3	5.3	4.8	1	1	4.7	3.5	3.8	11	21	9.78
TZEIOR 9 x TZEIOR 108	3132	5837	56	52	1.3	0.3	5	4.5	0.9	1.1	4	4	4.5	17	31	6.01
TZEIOR 9 x TZEIOR 11	838	2178	61	53	3.3	0.5	6.3	6.7	0.7	0.7	6	5.5	5.8	13	26	-7.11
TZEIOR 57 x TZEIOR 59	827	1697	62	57	1.8	1.3	6.5	7	0.9	0.9	6.7	5.5	6.3	15	21	-5.14
TZEIOR 60 x TZEIOR 56	779	1928	62	56	3.3	0.6	6.8	6.8	0.8	0.9	6.8	5.8	6.3	9	22	-6.07
TZEIOR 57 x TZEIOR 58	751	1619	63	58	1.3	0.6	6.8	6.8	0.8	0.9	6.8	6	6.3	12	19	-6.74
TZEIOR 122 x TZEIOR 127	729	1959	60	53	3.5	1.8	6.5	6.5	0.8	0.9	6.5	5.8	6.5	5	9	-5.81
TZEIOR 57 x TZEIOR 56	720	1958	63	57	2.0	0.8	6.5	6.7	0.7	1	6.8	5.5	6.5	10	25	-8.06
TZEIOR 11 x TZEIOR 12	620	1472	62	58	2.3	1.2	7	7	0.6	0.8	6.7	5.8	6.3	28	18	-9.82
TZEIOR 60 x TZEIOR 59	579	1282	61	56	2.3	1.2	6.8	6.3	0.8	0.9	7	5.8	7	30	32	-9.95
TZEIOR 60 x TZEIOR 58	522	1591	60	56	0.8	0.6	7	6.8	0.7	0.9	7	6	7	21	28	-11.2
TZEIOR 58 x TZEIOR 59	395	1176	60	57	1.5	1.0	7.3	7	0.7	0.8	7.5	6.3	6.8	15	26	-10.6
Check 3-(TZEI 11 x TZEI 23)	2844	4137	57	53	1.5	0.5	4.5	5	0.9	0.9	5.3	4.3	4.5	10	13	5.67
Check 4- (TZE-Y Pop DTSTR x TZEI 13)	2741	4566	58	55	1.8	1.5	5.3	5.5	0.8	0.9	5.3	4.8	5.3	23	37	0.2
Check 1- (TZEI 124 x TZEI 25)	2425	6147	57	52	1.8	1.0	5.8	4.8	0.8	1	4.2	4.8	5.8	12	10	0.44
Check 2- (TZEI 24 x TZEI 17)	2195	5015	57	51	1.5	0.3	4.8	5.2	1	1	5.3	4.3	4.8	11	10	4.76
Mean	2151	4818	57	52	1.9	0.3	5.7	5.3	0.9	1.0	5.4	1.7	1.9	3	3	
LSD	1267	1047	2	2	2	1	1.0	1.0	0.2	0.1	0.6	1.1	1.3	18	20	
Genotype	**	**	**	**	NS	**	**	**	NS	**	**	**	**	NS	*	
Environment	**	**	**	**	NS	*	**	**	*	**	**	**	**	**	**	
Genotype x Environment	NS	*	**	**	*	**	NS	**	NS	**	NS	NS	NS	NS	**	

STR=*Striga* infested environments, OPT= Optimal growing conditions, 8WAP= eight weeks after planting, 10WAP= ten weeks after planting.

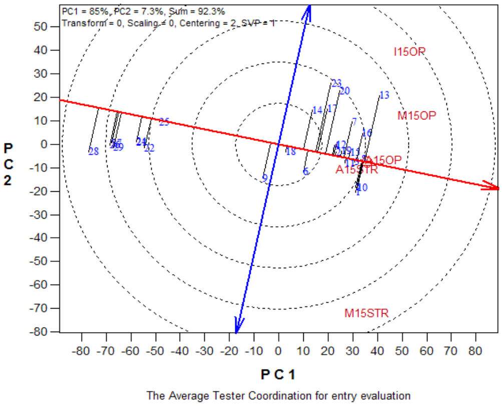

Fig. 4 - The "mean vs. stability" view of genotype x environment interaction of 25 early maturing provitamin A hybrids and four normal checks evaluated under Striga *infested and optimal growing conditions. I15OP= Ikenne optimal 2015; M15OP =Mokwa optimal 2015; A15OP= Abuja optimal 2015; A15STR= Abuja* Striga *infestation 2015 and M15STR= Mokwa* Striga *infestation 2015.*

Code	Genotype
1	TZEIOR 127 x TZEI 10
2	TZEIOR 42 x TZEI 10
3	TZEIOR 57 x TZEIOR 127
4	TZEIOR 42 x TZEI 17
5	TZEIOR 13 x TZEIOR 59
6	TZEIOR 13 x TZEIOR 58
7	TZEIOR 9 x TZEIOR 24
8	TZEIOR 57 x TZEIOR 108
9	Check 3- (TZEI 11 x TZEI 23)
10	TZEIOR 57 x TZEI 10
11	TZEIOR 118 x TZEI 17
12	TZEIOR 122 x TZEI 17
13	TZEIOR 108 x TZEI 17
14	Check 2- (TZEI 24 x TZEI 17)
15	TZEIOR 108 x TZEI 10
16	TZEIOR 9 x TZEIOR 108
17	TZEIOR 108 x TZEIOR 42
18	Check 4-(TZE-Y Pop DTSTR x TZEI 13)
19	Check 1- (TZEI 124 x TZEI 25)
20	TZEIOR 12 x TZEIOR 122
21	TZEIOR 9 x TZEIOR 11
22	TZEIOR 56 x TZEIOR 59
23	TZEIOR 11 x TZEIOR 122
24	TZEIOR 57 x TZEIOR 56
25	TZEIOR 57 x TZEIOR 60
26	TZEIOR 60 x TZEIOR 59
27	TZEIOR 11 x TZEIOR 12
28	TZEIOR 58 x TZEIOR 59

Discussions

The significant mean squares detected for Environments (E), Entries (G) and Entries x Environments (GEI) for most of the measured traits (Table 1), indicate that there is adequate genetic variability among the early maturing provitamin A hybrids to allow good progress from selection for the measured traits under the research environments (Badu-Apraku and Oyekunle, 2012). However the lack of significant GEI for *Striga* damage ratings at 8 and 10 WAP indicates that the expression of these traits could be consistent in different test environments. These results are consistent with the findings of Badu-Apraku *et al.* (2013); Badu-Apraku *et al.* (2011). The general combining ability (GCA) and specific combining ability (SCA) effects were significant for grain yield and most others agronomic traits indicating that both the additive and non-additive genetic effects were important in the inheritance of these characters. The closer the ratio of 2GCA/2GCA+SCA to unity, the greater the predictability based on GCA alone (Badu-Apraku *et al.*, 2013). In this study, GCA effects were greater than SCA effects for all traits across environments (Fig.1) indicating that the additive gene effects were more important for controlling these traits. The significant GCA x Environment mean squares for all measured traits indicates that the combining ability of the lines varies significantly under different environments. This suggests the need for multi-environments testing to identify high yielding and most stable *Striga* resistant hybrids. In contrast, the lack of significant SCA x Environment mean squares for *Striga* damage rating and *Striga* emergence count at 8 and 10 WAP shows that these traits were expressed consistently over different environments.

Fan *et al.* (2008) studied combining ability using crosses between CIMMYT germplasms and Chinese local germplasms and no crosses showed significant positive SCA effects among local lines. In general, parental inbred lines of related origin consistently produced lower yielding crosses; however there were some exceptions (Hallauer and Miranda, 1988). In this study, all the crosses with significant positive SCA effects involved crosses between the early provitamin A lines. Despite the fact that the early provitamin A inbred lines were developed from the same source populations, they were classified into different heterotic groups. This result suggests that there exists broad genetic diversity among the early maturing provitamin A inbred lines. Similar results were reported by Agbaje *et al.* (2008) in which the inbred lines derived from the same source populations were classified into different heterotic groups, confirming the broad genetic diversity within the source populations. Furthermore the source populations of the early maturing provitamin A inbred lines were developed from mixtures of different genetic composition (Agbaje *et al.*, 2008) by introgression followed by a cycle of backcrossing, selfing and recombination to form a population. These different breeding methods employed contributed in enhanced genetic diversity (Carena and Wicks III, 2006) of the source populations from which the early maturing provitamin A inbred lines were extracted.

The grouping of the inbred lines by HSGCA method was closely related to their pedigree data. This result corroborates the findings of Badu-Apraku *et al.* (2016) in which all IITA lines were classified by HSGCA method according their parentage. The HSGCA method appeared to be the most efficient for classifying this set of provitamin A inbred lines. It identified groups that allow the inter-heterotic group crosses to produce more superior hybrids than HGCAMT method. The latter method placed the inbreds with significant GCA effects for grain yield into the same group. These results are in agreement with the findings of Badu-Apraku *et al.* (2013) who reported that HGCAMT method tend to put most of the inbred lines with significant GCA effects for grain yield into the same group. The inbreds TZEIOR 108, TZEI 10 and TZEI 17 displayed significant positive GCA effects for grain yield indicating that these lines could be good combiners for improving this trait. In this study selection of potential testers was based on significant and positive GCA effects for grain yield of the inbreds, the ability to classify lines into heterotic groups and per se grain yield. Based on these criteria the inbreds TZEIOR 108, TZEI 10 and TZEI 17 were identified as potential testers. Akinwale *et al.* (2014) reported that the inbred which had consistently and negative SCA effects in cross combinations with other inbreds within the same group could be considered as the best tester for that group. Therefore the inbred lines TZEIOR 108, TZEI 10 and TZEI 17 could be the best testers for the groups 2, 3 and 1 respectively. However none of the lines in group 4 had significant positive GCA effects for grain yield and no tester could be identified for this group.

Significant negative GCA effects for *Striga* damage rating or *Striga* emergence count indicate tolerance or resistance to *Striga hermonthica*. Based on this, TZEIOR 122 and TZEIOR 127 were considered as *Striga* resistant, while TZEIOR 42 was classified as tolerant. Similarly TZEIOR 108 and TZEI 10 showed both tolerance and resistance to *Striga*. The percentage yield reduction of 55% in this study was comparable to the 64% reported by Akinwale *et al.* (2014), and suggested that the *Striga* infestation was severe enough to allow the identification of superior resistant genotypes. Therefore, the best single-cross early provitamin A hybrid TZEIOR 127 x TZEI 10 showed yield reduction of 18.6% while it has been 31% for the best *Striga* resistant commercial check hybrid TZEI 11 x TZEI 23. However it should be noted that high positive GCA effects for any genotype for *Striga* damage rating, EASP, PASP, husk cover (HUSK) and stem lodging (SL) will be considered as undesirable. Therefore TZEIOR 60, TZEIOR 56, TZEIOR 58, and TZEIOR 59 with significant positive GCA effects for these traits were not only susceptible to *Striga* and root lodging, but contributed also unfavorably to ear aspect, plant aspect and husk cover.

The performance and yield stability of the genotype are quite crucial in multi-environment test. According to Yüksel *et al.* (2006), an ideal genotype may not exist in reality, but it can be used as a reference to identify the most desirable genotypes. The 'mean performance vs. stability' view of the GGE Biplot ranked the genotypes relative to the ideal genotype (center of the concentric circle). Based on these results the following hybrids, TZEIOR 57 x TZEIOR 108, TZEIOR 57 x TZEIOR 127, TZEIOR 13 x TZEIOR 59, TZEIOR 57 x TZEI 10 and TZEIOR 127 x TZEI 10 were identified as the highest-yielding and most stable across environments and should be extensively tested to confirm the consistency of performance and commercialized in SSA. Furthermore most of the best performing and most stable hybrids involved at least one parent that is resistant to *Striga*. Therefore it should be noted that the resistance to *Striga* has played important role in conferring stability of performance of the provitamin A hybrids across test environments.

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