Association studies between grain yield and agronomic traits of a MARS maize (Zea mays L.) population under drought and non-stress condition

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Abstract: The study aimed at examining the associations between yield and other traits under drought stress and non-stress conditions. A total of 150 MARS testcrosses were evaluated under both conditions at the International Institute of Tropical Agriculture substation for two years during the dry season. Genotypic and phenotypic correlation, multiple stepwise regression and path co-efficient analyses were carried out to examine the relationship among the traits under both environments. Results showed anthesis-silking interval, days to silking, husk cover and plant aspect were significantly associated with yield under drought condition at both genotypic and phenotypic levels. Yield was positively correlated with plant and ear height but had a negative correlation with other agronomic traits under well-watered conditions. Regression analysis showed that ears per plant, plant aspect, ear aspect, days to silking, leaf death and plant height had a direct effect on yield, contributing a total of 71.1 % of observed variation under drought, while ears per plant, ear aspect, plant aspect, days to pollen shed, days to silking and plant height contributed about 31.42 % to yield under well-watered conditions. The study concluded that these traits be used as selection criteria as it will aid improvement of maize yield.

Key words: maize; association; grain yield; drought; well-watered; MARS; testcross

Raziskava povezav med pridelkom zrnja in agronomskimi lastnostmi populacij koruze (Zea mays L.) v razmerah suše in v nestresnih razmerah

Izvleček: Namen raziskave je bil preučiti povezave med pridelkom zrnja in drugimi lastnostmi koruze v razmerah suše in v nestresnih razmerah. Na International Institute of Tropical Agriculture je bilo ovrednotenih 150 križanj v obeh razmerah v dveh letih, v sušni sezone. Genotipska in fenotipska korelacija, postopna multipla regresija in koeficient korelacije med neposredno odvisnimi spremenljivkami so bili opravljeni za preučitev razmerja med lastnostmi v obeh rastnih razmerah. Rezultati so pokazali, da so bili znaki kot so obdobje antezis-sviljenje, dnevi do sviljenja in pokritost storža značilno povezani s pridelkom v sušnih razmerah na genotipski in fenotipski ravni. Pridelek je bil v pozitivni korelaciji z višino rastlin in višino storžev na rastlini, a je bil v negativni korelaciji z drugimi aspekti rastline in storža v razmerah dobre oskrbe z vodo. Analiza regresije je pokazala, da so imeli znaki kot so število storžev na rastlino, aspekt rastline in storža, dnevi do sviljenja, smrtnost listov in višina rastlin neposredno učinek na pridelek in so prispevali skupaj 71.1 % opažene spremenljivosti v sušnih razmerah, medtem ko so ti isti znaki prispevali le 31.42 % vpliva na pridelek v razmerah dobre oskrbe z vodo. Na osnovi te raziskave je bilo zaključeno, da bi lahko te lastnosti (znake) uporabili kot selekcijske kriterije, ki bi pomagali izboljšati pridelek koruze.

Ključne besede: koruza; povezava; pridelek zrnja; suša; dobra oskrba z vodo; MARS; testna križanja
1 INTRODUCTION

Maize (Zea mays L.) is presently the second most abundant crop in the world (Ort and Long, 2014) and it has a higher yield potential compared with rice and wheat theoretically, being a C₄ plant (Gong et al., 2015). Despite its potential, the average grain yield of maize in the West and Central Africa (WCA) sub region is estimated to be 1.8 t ha⁻¹ (www.fao.org), this is quite low compared with the yield recorded by many other regions where maize is grown, both in the developing and developed regions of the world (Semagn et al., 2015). Maize yields in Africa are considerably lower than the world average because the cultivation of maize is often prone to abiotic stresses such as drought and low soil fertility in addition to biotic stresses principal among which are Striga species and stem borers (FAO, 2010). Drought stress is the most restrictive agronomic problem confronting maize production, reducing crop yields particularly in regions of the world faced with water limitation, a region with an appreciable number of resource poor farmers (Mhike et al., 2013). In comparison to other abiotic stress factors, drought is the causative factor for the major losses recorded in crop production (Ober, 2008). Although maize has it origin in the tropics, it is extremely prone to drought and heat, particularly at silk emergence and/or when flowers are ready for pollination (Boyer and Westgate, 2004; Lobell et al., 2011, 2014; Frey et al., 2015). A number of studies have reported a significant decrease in the ear traits and also in the commercial value of maize under drought (Edmeades et al., 1995; Ti-da et al., 2006; Mohammadai et al., 2012). The change in the climate is expected to increase the occasions of drought in Africa (Williams and Funk, 2011), together with the fact that maize production is extending into regions that are predisposed to drought stress (Bankole et al., 2017). The world's grain supply is inadequate compared with the demand for food and feed. Furthermore, a gross limitation in crop production worldwide has been predicted as a result of changes in climate, particularly extreme temperatures and drought (Cooper et al., 2014; Frey et al., 2015; Horton et al., 2015). Consequently, it becomes more difficult for a small-scale producer of maize with little or no access to irrigation facilities, who plants varieties that are susceptible to drought stress in sub-Saharan Africa to survive these myriads of challenges (Derera et al., 2008).

Some maize producers have adjusted their planting dates to fit the rainy season which enables the planting periods to correspond with the onset of the rains. Others producers have substituted maize with tree crops because they are more tolerant of changes in temperature and rainfall regimes (Barimah et al., 2014). Subsequently, improvement of crops for tolerance to drought has become imperative of crops under the changing environmental conditions.

The conventional breeding method which exploits inherent genetic variation and uses selection as a tool to integrate desirable traits into adapted genotypes seems to be the most common method in breeding for drought tolerance (Xoconostle-Cazares et al., 2011). The comparative performance of genotypes under drought stress and non-stressed conditions seem to be a common criterion for identifying desirable genotypes for erratic rain-fed conditions (Nouri et al., 2011). Selection for traits such as yield under drought condition proves more challenging as a result of low heritability for the trait under stress conditions (Edmeades et al., 1999; Venuprasad et al., 2007; Ziyomo and Bernado, 2013) rendering the selection process quite ineffective. However, some secondary traits such as anthesis silking interval and ears per plant show high estimates of heritability and genetic correlations with grain yield under drought stress (Bolaños and Edmeades, 1996; Bänziger and Lafitte, 1997; Badu-Apraku et al., 2004). Therefore, an estimate of correlation and path coefficient between the primary trait and other component traits influencing yield is requisite for selection of superior maize genotypes for a successful breeding programme.

Correlation coefficient analyses are useful for selecting the traits that influence grain yield simultaneously (Menkir, 2008). It usually exploits the degree of association among continuous traits (Malik et al., 2005). Despite the usefulness of these estimates in the understanding of complex traits such as grain yield, direct and indirect influences of these traits on productivity are not defined (Baretta et al., 2016). In this context, a method was proposed by Wright (1921) which partitions correlation coefficients into the components of direct and indirect effects known as path coefficient analysis. The analysis not only partitions the correlation coefficient into direct and indirect effects, it also provides the information on the actual contribution of a trait on the yield (Dewey and Lu, 1959).

Several maize breeders have used this method for identifying secondary traits as opposed to the use of genetic correlation (Barros et al., 2010; Chinnadurai and Pothraj, 2011; Begum et al., 2016; Baretta et al., 2016, Talabi et al., 2017; Matin et al., 2017). However, the use of path coefficient analysis to identify and validate secondary traits for selection of improved grain yield under water stress and non-stress condition remains a relevant research focus in maize breeding (Bolaños and Edmeades, 1993, 1996; Bänziger and Lafitte, 1997; Badu-Apraku et al., 2004). The study reported herein examined the nature of inter-trait associations between grain yield and other traits under managed drought stress and non-stress con-
Association studies between grain yield and agronomic traits of a MARS maize (Zea mays L.) population under drought and non-stress condition

2 MATERIALS AND METHOD

2.1 DEVELOPMENT OF THE MARS POPULATION

The details of the development of the marker assisted recurrent selection (MARS) population and data collected have been provided in an earlier report (Bankole et al., 2017). Briefly, a MARS population was developed by crossing two elite drought tolerant maize inbreds (DTPL-W-C7-S2-7-1-1-1-B-5-B*4 and Babangoyo/ MO17LPA/Babangoyo-23-4-3-3-B*6) selected for desirable agronomic traits, resistance to foliar diseases. The resulting F_1 was selfed to generate F_2 bulk seeds, which was grown in 50 rows of 5 m length with a spacing of 0.75 m and self-pollinated to generate 300 F_2:3 lines. A total of 250 F_2:3 lines from this population were planted each in a row and crossed to an inbred tester of the opposite heterotic group. The testcrosses were evaluated under drought stress (DS) and well watered (WW) conditions at the International Institute of Tropical Agriculture (IITA) Ikenne station during the dry season in 2014 and 2015.

2.2 EXPERIMENTAL LAYOUT AND CULTURAL PRACTICES FOR DROUGHT AND WELL-WATERED EXPERIMENTS

A trial which consist of 150 testcrosses of randomly selected 5 lines from a MARS population were evaluated under drought and well-watered conditions at Ikenne during the 2014 and 2015 dry seasons. The testcrosses were arranged in a lattice design with two replications. Each of testcross was planted in a single row of 5 m and 0.75 m spacing between the rows and the plants were spaced 0.25 m apart in the rows. In the DS trial, irrigation was withdrawn from six weeks after planting up to the harvest to elicit drought stress at flowering and grain filling stages, whereas the well-watered trial received irrigation until physiological maturity. Compound fertilizer was application at the rates of 60 kg N, 60 kg P, and 60 kg K ha⁻¹ was done at the time of sowing and an additional 60 kg N ha⁻¹ was added four weeks later. Gramoxone and atrazine were applied for each of the trial as pre-emergence herbicides at 5.0 l ha⁻¹ and supplemented with manual weeding to keep the experiments free from weed.

2.3 DATA COLLECTION

Days to anthesis (DP) and silking (DS) were recorded in each of the plot as the number of days from sowing to when half of the plants were shedding pollen grains and showed emerged silks, respectively. Anthesis-silking interval (ASI) was computed as the interval in days between silking and anthesis. Plant height (PH) and ear height (EH) were measured in centimetres as the distance from the base of the plant to the height of the first tassel branch and the node bearing the upper ear, respectively. Plant aspect (PA) was rated on a scale of 1 to 5, where 1 = excellent overall phenotypic appeal and 5 = poor overall phenotypic appeal. Ear aspect (EA) was scored on a scale of 1 to 5, where 1 = clean, uniform, large, and well-filled ears and 5 = rotten, variable, small, and partially filled ears. Visual leaf death (LD) was scored only under drought at 12 weeks after planting (WAP) on a scale of 1 to 9, where 1 = less than 10 % senesced leaf and 9 = more than 80 % senesced leaf area below the ear. The number of ears per plant (EPP) was the proportion of total number of ears divided by the number of harvested plants. All ears harvested from each plot were shelled to determine percentage moisture and grain yield (GY) adjusted to 15 % moisture.

2.4 STATISTICAL ANALYSES

SAS version 9.3 (SAS institute 2011) was used for the regression analyses. The multiple stepwise regression was then used to describe the contributory relationship among the traits under both the drought stress and well-watered environments using the protocol proposed by Mohammadi et al., 2003. The first and second order traits were organized into the path coefficient analysis based on their contribution to the total variation in grain yield with the use of the multiple stepwise regression analysis. Initially all measured traits were regressed on GY and the first order traits that made contributions to GY through the first order traits were identified by their significant percentage contribution to GY at 5 % probability level. The other traits that made contributions to GY through the first order traits were classified as second order traits. Estimates of genotypic and phenotypic correlation coefficient between GY and other agronomic and yield related traits were carried out using SAS version 9.3 version (SAS Institute, 2011). The Delta method was used to compute the standard errors as proposed by Holland (2006). The genotypic correlation greater than twice the value of its standard error was considered to be significant statistically (Kolawole et al., 2018).
3 RESULTS AND DISCUSSION

The genotypic and phenotypic correlations between grain yield, the primary trait of selection, and other agronomic traits for the MARS testcrosses under drought stress and well-watered conditions are presented in Tables 1 and 2 respectively. For some of the traits such as ear aspect it was not possible to estimate genotypic correlation because of the null values of genetic variance which was identified as 0.00. The results revealed that though the direction and magnitude of the estimates of both correlations were alike for almost all measured traits, the estimates of genotypic correlation were a little higher than those of the phenotypic correlation. This suggests the presence of environmental influence which is responsible for a reduction in estimates of phenotypic correlation despite the strong association between the traits considered. Similar results were reported by Gazal et al., (2018), who observed higher values for genotypic correlation compared with those of the phenotypic correlation.

Significant ($\rho \leq 0.01$) but negative genotypic and phenotypic correlations were observed between grain yield (GY) and flowering traits viz: anthesis silking interval (ASI), days to anthesis (DP) and silking (DS) under drought stress condition. This observation suggests that improvement in GY is associated with a reduction in ASI, DS, PA and EA under drought stress condition (Table 1).

Significant ($\rho \leq 0.01$) but positive genotypic and phenotypic correlations were observed between grain yield (GY) and plant height (PH), and also with ear height (EH). The association between GY and plant aspect (PA) well as between GY and ear aspect (EA) was significant and negative at both genotypic and phenotypic correlations under well-watered condition (Table 2). The improvement in the yield of grains can therefore be associated with an increase in the PH and EH under well-watered conditions which suggests that both traits are good predictors of GY. Lodging is however increased when selection is done for increased PH under non-stressed conditions and this will have a negative impact on GY (Talabi et al., 2017).

Association between ASI, DP, DS, PH, EA and GY under drought stressed and non-stressed conditions have been earlier reported (Bolanos and Edmeades, 1996; Ribaut et al., 1997; Messmer et al., 2009; Zheng et al., 2009 and Liu et al., 2011). Of all the secondary traits measured under drought stress condition only ASI showed significant association with GY at the genotypic level whereas these traits were weakly or not correlated under well-watered condition. This indicates that ASI is a secondary trait that provides the most important adaptive mechanism for drought tolerance in maize. The strong observed correlations of GY with ASI under drought conditions, was in line with the findings of Betran et al. (2003), Cattivelli et al. (2008). Bolanos and Edmeades (1996) reported strong genetic correlations between GY and ASI ($r = -0.60$) under drought stress and suggested that selection done for GY under drought stress was less efficient than selection for ASI.

The results of the stepwise multiple regression analysis under drought stress conditions showed that EPP, PA, EA, DS, LD and PH had a direct effect which was significant on grain yield with a contribution of about 71.1 % to the total observed variation (Table 3). EPP accounted for the largest proportion of the contribution to grain yield under drought stress (52 %) which is an indication that it is the most important trait in determination of grain yield under drought conditions. With a cumulative contribution of 71.1 % to the observed variation in grain yield, this suggests that these traits can be used in selection programmes as secondary traits for yield improvement under drought stress. Grain yield is therefore expected to increase in response to an increase in any of the traits. Badu-Apraku et al. (2012a) had earlier identified EA, PA, ASI, PH, EH, DS and EPP among some extra early maturing inbred lines as secondary traits to be considered during selection under drought conditions.

Under well-watered condition, EPP also had the highest contribution of 31.42 % to grain yield followed by EA, PA, DP, DS and PH which contributed 15.94, 5.37, 3.13, 10.7 and 0.037 % respectively with a cumulative total of 56.97 %. Similar to drought stress condition, a contribution of these traits was highly significant ($\rho \leq 0.01$) except DS which was significant at 5 % probability level. This is an indication of the level to which these traits are able to influence yield under well-watered condition. EPP has proven to be a trait to be considered when improvement is being carried out for grain yield under both drought stress and well-watered conditions (Bazinger et al., 2000).

Grain yield is complex being an effect of the interrelatedness of several other plant components that influences growth and development, thus deducing with reference to the method of accumulation, combination of its processes through its life cycle proves difficult (Anjum et al., 2011). Tadesse et al. (2018) hypothesized that selection based on simple correlation without taking into cognisance the direct effect of the one variable (independent) on the other (dependent) is ineffective in determining the actual interrelatedness among traits. The use of path coefficient analysis will therefore aid the breeder in deciphering the cause, consequences and importance of the variables therefore providing a more effective means of interpreting the association.

The partitioning of genotypic correlation coefficient with path coefficient analysis showed that DS, PH, PA,
Table 1: Estimates of genotypic (upper diagonal) and phenotypic (lower diagonal) correlation coefficient and standard errors between grain yield and other agronomic traits for testcrosses of F1 lines derived from three cycles of marker assisted recurrent selection evaluated under drought stress condition in 2014 and 2015.

<table>
<thead>
<tr>
<th>Days to anthesis</th>
<th>Days to silking</th>
<th>Anthesis silking interval</th>
<th>Plant height</th>
<th>Ear height</th>
<th>Husk cover</th>
<th>Plant aspect</th>
<th>Ear aspect</th>
<th>Ears per plant</th>
<th>Grain yield</th>
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<tr>
<td>1.00**</td>
<td>-1.00</td>
<td>0.80**</td>
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<td>1.00**</td>
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*,** Significant at 0.05 and 0.01 probability levels respectively.

Table 2: Estimates of genotypic (upper diagonal) and phenotypic (lower diagonal) correlation coefficient and standard errors between grain yield and other agronomic traits for testcrosses of F1 lines derived from three cycles of marker assisted recurrent selection evaluated under well-watered condition in 2014 and 2015.

<table>
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<th>Days to anthesis</th>
<th>Days to silking</th>
<th>Anthesis silking interval</th>
<th>Plant height</th>
<th>Ear height</th>
<th>Husk cover</th>
<th>Plant aspect</th>
<th>Ear aspect</th>
<th>Ear per plant</th>
<th>Grain yield</th>
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<tr>
<td>1.00**</td>
<td>-1.00</td>
<td>0.90**</td>
<td>-0.10**</td>
<td>0.00</td>
<td>0.00</td>
<td>-0.30**</td>
<td>0.00</td>
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<tr>
<td>1.00**</td>
<td>-1.00</td>
<td>0.90**</td>
<td>-0.10**</td>
<td>0.00</td>
<td>0.00</td>
<td>-0.30**</td>
<td>0.00</td>
<td>-0.20**</td>
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*,** Significant at 0.05 and 0.01 probability levels respectively.
EA, LD and EPP as having a direct effect on GY. These six first order traits were recognized as being important in contributing to the observed variation in GY as shown from the path coefficient analysis (Fig 1) thus signifying them as potential secondary traits used as a selection criterion under drought stress. These findings corroborate those of Talabi et al. (2016) who identified EA, EPP, PA, STGR, DS, EHT and SL as traits with direct significant traits affecting GY under drought stress conditions.

EPP had the highest direct effect (0.37) on GY followed by DS (0.22) under drought stress condition. However, only the effects of EPP and PH were positive while the other direct effects were negative. Four traits (DP, ASI, EH and HC) also made contributions to GY but the contributions were indirect through DS (DP and ASI), PH (EH and HC), PASP (DP and EH) and EPP (DP, ASI, EH). EH and HC also contributed indirectly to grain yield through LD. ASI a known secondary trait contributed to GY indirectly through the six first order traits indicating that it should be considered as trait of importance during selection under drought stress. DP had the highest indirect positive effect on GY through DS (0.74) followed by EH through PH (0.73). The high coefficient reported for EPP corroborates the findings of Badu Apraku et al. (2018) who reported the highest path coefficient for EPP among extra early maize cultivars under drought stress conditions.

Under well-watered condition, the first order traits include EPP, EA, PA, DP and PH (Fig 2) with EPP (0.56) having the highest direct effect among the traits followed by EA (-0.40). Similar to the result under drought stress, only EPP and PH had positive direct effect on GY while other first order traits were also negative. None of the second order traits which include DP, ASI, EH and HC made contributions through all the first order traits simultaneously. DS had the highest contribution to GY through DP (0.91) followed by the contributions of EH through PH (0.87).

Increased EPP together with some other traits had been identified as a secondary trait used as a selection criterion (Bazinger et al., 1997) under drought stress conditions, including PH which also had a direct positive effect on GY. Selection for increased EPP may be accomplished without an adverse effect under well-watered condition, but selection for increased plant height though may aid the plant to synthesize more assimilates resulting in production of more grains but usually at a price. Increased plant height usually predisposes the plant to lodging which adversely affects grain production. The proportion of the variation in the dependent variable that is influenced by the other variables (independent) is less than half under drought as reflected by the coefficient of determination. The high positive and direct effect of plant

### Table 3: Multiple stepwise regression of other agronomic traits on grain yield of marker assisted recurrent selection maize testcrosses evaluated under drought stress and well-watered conditions at Ikenne in 2014 and 2015

<table>
<thead>
<tr>
<th>Trait</th>
<th>Drought stress condition</th>
<th>Well-watered condition</th>
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<tr>
<td>Trait</td>
<td>R²</td>
<td>Percent contribution to grain yield</td>
</tr>
<tr>
<td>Ear per plant</td>
<td>0.52</td>
<td>607.47**</td>
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<tr>
<td>Plant aspect</td>
<td>0.61</td>
<td>142.45**</td>
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<tr>
<td>Ear height</td>
<td>0.67</td>
<td>89.32**</td>
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<tr>
<td>Days to silking</td>
<td>0.69</td>
<td>49.78*</td>
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<tr>
<td>Leaf death</td>
<td>0.70</td>
<td>18.21*</td>
</tr>
<tr>
<td>Plant height</td>
<td>0.71</td>
<td>15.59*</td>
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height on the yield corroborates the findings of Adesoji et al. (2015) who reported the highest positive and direct effect of plant height to grain yield in a maize population grown under legume incorporation and nitrogen. Therefore, the use of PH as a selection criterion may not be practical due to its non-beneficial effect on production of grains.

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4 CONCLUSION

It can be concluded that though varying results were obtained with the use of correlation with respect to the measured traits associated with grain yield under both conditions, similar results were however obtained for the MARS testcrosses under both conditions with the use of multiple stepwise regression and path coefficient analysis. The coefficient of determination however showed variation: with the use of regression the dependent trait influenced the first order traits more under drought condition but reverse was the case with the use of path analysis. Ear aspect, plant aspect and ears per plant were identified as secondary traits that will aid selection for improved yield of maize. Leaf death; which is not one of the observed traits under well-watered condition was also identified as a secondary trait under drought stress. These traits are rated based on phenotypic appeal therefore their determination is highly subjective. Extra caution should therefore be taken when scoring to increase precision.

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