

## Article

# Performance and Stability Analysis of Extra-Early Maturing Orange Maize Hybrids under Drought Stress and Well-Watered Conditions

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**Abstract:** The consistently low yield turnout of maize on farmers' fields owing to drought and the nutritional challenges attributable to the consumption of white endosperm maize pose a major threat to food and nutritional security in Sub-Saharan Africa (SSA). The objectives of this study were to assess the performance of newly developed extra-early maturing orange hybrids under managed drought and well-watered conditions, compare the outcomes of multiple-trait base index and multi-trait genotype–ideotype distance index selection procedures, and identify drought-tolerant hybrids with stable performance across contrasting environments for commercialization in SSA. One hundred and ninety orange hybrids and six checks were evaluated under managed drought and well-watered conditions at Ikenne for two seasons between 2021 and 2023. A 14 × 14-lattice design was used for the field evaluations under both research conditions. Drought stress was achieved by the complete withdrawal of irrigation water 25 days after planting. Results revealed significant differences among the hybrids under drought and well-watered conditions. Grain yield, ears per plant, and plant aspect under managed drought were correlated to the same traits under well-watered conditions, suggesting that the expression of these traits is governed by common genetic factors. Twenty-nine hybrids were identified as top-performing drought-tolerant hybrids by the multiple-trait base index and the multi-trait genotype–ideotype distance index. Of the selected outstanding 29 hybrids, 34% were derived from crosses involving the tester TZEEIOR 197, demonstrating the outstanding genetic potential of this inbred line. Further analysis of the 29 selected hybrids revealed TZEEIOR 509 × TZEEIOR 197 as the hybrid that combined the most drought-tolerant adaptive traits. However, the hybrids TZEEIOR 526 × TZEEIOR 97, TZEEIOR 384 × TZEEIOR 30, TZEEIOR 515 × TZEEIOR 249, TZEEIOR 510 × TZEEIOR 197, TZEEIOR 479 × TZEEIOR 197, and TZEEIOR 458 × TZEEIOR 197 were identified as the most stable hybrids across drought and well-watered conditions. These hybrids should be extensively tested in multi-location trials for deployment and commercialization in SSA.

**Keywords:** drought tolerance; multiple-trait base index; multi-trait genotype–ideotype distance index; stability; sub-Saharan Africa



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## 1. Introduction

Maize is a major crop for human consumption and as animal feed in sub-Saharan Africa (SSA). The crop has the potential to combat the food insecurity and malnutrition challenges presently facing the region. It currently provides 30% of dietary calories for

millions of people in SSA [1]. Maize is cultivated on approximately 42.5 million hectares in Africa [2]. Most of the varieties cultivated by farmers are deficient in essential micronutrients, particularly vitamin A [3,4], which cannot be synthesized by the human body. The essentiality of this nutrient is demonstrated by the vulnerability of people who depend largely on maize for food to several diseases such as river blindness and diarrhea [5]. Vitamin A deficiency (VAD) is also known to impair the functionality of the immune system, increase susceptibility to diseases, and cause night or complete blindness and death from severe illnesses [6]. Given its importance as a major staple cereal crop in African homes, it is imperative to develop and commercialize improved maize genotypes biofortified with essential nutrients such as provitamin A (PVA) with tolerance/resistance to major production constraints faced by SSA farmers. These constraints include drought, low soil nitrogen (low N), *Striga hermonthica* parasitism, fall armyworm infestation, and many diseases [7–9]. On farmers' fields, these constraints usually occur jointly with devastating effects on maize grain yield. This, coupled with the low genetic potentials of varieties cultivated by most farmers has resulted in the low grain yields frequently observed in farmers' field.

In the absence of other stress factors during the production cycle, drought is capable of causing grain yield reduction between 40 and 90% depending on the stage of plant growth and development when the drought occurs, and its severity [10–13]. The flowering (tassel and silk emergence) and post-flowering (grain filling) stages are the major determinants of photo-assimilate partitioning from the source to the sink and consequently the amount of grain yields obtained at harvest. These stages are widely considered as the most sensitive stages of maize development which, if affected by drought, can lead to considerable reduction in maize production and productivity [14–17]. To address the dietary needs of the populace in SSA and achieve increased maize productivity in the sub-region, it is of utmost importance to develop new and improved maize hybrids that combine enhanced levels of PVA and high levels of tolerance to drought for commercialization. Based on the findings from some studies high PVA contents are correlated to the orange coloration of the kernel endosperm [18,19]. Therefore, orange maize could be a source of PVA content to address malnutrition. The first step to the realization of drought-tolerant PVA-rich maize began in 2007 by the International Institute of Tropical Agriculture Maize Improvement Program (IITA-MIP). The program had as its aim the development of extra-early and early (80–85 and 90–95 days to maturity, respectively) varieties that combine high levels of drought tolerance and provitamin A for SSA farmers. Through this program, numerous extra-early inbred lines possessing drought tolerance genes and elevated levels of PVA were developed, commercialized, and are presently serving as invaluable germplasm resource for developing drought-tolerant PVA-rich hybrids in several national and international breeding programs [20]. In SSA, hybrid development and commercialization are the major focus of many breeding programs owing to the superior performance of hybrids over open pollinated varieties (OPVs). When new hybrids are developed, it is important to understand their responses to drought in multiple locations to identify tolerant hybrids for commercialization. To achieve this goal, a good selection strategy is required.

Plant breeders usually develop new genotypes with superior performance by combining several desirable traits in these new genotypes [21]. The main difficulties that frequently prevent breeders from selecting optimal genotypes using this approach are a definition of the economic value of such traits and converting these into realistic economic weights [22]. These have been the major drawbacks of the commonly used the multiple-trait base index (MBI) method. To overcome these limitations, a multivariate selection index, the multi-trait genotype–ideotype distance index (MGIDI) has been developed [21]. This index takes multicollinearity into account and identifies all variables that should be taken into account to lead to significant genetic gains from selection [21]. Using this selection approach will enable maize breeders to identify genotypes that combine high yield potential with valuable characters conditioning drought tolerance to mitigate the effects of drought on maize production in SSA. This study was conducted to (i) assess the performance of newly developed multiple stress-tolerant extra-early orange hybrids under managed drought and

well-watered research conditions, (ii) compare the selection outcomes resulting from the use of the MBI and the MGIDI, and (iii) identify outstanding drought-tolerant and stable hybrids across drought stress and well-watered environments.

## 2. Materials and Methods

### 2.1. Genetic Material

In an effort by the IITA Maize Improvement Program (MIP) to develop multiple-stress-tolerant and/or -resistant, extra-early maturing cultivars with elevated levels of PVA for farmers in SSA, the extra-early *Striga*-resistant cultivar, 2004 TZEE-Y STR C4 was crossed to Syn-Y-STR-34-1-1-1-2-1-B-B-B-B/NC354/SYN-Y-STR-34-1-1-1 (OR1) that possess high levels of PVA, in 2007. This was aimed at incorporating genes conditioning high  $\beta$ -carotene into 2004 TZEE-Y STR C4. The resulting F<sub>1</sub> from this cross was taken through a generation of backcrossing with 2004 TZEE-Y STR C4 to retrieve the recurrent parent genome (2004 TZEE-Y STR C4). From the resulting BC<sub>1</sub> generation, kernels with a deep orange color were targeted for selection and advanced to F<sub>2</sub> and F<sub>3</sub> through inbreeding. At F<sub>3</sub>, the lines characterized by intense orange coloration were selected for recombination to form the extra-early PVA-rich cultivar 2009 TZEE-OR1 STR. This broad-based cultivar was evaluated for under artificial *Striga* infestation and managed drought since 2010 and has shown outstanding performance. The cultivar represent the first extra-early PVA maize germplasm from where the first set of PVA inbreds development was initiated in 2011. As of 2014, a total of 224 S<sub>6</sub> inbreds characterized by a deep orange color were successfully extracted from this broad-based population. The new inbreds were evaluated under managed drought during the minor seasons of 2014 and 2015. Following the performance assessment under managed drought, the inbreds were advanced to S<sub>7</sub> and S<sub>8</sub> and kernels from the S<sub>8</sub> were sampled for PVA analysis in the laboratory at the Food and Nutrition Laboratory of IITA, Ibadan [20]. After the generation of the PVA inbred lines from the extra-early PVA cultivar 2009 TZEE-OR1 STR, a new set of PVA inbred lines was generated from the biparental populations of TZdEEI 12  $\times$  TZdEEI 95 and TZdEEI 7  $\times$  TZdEEI 12. The F<sub>1</sub> hybrids from these crosses were advanced through several cycles of repeated inbreeding and selection based on a deep orange kernel color. At the S<sub>5</sub> stage, selected lines were evaluated under combined heat and drought stress at Kadawa (Nigeria) in 2021 (Badu-Apraku Unpublished). Based on the performance, 27 inbred lines (11 from TZdEEI 12  $\times$  TZdEEI 95 and 16 from TZdEEI 7  $\times$  TZdEEI 12) were selected. These inbred lines plus nine other inbred lines extracted from 2009 TZEE-OR1 STR (making a total of 36 inbred lines) and five drought-tolerant inbred testers, are the genetic materials used for the development of the new orange hybrids evaluated in this study. The pedigree information of the inbreds is presented in Table S1.

### 2.2. Generation of Crosses

The 36 extra-early PVA inbred lines were crossed to the 5 drought-tolerant PVA testers using the line by tester (L  $\times$  T) design to generate 180 testcrosses at IITA-Ikenne breeding nursery in 2021. In addition, the testers were crossed in a Diallel fashion to generate 10 hybrids, which were added to the testcross hybrids for this study. A total of 6 extra-early normal yellow endosperm hybrids were included as checks to make 196 hybrids.

### 2.3. Field Evaluation

The 196 orange hybrids were evaluated for agronomic performance at Ikenne (7°52' N, 30°44' E, 61 m a.s.l., 1200 mm mean annual precipitation) under managed drought during the dry seasons (November to February) of 2021–2022 and 2022–2023 and under well-watered conditions during the rainy seasons (June to September) of 2022 and 2023. The managed drought experiment was achieved by supplying 17 mm of sprinkler irrigation water per week up to 25 days after sowing. The plants were allowed to depend on the available soil moisture to reach physiological maturity. Plots consisted of single rows, each 3 m long, with inter- and intra-row spacing of 0.75 and 0.40 m, respectively. Three seeds

were sowed per hole and the seedlings were thinned to two plants per hill two weeks after sowing to give a final plant population density of 66,666 plants/ha. A 14 × 14 lattice design with two replications was used for each experiment. Compound fertilizer, NPK 15:15:15, was applied at sowing at the rate of 60 kg/ha of N, P, and K for managed-drought conditions while a similar rate of application was applied at 2 weeks after sowing (WAS) for well-watered conditions. Top-dressing was performed using urea (46% N) at the rate of 30 kg/ha N at 3 WAS for managed-drought conditions and 5 WAS for well-watered conditions. Weed control was achieved with the application of an herbicide formulation containing gramoxone and primextra at the rate of 5 L/ha as pre-emergence while subsequent weed control was by hand weeding.

#### 2.4. Data Collection

Observations were made on grain yield (GY), days to 50% anthesis (DA), days to 50% silking (DS), anthesis–silking interval (ASI), plant height (PHT), ear height (EHT), ear per plant (EPP), plant aspect (PASP), ear aspect (EASP), stay green characteristic (STGC), and husk cover (HCV) as described in Table 1.

**Table 1.** Description of the measured traits of the extra-early PVA hybrids evaluated under drought stress and well water conditions between 2021 and 2023.

Traits	Stage	Description
DA	Flowering	Number of days from sowing 50% of the plants in a row had pollen shed
DS	Flowering	Number of days from sowing 50% of the plants in a row had silk emergence
ASI	Flowering	The interval between 50% anthesis and silking
PHT	Post flowering	Distance in centimeters from the base of the plant and the first tassel branch
EHT	Post flowering	Distance in centimeters between the base of the plant and the uppermost ear
PASP	Post-flowering	The general phenotypic appearance of the plants in a plot at 70 days after sowing scored on a scale of 1–9, where 1 = excellent and 9 = very poor
HCV	Post-flowering	The protective outer covering of the cob is scored on a scale of 1 to 9 at 70 days after sowing, where 1 = husks tightly arranged and extended beyond the ear tip and 9 = ear tips exposed
STGC	Post-flowering	The ability of the plant to retain the greenness of the leaves at 70 days after sowing is scored on a scale of 1 to 9, where 1 = almost all leaves green and 9 = virtually all leaves dead
EASP	Harvest	The phenotypic appearance of the cob after harvest scored on a scale of 1–9, where 1 = clean, uniform, large, and well-filled ears and 9 = ears with undesirable features, such as diseases, small ears, ears rot and ears with poorly filled grains
EPP	Harvest	Calculated by dividing the number of ears harvested by the number of plants from where the ears were harvested.
GY	Harvest	Computed from the weight of the shelled grain adjusted to 80% shelling percentage and corrected for 15% moisture content

DA: days to anthesis; DS: days to silking; GY: grain yield; ASI: anthesis–silking interval; PHT: plant height; EHT: ear height; PASP: plant aspect; EASP: ear aspect; EPP: ear per plant; HCV: husk cover; STGC: stay green characteristic.

#### 2.5. Data Analysis

Combined analysis of variance (ANOVA) was conducted for each of the research conditions using the mixed linear model (MLM) (Equation (1)) implemented in lmerTest package in R software 4.3.1 [23]. In the MLM, a combination of year and research condition (managed drought or well-watered) was considered as an environment. Thus, four environments namely, WW\_22 (well-watered 2022), WW\_23 (well-watered 2023), DS\_22 (managed drought 2022), and DS\_23 (managed drought 2023) were involved. Environment,

block, and replications were considered as random effects while genotype was considered as fixed effect. The model for the analysis of variance is expressed as:

$$Y = \mu + Rep + Rep (Blk) + G + E + G \times E + e \quad (1)$$

where  $Y$  = phenotype;  $\mu$  = mean of the trait;  $G$  = genotype;  $E$  = environment;  $Rep$  = replication;  $Rep (Blk)$  = replication nested in block;  $G \times E$  = genotype by environment interaction;  $e$  = residual.

The best linear unbiased predictions (BLUPs) under both research conditions were estimated from the ANOVA. The estimates were used to compute Pearson phenotypic correlation between grain yield and other traits under each research condition and between research conditions using the metan package v.1.18 [24]. The MGIDI proposed by Olivoto and Nardino [21] was used to select maize hybrids that combine drought-tolerant adaptive traits and high yield. The MGIDI is based on four principles namely (i) traits rescaling to ensure they have a 0–100 range, (ii) accounting for correlation structure and dimensionality reduction in the data, (iii) using desired traits value to plan ideotype, and (iv) computing the distance between the planned ideotype and the genotype.

Traits rescaling was performed using the equation below

$$rX_{ij} = \frac{\eta_{nj}}{\eta_{oj}} - \frac{\varphi_{nj}}{\varphi_{oj}} * (\theta_{ij} - \eta_{nj}) + \eta_{nj} \quad (2)$$

where  $\varphi_{oj}$  and  $\eta_{oj}$  represent the original minimum and maximum values of the  $j$ th trait of interest;  $\varphi_{nj}$  and  $\eta_{nj}$  are the new minimum and maximum values for  $j$ th trait of interest after rescaling; and  $\theta_{ij}$  is the original value for  $j$ th trait of interest for the  $i$ th genotype.

The values for  $\eta_{nj}$  and  $\varphi_{nj}$  were chosen in a manner that for traits in which negative gains are desired,  $\eta_{nj} = 0$  and  $\varphi_{nj} = 100$  was used while for traits where positive gains are desired,  $\eta_{nj} = 100$  and  $\varphi_{nj} = 0$ . In the final rescaled table ( $rX_{ij}$ ), the column has a 0–100 range.

Following traits rescaling, explanatory factor analysis was carried out. The rescaled values were first used to group correlated traits into factors before the factorial scores were estimated for the traits using Equation (3) below.

$$X = \mu + Lf + e \quad (3)$$

where  $X$  is a  $p \times 1$  vector of the observations that were rescaled;  $\mu$  is a  $p \times 1$  vector of the standardized means;  $L$  is a  $p \times f$  matrix of the factorial loadings;  $f$  is a  $p \times 1$  vector of the common factors; and  $e$  is a  $p \times 1$  vector of error.

For  $p$  and  $f$ , which represent the number of traits and common factors retained, respectively, the correlation matrix of the rescaled values was used to generate the eigen values and eigen vectors. For the initial loadings, factors with eigen values higher than one were considered before the varimax rotation criteria was implemented for analytic rotation and estimation of final loadings according to (Kaiser) [25]. The final scores were obtained using Equation (4) below:

$$F = Z \left( A^T R^{-1} \right)^T \quad (4)$$

where  $F$  is a  $g \times f$  matrix with the factorial scores;  $Z$  is a  $g \times p$  matrix with the rescaled means;  $A$  is a  $p \times f$  matrix of canonical loading, and  $R$  is a  $p \times p$  correlation matrix between the traits. The  $g$ ,  $f$ , and  $p$  represent the number of genotypes, the factor kept, and the traits measured, respectively.

Ideotype planning was performed such that the ideotype has the highest rescaled value of 100 for all the analyzed traits. The MGIDI was finally estimated using Equation (5).

$$MGIDI = \sum_{i=1}^f \left[ (Y_{ij} - Y_j)^2 \right]^{0.5} \quad (5)$$

where the *MGIDI* is the multi-trait genotype–ideotype distance index for the *i*th genotype;  $Y_{ij}$  is the score of the *i*th genotype in the *j*th factor being *g* and *f*; and  $Y_j$  is the *j*th score of the ideotype.

From the above equation, the lower the *MGIDI* score of a genotype, the closer the genotype is to the ideotype. Thereafter, the proportion of the *MGIDI* of the genotypes explained by the correlated factor is used to display the strengths and weaknesses of the genotypes based on Equation (6).

$$w_{ij} = \frac{\sqrt{D_{ij}^2}}{\sum_{i=1}^f \sqrt{D_{ij}^2}} \quad (6)$$

where  $w_{ij}$  is the proportion of the *MGIDI* of the *i*th genotypes explained by the correlated *j*th factor;  $D_{ij}^2$  is the distance between the *i*th genotype and the ideotype for the *j*th factor.

Data processing and the computation of the index were carried out using the *Metan* package in R [24].

To ascertain the effectiveness of the *MGIDI*, the results obtained from the latter were compared to those from a multiple-trait base index (*MBI*) proposed by Badu-Apraku et al. [26] that incorporates grain yield, plant and ear aspects, stay green characteristic, number of ears per plant, plant and ear height, husk cover, and anthesis–silking interval. The *MBI* has been used for the selection of hybrids that combine drought tolerance with outstanding yield performance based on Equation (7) below:

$$MBI = (2 \times GY) + EPP + PHT + EHT - PASP - EASP - ASI - STGC - HCV \quad (7)$$

where *MBI* = multiple-trait base index, *GY* = grain yield, *EPP* = ear per plant, *PASP* = plant aspect, *EASP* = ear aspect, *ASI* = anthesis–silking interval, *STGC* = stay green characteristic, *HCV* = husk cover, *PHT* = plant height, and *EHT* = ear height.

The traits used in the *MBI* were standardized to reduce the effects of different scales of the traits. A positive *MBI* value indicates tolerance to drought while a negative value indicates susceptibility to drought. Yield reduction attributable to drought stress was estimated using Equation (8).

$$Yield\ reduction\ (YR\ \%) = \frac{(\text{yield under well} - \text{watered} - \text{yield under managed drought}) \text{conditions}}{\text{yield under well} - \text{watered conditions}} \times 100 \quad (8)$$

Forty hybrids from the *MGIDI* selection comprising the 20 best, 10 average, and 10 worst performers were selected for *GGE* biplot analysis using *Metan*. The “mean vs. stability” view of the *GGE* biplot was used to determine hybrids with stable and high grain yields across managed drought and well-watered research conditions. The data were neither transformed nor standardized and were environment centered. The model for the *GGE* biplot is shown in Equation (9) below:

$$Y_{ij} - Y_j = \lambda_1 \epsilon_{i1} \eta_{j1} + \lambda_2 \epsilon_{i2} \eta_{j2} + \epsilon_{ij} \quad (9)$$

where  $Y_{ij}$  is the average yield of *i*th genotype in *j*th environment,  $Y_j$  is the average yield across genotypes in environment *j*,  $\lambda_1$  and  $\lambda_2$  are the singular values for principal components I and II, respectively,  $\epsilon_{i1}$  and  $\epsilon_{i2}$  are the scores of the *i*th genotype for the principal components I and II,  $\eta_{j1}$  and  $\eta_{j2}$  are the scores of the *j*th genotype for the principal components I and II, and  $\epsilon_{ij}$  is the error associated with the *i*th genotype in *j*th environment.

Drought susceptibility index was calculated for the 196 hybrids to assess the tolerance of the 196 hybrids using performance based on grain yield alone according to [27] as shown below.

$$DSI = 1 - \frac{\left(\frac{YMD}{YWW}\right)}{1 - \left(\frac{XMD}{XWW}\right)} \quad (10)$$

where *DSI*: drought susceptibility index; *YMD*: grain yield under managed drought; *YWW*: grain yield under well-watered conditions; *XMD*: mean of grain yield under managed drought; *XWW*: mean of grain yield under well-watered conditions.

### 3. Results

#### 3.1. Analysis of Variance for Grain Yield and Other Traits under Managed Drought and Well-Watered Conditions

The ANOVA results across well-watered conditions presented in Table 2 revealed significant ( $p < 0.05$ ) genotype mean squares for all traits except EASP. Environment mean square was significant ( $p < 0.05$ ) for GY, ASI, PASP, and EPP. Under managed drought, a significant ( $p < 0.001$ ) genotype mean square was observed for all traits. The environment mean square was significant ( $p < 0.05$ ) for ASI and HCV, while genotype  $\times$  environment mean square was significant for ASI, PHT, EPP, and STGC (Table 3).

**Table 2.** Mean squares of grain yield and other measured traits across well-watered environments.

Source of Variation	DF	GY	ASI	PHT	EHT	PASP	EASP	EPP	HCV
Environment (Env)	1	37,593,481 *	46.76 *	6.65 <sup>ns</sup>	50.26 <sup>ns</sup>	37.12 *	3.46 <sup>ns</sup>	1.25 *	3.71 <sup>ns</sup>
Genotype (Gen)	195	4,330,349 ***	0.71 **	422.55 ***	192.61 ***	2.07 ***	0.85 <sup>ns</sup>	0.02 ***	7.19 ***
Env $\times$ Gen	195	1,174,838 <sup>ns</sup>	0.42 <sup>ns</sup>	67.95 <sup>ns</sup>	54.75 <sup>ns</sup>	0.65 <sup>ns</sup>	1.00 <sup>ns</sup>	0.01 <sup>ns</sup>	0.82 <sup>ns</sup>
Residual		1,108,155	0.40	69.28	52.12	0.89	0.75	0.01	0.84
Min		590.5	0.00	129.2	57	2	2	0.38	1
Max		10,064	4.00	213.8	180	8	8	1.27	8
Mean		5568	0.62	179.3	83.56	4.44	4.68	0.92	3.92

DF: degree of freedom; Env: environment; GY: grain yield; ASI: anthesis–silking interval; PHT: plant height; EHT: ear height; PASP: plant aspect; EASP: ear aspect; EPP: ear per plant; HCV: husk cover; \*, \*\*, \*\*\*: significant at 0.05, 0.01 and 0.001 probability levels, respectively. <sup>ns</sup>: non-significant.

**Table 3.** Mean squares of grain yield and others agronomic traits across drought stress environments.

Source of Variation	DF	GY	ASI	PHT	EHT	PASP	EASP	EPP	HCV	STGC
Environment (Env)	1	1,560,132 <sup>ns</sup>	35.57 *	673.19 <sup>ns</sup>	2.05 <sup>ns</sup>	8.51 <sup>ns</sup>	2.45 <sup>ns</sup>	0.19 <sup>ns</sup>	81.67 ***	0.10 <sup>ns</sup>
Genotype (Gen)	195	1,464,750 ***	3.085 ***	485.84 ***	193.81 ***	1.55 ***	1.61 ***	0.05 ***	3.90 ***	1.93 ***
Env $\times$ Gen	195	979,359.6 <sup>ns</sup>	2.16 *	258.2 **	70.25 <sup>ns</sup>	1.08 <sup>ns</sup>	1.15 <sup>ns</sup>	0.04 **	0.95 <sup>ns</sup>	1.07 ***
Residual		835,346	1.57 <sup>ns</sup>	191.5	65.2	0.98	1	0.03	0.78	0.99
Min		0	0	65	34	2	1	0	1	2
Max		8407	9	205.8	128.4	9	9	1.17	8	9
Mean		2138.74	1.5	147.9	76.27	5.42	5.13	0.71	3.97	5.02

DF: degree of freedom; Env: environment; GY: grain yield; ASI: anthesis–silking interval; PHT: plant height; EHT: ear height; PASP: plant aspect; EASP: ear aspect; EPP: ear per plant; HCV: husk cover; STGC: stay green characteristic; \*, \*\*, \*\*\*: significant at 0.05, 0.01 and 0.001 probability levels, respectively; <sup>ns</sup>: non-significant.

#### 3.2. Selection of Outstanding Orange Hybrids with Drought Tolerance Using the MGIDI Selection Method

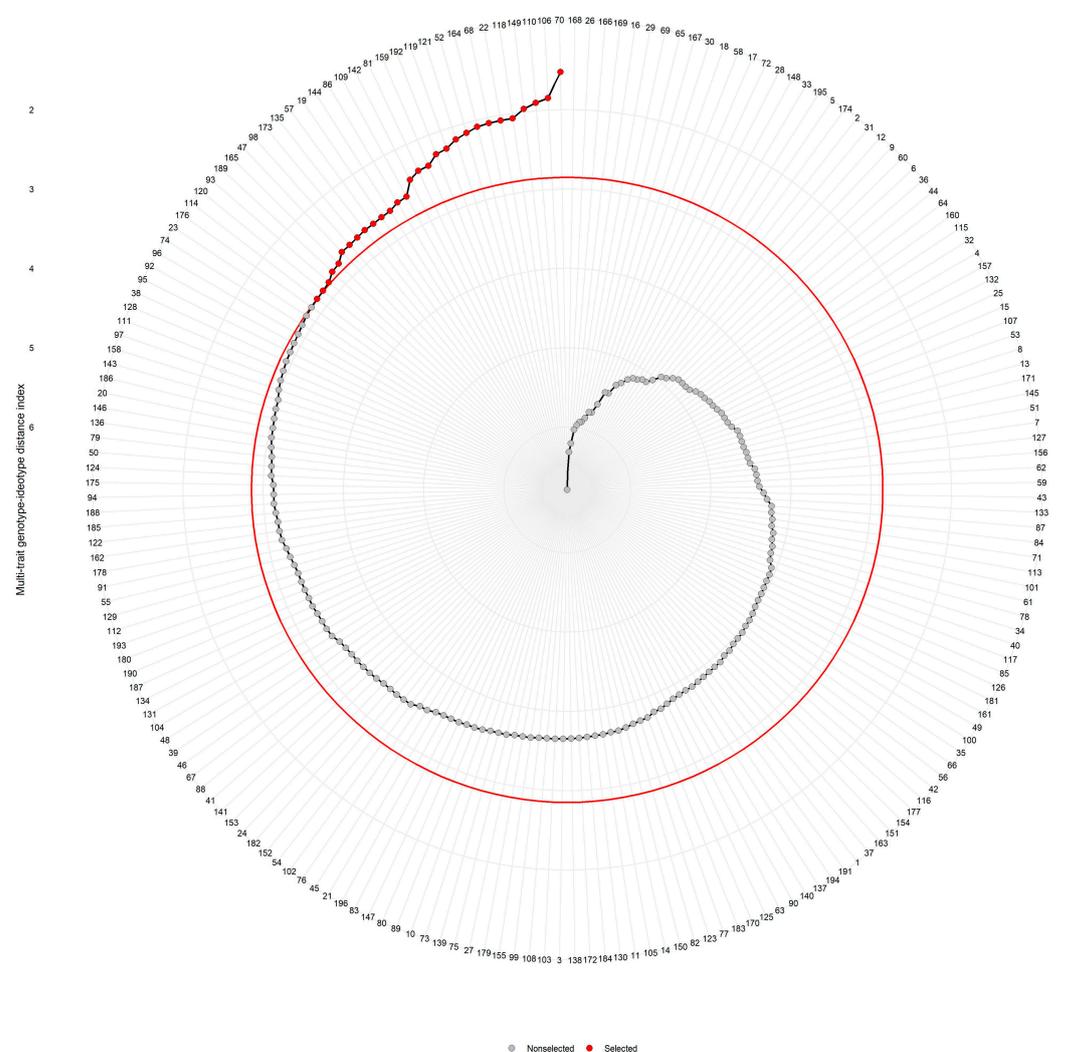
The MGIDI partitioned the measured traits into three factors. The first factor (FA1) was correlated with GY, EPP, PASP, EASP, EPP, ASI, and STGC. The second factor (FA2) was associated with HCV while the third factor (FA3) was correlated with PHT and EHT. All the measured traits showed desirable predicted gains from selection observed in the sense of selection and perfect goal. Generally, the MGIDI provided a total predicted gain from selection of 11.96% for traits for which higher positive expression was desirable and  $-16.65\%$  for traits for which higher negative expression was desirable (Table 4). Using a selection intensity of 15%, 29 hybrids comprising 28 new hybrids and one commercial check (TZEEI 79  $\times$  TZEEI 82) were selected as drought-tolerant hybrids (Figure 1 and Table S2). Of the selected testcross hybrids, 10 hybrids (34%) were developed from crosses involving the tester TZEEIOR 197. The best five hybrids based on the MGIDI were TZEEIOR 510  $\times$  TZEEIOR 97, TZEEIOR 510  $\times$  TZEEIOR 249, TZEEIOR 321  $\times$  TZEEIOR 197, TZEEIOR 384  $\times$  TZEEIOR 30, and TZEEIOR 458  $\times$  TZEEIOR 197. Similarly, the worst

five hybrids were TZEEIOR 523 × TZEEIOR 9A, TZEEIOR 221 × TZEEIOR 9A, TZEEIOR 24 × TZEEIOR 30, TZEEIOR 41 × TZEEIOR 30, and TZEEIOR 45 × TZEEIOR 30 (Table S2).

**Table 4.** Factorial loadings and predicted genetic gains of 196 orange hybrids based on the MGIDI.

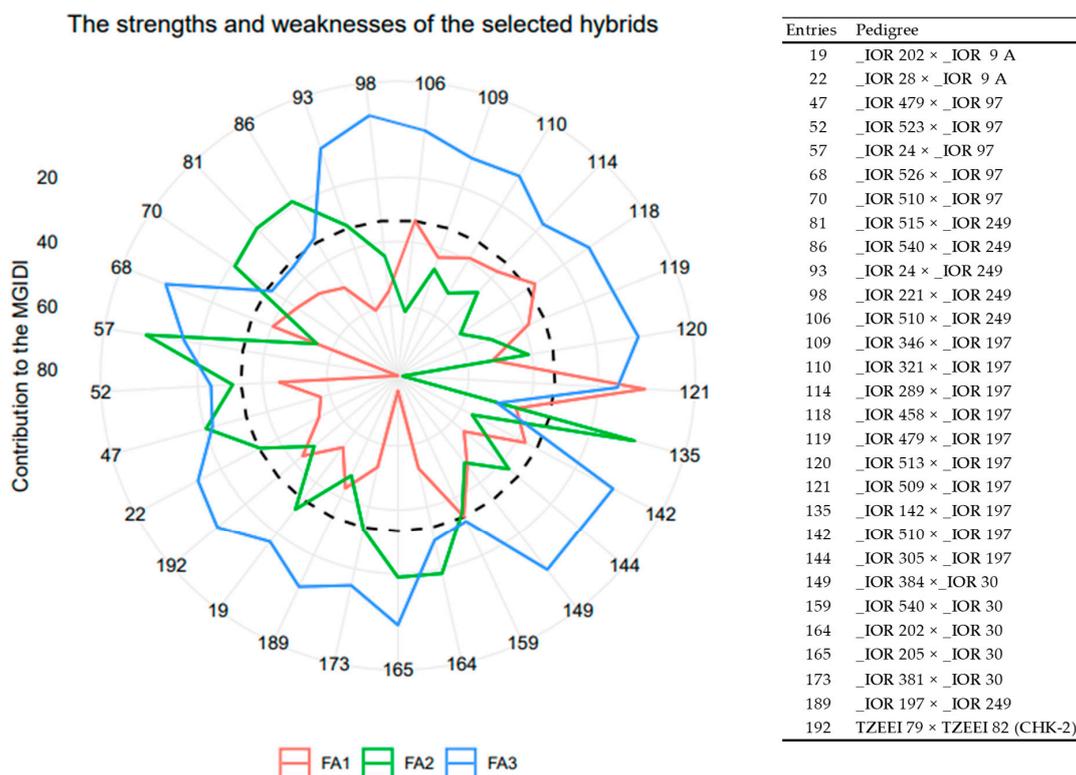
Traits	FA1	FA2	FA3	Xo	Xs	Predicted Gain	Sense	Goal
ASI	−0.62	0.49	−0.13	1.51	1.49	−1.03	decrease	100
PHT	−0.28	−0.33	<b>0.80</b>	147.87	148.24	0.25	increase	100
EHT	−0.11	0.13	<b>0.93</b>	76.36	78.69	3.04	increase	100
HCV	0.00	−0.90	−0.02	3.97	3.57	−10.13	decrease	100
PASP	−0.73	−0.20	0.41	5.42	5.41	−0.17	decrease	100
EASP	−0.89	−0.08	0.19	5.13	5.06	−1.24	decrease	100
EPP	−0.85	0.02	0.11	0.71	0.73	2.63	increase	100
GY	−0.91	−0.02	0.19	2138.74	2267.90	6.04	increase	100
STGC	−0.48	−0.47	0.27	5.02	4.82	−4.08	decrease	100

NB: bold values represent traits that are correlated to each of the factor analysis 1, 2, and 3. FA1: factor analysis 1; FA2: factor analysis 2; FA3: factor analysis 3; Xo: population mean before selection; Xs: population mean of the selected individuals; ASI: anthesis–silking interval, PHT: plant height, EHT: ear height, HCV: husk cover, PASP: plant aspect, EASP: ear aspect, EPP: ear per plant, GY: grain yield, STGC: stay green characteristic.



**Figure 1.** View of the selected (red dots) and non-selected (grey dots) hybrids based on multi-traits genotype–ideotype distance index. The lower the MGIDI of a genotype on the left side of the graph, the closer the genotype is to an ideotype. The red circle is the cut-off MGIDI value that determines the genotype to be selected based on their individual MGIDI estimate.

The result of the analysis to unmask the strengths and weaknesses of the selected extra-early maize hybrids is presented in Figure 2. In this figure, the proportion of each factor to the MGIDI accounts for the strengths and weaknesses. The red, green, and blue lines represented the three factors, and the factor protruding the most to a genotype is indicated as the strength of the genotype. Based on this, entry 121 had strength for FA1, which is correlated to GY, EASP, PASP, EPP, and STGC. Entries 135, 86, 81, 70, 57, 47, and 164 had strength for FA2, which is correlated to PHT and EHT. Entries 22, 192, 19, 189, 173, 165, 159, 149, 144, 142, 120, 119, 118, 114, 110, 109, 106, 98, 93, 68, and 52 showed strength for FA3, which is correlated to ASI and HCV (Table 5 and Figure 2).



**Figure 2.** The radar plot showing the strengths and weaknesses of the MGIDI selected hybrids. The red, green and blue lines represent the three factors and the factor protruding the most to a genotype is indicated as the strength of the genotype. The black broken circle shows the theoretical value if all the factors contributed equally. \_IOR: TZEEIOR.

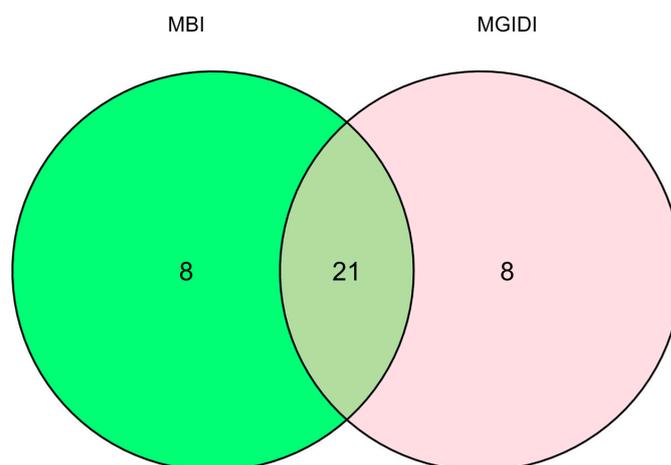
**Table 5.** Factorial loadings, communalities and uniqueness of 29 selected orange hybrids based on the multi-trait genotype–ideotype distance index.

Traits	FA1	FA2	FA3	Communality	Uniqueness
ASI	0.13	0.26	<b>0.87</b>	0.84	0.16
PHT	0.15	− <b>0.84</b>	−0.05	0.73	0.27
EHT	−0.27	− <b>0.71</b>	0.10	0.59	0.41
HCV	−0.02	0.36	− <b>0.74</b>	0.69	0.31
PASP	<b>0.72</b>	0.21	0.01	0.57	0.43
EASP	<b>0.91</b>	−0.06	0.03	0.82	0.18
EPP	<b>0.74</b>	0.00	0.14	0.57	0.43
GY	<b>0.81</b>	−0.08	0.19	0.69	0.31
STGC	<b>0.38</b>	0.09	−0.18	0.18	0.82

NB: bold values represent traits are correlated to each factor analysis 1, 2, and 3. FA1: factor analysis 1; FA2: factor analysis 2; FA3: factor analysis 3; ASI: anthesis–silking interval, PHT: plant height, EHT: ear height, HCV: husk cover, PASP: plant aspect, EASP: ear aspect, EPP: ear per plant, GY: grain yield, and STGC: stay green characteristic.

### 3.3. Selection of Drought-Tolerant Hybrids Using the Base Index Derived from Multiple Traits

Based on the multiple-trait base index (MBI) that integrated grain yield, plant aspect, ear aspect, anthesis–silking interval, stay green characteristic, ears per plant, plant height, ear height, and husk cover, 99 hybrids had positive selection base indices. The positive selection base index varied from 0.06 for Entry 150 to 15.93 for Entry 121. Three checks (TZEEI 79 × TZEEI 82, TZdEEI 1 × TZdEEI 12, and TZEEIY Pop STR C5 × TZEEI 58) were identified as drought-tolerant hybrids among the six checks used in this study. The best five top-performing hybrids under managed drought stress were hybrids TZEEIOR 509 × TZEEIOR 197, TZEEIOR 458 × TZEEIOR 197, TZEEIOR 510 × TZEEIOR 30, TZEEIOR 510 × TZEEIOR 97, and TZEEIOR 510 × TZEEIOR 249. Three of these five hybrids, namely TZEEIOR 510 × TZEEIOR 97, TZEEIOR 510 × TZEEIOR 249, and TZEEIOR 458 × TZEEIOR 197, were also identified to be superior for drought tolerance using the MGIDI procedure. The five worst hybrids under managed drought were hybrids TZEEIOR 41 × TZEEIOR 30, TZEEIOR 24 × TZEEIOR 30, TZEEIOR 221 × TZEEIOR 9 A, TZEEIOR 45 × TZEEIOR 30, and TZEEIOR 525 × TZEEIOR 97 (Table S3). The selected hybrids from the MGIDI were included in the list of the hybrids identified as drought-tolerant using the MBI without consideration for selection intensity. However, when 15% selection intensity was applied as performed for the MGIDI, 21 of 29 hybrids (72%) selected were similar to the selection results from the MGIDI (Figure 3).



**Figure 3.** Venn diagram showing the similarity level (as the intersection) between the multiple-trait base index (MBI) and the multi-trait genotype–ideotype distance index (MGIDI) methods.

### 3.4. Yield Performance and Grain Yield Reduction under Drought

Under drought stress condition, grain yield ranged from 198 kg/ha for hybrid TZEEIOR 41 × TZEEIOR 30 to 4044 kg/ha for TZEEIOR 509 × TZEEIOR 197 with an average of 2139 kg/ha. The best-yielding hybrid (TZEEIOR 509 × TZEEIOR 197) had 34% higher grain yield than the best check (TZEEI 79 × TZEEI 82) that had a grain yield of 3015 kg/ha. Under well-watered condition, grain yield varied from 1251 kg/ha for TZEEIOR 28 × TZEEIOR 30 to 7455 kg/ha (TZEEIOR 509 × TZEEIOR 197) with a mean of 5568 kg/ha. The best check (TZEEIOR 12 × TZEEIOR 223) had a grain yield of 5732 kg/ha, which translates to 30% grain yield reduction compared to the best hybrid (Table S4).

In general, the grain yield reduction in the hybrids under managed drought stress compared with that of the well-watered conditions varied from 35% for TZEEIOR 321 × TZEEIOR 249 to 89% for TZEEIOR 41 × TZEEIOR 30 with an average of 61%. The grain yield reduction in the selected drought-tolerant hybrids from the MGIDI under drought compared to that of the well-watered varied from 36% for the check hybrid TZEEI 79 × TZEEI 82 to 69% for TZEEIOR 202 × TZEEIOR 9A. In addition, 10 (~35%) and 26 (~90%) hybrids from the MGIDI selection had better performance than the selected commercial check under managed drought and well-watered conditions, respectively. The yield advantage

of the hybrids with better performance than the selected check was 12 and 33% under managed drought and well-watered conditions, respectively. For the selected hybrids from the MBI, grain yield reduction varied from 35% for TZEEIOR 321 × TZEEIOR 249 to 62.71% for TZEEIOR 510 × TZEEIOR 197. Furthermore, 16 (~55%) and 27 (~93%) hybrids from the MBI selection had better performance than the selected commercial check under managed drought and well-watered conditions, respectively (Table 6). Considering hybrids grain yield potential under managed drought and well-watered conditions alone, 25 of 29 hybrids selected by the MGIDI had drought susceptibility index (DSI) estimates lower than the average DSI of the hybrid population (0.99). When similar observation was made for the MBI, 26 of 29 hybrids had DSI estimates lower than the average DSI of the hybrid population (Tables 5 and S2).

Drought stress reduced PHT by 17%, EHT by 9%, EPP by 22%, DP by 2%, but increased ASI by 181%, DS by 0.2%, HCV by 7%, PASP by 25%, and EASP by 11% (Table S5).

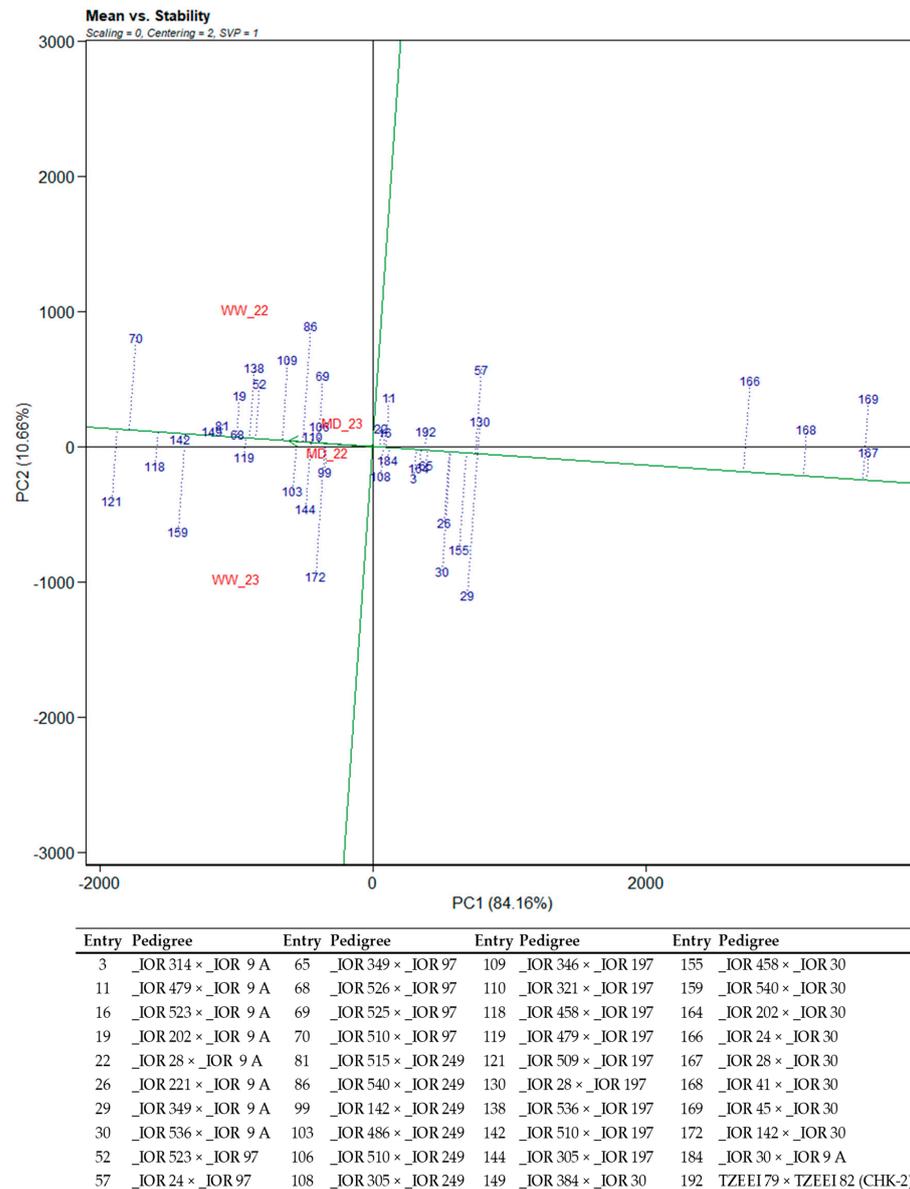
**Table 6.** Grain yield (kg/ha) of selected drought-tolerant extra-early maturing hybrids identified from both the MGIDI and the multiple-trait base index with a 72% level of similarity.

Entries	Genotypes	GY (kg/ha) under MD	GY (kg/ha) under WW	YR%	MBI	MGIDI	DSI
19	_IOR 202 × _IOR 9 A	2098	6714	68.75	5.39 *	2.65	1.12
22	_IOR 28 × _IOR 9 A	2501	5341	53.17	7.79	2.06	0.86
39	_IOR 314 × _IOR 97	3604	6993	48.46	8.38	3.37 *	0.79
47	_IOR 479 × _IOR 97	2858	5248	45.54	7.7	2.69	0.74
52	_IOR 523 × _IOR 97	3054	6386	52.17	10.5	2.12	0.85
57	_IOR 24 × _IOR 97	2503	4392	43.02	6.41	2.66	0.70
68	_IOR 526 × _IOR 97	3319	6436	48.43	9.69	2.07	0.79
70	_IOR 510 × _IOR 97	3569	7380	51.64	15.07	1.53	0.84
74	_IOR 321 × _IOR 249	3208	4918	34.78	9.31	2.89 *	0.56
81	_IOR 515 × _IOR 249	2917	6722	56.6	9.81	2.36	0.92
86	_IOR 540 × _IOR 249	2923	5818	49.76	9.02	2.6	0.81
92	_IOR 205 × _IOR 249	2559	5240	51.17	8.46	2.91 *	0.83
93	_IOR 24 × _IOR 249	1989	4367	54.46	2.29 *	2.83	0.88
98	_IOR 221 × _IOR 249	2157	5186	58.41	4.19 *	2.68	0.95
106	_IOR 510 × _IOR 249	3295	5846	43.64	12.15	1.85	0.71
109	_IOR 346 × _IOR 197	2438	6398	61.89	6.58	2.59	1.00
110	_IOR 321 × _IOR 197	3116	5714	45.46	11.24	1.9	0.74
114	_IOR 289 × _IOR 197	2873	7341	60.86	7.61	2.85	0.99
118	_IOR 458 × _IOR 197	3643	7062	48.42	15.91	2.06	0.79
119	_IOR 479 × _IOR 197	3074	6428	52.18	10.41	2.24	0.85
120	_IOR 513 × _IOR 197	2799	6953	59.75	6.03 *	2.85	0.97
121	_IOR 509 × _IOR 197	4044	7455	45.75	15.94	2.17	0.74
128	_IOR 205 × _IOR 197	2308	5990	61.46	6.71	2.95 *	1.00
135	_IOR 142 × _IOR 197	2578	5954	56.69	6.03 *	2.66	0.92
142	_IOR 510 × _IOR 197	2629	7051	62.72	8.9	2.42	1.02
144	_IOR 305 × _IOR 197	2648	5885	55	6.08 *	2.64	0.89
149	_IOR 384 × _IOR 30	3415	6753	49.43	10.71	1.96	0.80
158	_IOR 509 × _IOR 30	3097	5469	43.37	6.84	3.01 *	0.70
159	_IOR 540 × _IOR 30	3261	7086	53.98	10.69	2.36	0.88
164	_IOR 202 × _IOR 30	2297	4918	53.29	8.97	2.08	0.87
165	_IOR 205 × _IOR 30	2360	6280	62.42	4.80 *	2.76	1.01
173	_IOR 381 × _IOR 30	3000	5898	49.13	6.97	2.67	0.80
175	_IOR 536 × _IOR 30	3498	7390	52.67	6.34	3.13 *	0.86
178	_IOR 525 × _IOR 30	3217	5618	42.73	8.85	3.17 *	0.69
179	_IOR 510 × _IOR 30	3805	6392	40.47	15.18	3.63 *	0.66
189	_IOR 197 × _IOR 249	2300	5464	57.91	4.53 *	2.78	0.94
192	TZEEI 79 × TZEEI 82 (CHK-2)	3015	4720	36.13	7.91	2.26	0.59

\_IOR: TZEEIOR; GY: grain yield; YR: yield reduction; MBI: multiple-trait base index; MGIDI: multi-trait genotype-ideotype distance index; DSI: drought susceptibility index; \*: hybrids that were not selected as drought-tolerant using the alternative selection method.

### 3.5. Stability Analysis of Forty Orange Hybrids Comprising Best, Average, and Worst Performing Genotypes across All Environments

The GGE biplot view of the 40 extra-early provitamin A hybrids comprising the 20 best, 10 average, and 10 worst performing hybrids from the MGIDI selection analyzed for performance stability is presented in Figure 4. Approximately 95% of the variability in grain yield of the orange hybrids was accounted for by the first two principal components.



**Figure 4.** Mean performance and stability of 40 extra-early provitamin A maize hybrids based on grain yield as measured by the principal components across four environments at Ikenne. \_JOR: TZEEIOR.

The best-yielding and most stable extra-early provitamin A hybrids were identified using the “mean vs. stability” biplot view across the test environments. The vertical line passing through the biplot origin is the average tester coordinate (ATC) which separates the hybrids based on yield performance. The more the distance between the hybrid and the ATC to the left, the better the yield performance of the hybrid and vice versa. The length of the hybrid projection from the ATC abscissa (horizontal line) determines their stability. The shorter the projection, the more stable the hybrid and vice versa. Based on these criteria, the five top-yielding hybrids across the test environments in descending order included TZEEIOR 509 × TZEEIOR 197, TZEEIOR 510 × TZEEIOR 97, TZEEIOR 458 × TZEEIOR



#### 4. Discussion

Genetic variability is crucial for the improvement of desired traits in crop improvement programs [28–30]. Significant genotype mean squares observed for grain yield and other traits under the contrasting research conditions was indicative of the presence of genetic variability among the extra-early orange hybrids under managed drought and well-watered conditions. These results provide a basis for rapid genetic gains from selection and identification of hybrids with desirable traits for each research condition [31,32]. Significant environment mean squares observed for anthesis–silking interval and husk cover under managed drought indicated that the difference in environmental conditions that prevailed in the two years of evaluation affected these traits [33]. A similar trend of results was observed for grain yield, anthesis–silking interval, plant aspect, and ears per plant under well-watered conditions. The well-watered trials were carried out under rain-fed conditions. Considerable variation in rainfall occurs from year to year in Nigeria, even for the same site. Consequently, evaluation in several years is necessary, to expose materials being evaluated to the possible variation in rainfall, towards ensuring that materials that are selected are productive under wide rainfall regimes. Significant genotype  $\times$  environment mean squares observed for anthesis–silking interval, plant height, ears per plant, and stay green characteristic under managed drought indicated that the genotypes response pattern differ under the two managed drought environments considered in this study. In contrast, the lack of significant environment and genotype  $\times$  environment interaction mean squares for grain yield indicate that the relative performances of the genotypes were stable under the managed drought condition investigated. This is also true for the well-watered condition for which the mean squares for genotype by environment interactions were not significant. These results indicate that plant traits differ in their response to environmental conditions.

In the present study, both the MGIDI and the MBI were each used to identify 29 genotypes combining drought tolerance with high yield. Of the selected genotypes from both selection procedures, 72% similarity was observed. As reported by several authors [21,24,34,35], the multiple-trait base index is usually affected by multicollinearity of traits which in turn affects the efficiency of selection. The MGIDI is, however, not affected by traits multicollinearity making it a better selection procedure when ideotype design is of major interest. In addition, the MGIDI provides knowledge of the strengths and weaknesses of selected ideotype, which could guide plant breeders in identifying traits for further improvement. The MGIDI has been used to develop climate-resilient maize hybrids through the development of better selection approaches and optimization of breeding strategies [36]. Improvement in crop performance, the goal of all plant breeding programs, is usually pursued through the assembly in new varieties of genes for various desirable plant traits associated with high grain productivity [37]. The selected 29 drought-tolerant orange hybrids from the MGIDI comprised of 28 new hybrids and one commercial check. The 10 and 26 hybrids ranked above this commercial check under managed drought and well-watered conditions have an average yield advantage of 12% and 33%, respectively. These hybrids show promise for commercialization in SSA, as they are not only drought-escape as being extra-early genotypes, but also have the inherent ability to withstand drought occurring during the growing season. These hybrids could also be useful for developing open-pollinated varieties, three-way and double cross hybrids for drought-prone areas in SSA. Ten hybrids from those selected by the MGIDI had the tester TZEEIOR 197 as a parent. In a previous study [38], this tester was reported as an outstanding inbred line with positive and significant general combining ability effects for grain yield under drought. These results indicate TZEEIOR 197 to possess genes for drought tolerance as well as show it as an excellent combiner that could be used for the development of more drought-tolerant extra-early orange hybrids. In the present study, the superior combining ability was further demonstrated by the fact that hybrid combinations involving this tester were more in number than those from any other inbred tester among the selected hybrids combining drought tolerance with high yield.

Examination of the strengths and weaknesses of the MGIDI selected hybrids revealed hybrid TZEEIOR 509 × TZEEIOR 197 as the only genotype associated with the first factor. This factor correlates with high grain yield, increased ears per plant, and improved ear and plant aspects under drought conditions, an indication that this hybrid has desirable genes for these four traits under managed drought conditions. This provides a basis for its high grain yield under drought stress as well as under well-watered environments. Hybrids associated with the second factor could be selected and used as parent for plant and ear height improvement while hybrids associated to the third factor could be used as parent in a breeding program for the improvement of anthesis–silking interval and husk cover improvement. The initiation of three-way and double-cross hybridization program among these promising hybrids could further increase the number of hybrids with desired performance under drought stress.

In this study, the extended anthesis–silking interval combined with reduction in ears per plant, plant height, ear height and increased plant and ear aspects, and husk cover observed under drought stress contributed to the reduction in grain yield under drought. The imposed drought stress covered the flowering and grain filling periods. Increased anthesis–silking interval is known to increase the incidence of barrenness [39,40]. The grain yield loss obtained in the present study ranged from 35 to 89% with an average of 61% under managed drought conditions. These yield losses fall within the range reported by other authors [16,40–43]. The magnitude of grain yield reduction as a result of drought is known to depend on the duration and intensity of the drought stress [44–47].

Given that the occurrence of drought is unpredictable and crop varieties must optimize productivity under diverse environmental conditions, improved varieties must show outstanding and stable performance under both drought and well-watered conditions. Hybrids that showed these attributes in the present study and whose superior performances were stably expressed viz. TZEEIOR 509 × TZEEIOR 197, TZEEIOR 458 × TZEEIOR 197 and TZEEIOR 510 × TZEEIOR 97, have great potential for commercialization for cultivation in drought-prone environments and qualify for more extensive testing under diverse conditions.

Grain yield, plant aspect, and ears per plant under managed drought were correlated to the same traits under well-watered conditions, suggesting that the expression of these traits is governed by common genetic factors. Similar results have been reported by several authors [41,48,49]. The presence of strong associations between grain yield and ears per plant, plant and ear heights, anthesis–silking interval, plant and ear aspect, and the stay green characteristic under managed drought are indicative of potential of the latter for use as secondary traits in indirect selection in grain yield improvement. Similar results have been found by Bankole et al. [50]. These traits have been used to develop selection index for simultaneous improvement of grain yield and drought tolerance with considerable success [51,52]. In the present study, the relationship between grain yield and anthesis–silking interval under well-watered conditions was not significant but significant under managed drought conditions. These results are consistent with the findings of Bolanos and Edmeades [53] and Bankole et al. [50] who reported that grain yield was weakly correlated to anthesis–silking interval under optimum condition but both are strongly correlated under moisture stress. Knowledge of trait correlations and how the relationships that they define change under contrasting research conditions is useful in guiding plant breeders in the implementation of a breeding program targeting improvements in one trait through improvement in another that is easier to measure [36,54].

## 5. Conclusions

The results of this study revealed significant genetic variability among the newly developed extra-early maturing orange hybrids. Some of the hybrids have considerable potential for mitigating food insecurity and reducing malnutrition in SSA. The inbred line TZEEIOR 197 possesses outstanding attributes for the development of extra-early provitamin A-rich hybrids for drought-prone environments in SSA. Both the multiple-trait

base index and the multi-trait genotype–ideotype distance index selection outcome showed 72% similarity. The hybrids TZEEIOR 509 × TZEEIOR 197, TZEEIOR 458 × TZEEIOR 197 and TZEEIOR 510 × TZEEIOR 97 were identified as the most outstanding drought-tolerant hybrids based on the per se performance, yield stability analysis, and multiple-traits genotype–ideotype distance index. These hybrids should be further tested in multiple stress environments towards efforts to commercialize them in SSA. Additionally, the hybrids could be used as parents for population development and extraction of new and improved inbred lines for further development of new maize ideotypes for farmers in SSA.

**Supplementary Materials:** The following supporting information can be downloaded at: <https://www.mdpi.com/article/10.3390/agronomy14040847/s1>, Table S1: Pedigree information of the genetic materials used for this study; Table S2: Ranking of 196 hybrids using the multi-trait genotype–ideotype distance index (MGIDI); Table S3: Ranking of 196 hybrids using the multiple-trait base index (MBI); Table S4: Grain yield performance of 196 hybrids evaluated under managed drought and well-watered conditions at Ikenne during 2021–2022 and 2022–2023 the dry seasons and the 2022 and 2023 main seasons; Table S5: Trait reduction under managed drought compared to under well-watered conditions.

**Author Contributions:** Conceptualization, T.O.B., B.B.-A. and V.O.A.; methodology, T.O.B., B.B.-A. and I.I.A.; supervisions, B.B.-A. and V.O.A.; formal analysis, T.O.B. and I.I.A.; writing—original draft preparation, T.O.B.; writing—review and editing, B.B.-A., V.O.A., I.I.A. and K.R.N. All authors have read and agreed to the published version of the manuscript.

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**Data Availability Statement:** The datasets used in the present study have been deposited at the IITA CKAN repository.

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**Conflicts of Interest:** The authors declare no conflict of interest.

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