Inheritance of stay green physiological determinants in drought tolerant maize inbred lines

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Received 16 June, 2021; Accepted 31 August, 2021

Stay green is the plant's ability to sustain photosynthetically functional green leaf area for longer periods, even with excessive water stress. This study was done to understand the gene action that controlled inheritance of the stay green trait with the general objective of contributing to breeding more drought tolerant maize varieties. 10 F1 hybrids from crosses between highly performing and the least performing drought tolerant inbred lines as regards selected stay green physiological determinants were planted under a rain out shelter with a well-watered control and excessive water-stress treatments applied from six weeks after planting and eight weeks after planting. With progress in days after flowering, data were collected on relative chlorophyll per unit leaf area (RC: LAUG), maximum quantum yield of photosystem II photochemistry (ΦII), non-photochemical quenching (NPQ) and linear electron flow (LEF). RC: LAUG and NPQ were mainly inherited by additive gene action. ΦII was controlled more by the environment than interallelic interactions while LEF inheritance was controlled mainly by interallelic interactions.

Key words: Additive, inheritance, non-additive, photosynthesis, stay green.

INTRODUCTION

Stay green is the ability of a plant to maintain functional photosynthetic leaf area for longer periods after physiological maturity, even under excessive shortage of water (Luo et al., 2006). Due to their high photosynthetic activity beyond physiological maturity, such plants have a higher deposition of assimilates in their sinks, increased resistance to diseases and pests, along with the pronounced tolerance to water stress (Thomas and Smart, 1993). Breeding programs targeting water stress prone ecologies make use of secondary traits such as stay green when selecting and advancing populations that are suitable for these limiting environments (Edmeades et al., 1997). Such programs make use of knowledge on gene actions controlling the inheritance of the traits to effectively design breeding approaches that can maximize the breeding value of selected germplasm.
A number of annual crop species inclusive of maize exhibit genetic variation for the degree of retention of leaf pigmentation under water stress (Thomas and Smart, 1993). However, little has been established about the inheritance of stay green ability in maize, which is a prerequisite for the successful improvement of cultivars for the trait (Belicuas et al., 2014). In other crops such as sorghum, green leaf area retention, was reported to be regulated by both dominant and recessive epistatic interactions (Tenkouano et al., 1993), with the level of dominant gene action dependent on the environment (Walulu et al., 1994).

The few studies on stay green inheritance in maize such as that by Belicuas et al. (2014), reported that both the additive and dominance variances for the trait were significant, with the additive variance exceeding the dominance variance by a factor of 6.41 therefore classifying the average level of dominance as partial dominance. Indeed, other studies such as those by Guei and Wasson (1996), Banziger et al. (2000), Lee et al. (2005) and Costa et al. (2008) also reported that inheritance of the stay green trait was controlled mostly by additive effects than non-additive effects. Joshi et al. (2007) also reported that the trait had medium to high heritability with strong contribution of additive effects. Traits controlled largely by additive effects have the advantage of being easily improved for in populations using conventional approaches, which are cheaper, and have established legal systems in the developing world (La Rovere et al., 2010).

Majority of the previous studies on inheritance of the stay green trait in maize, however, made use of visual scoring while assessing genotypes for stay green (Belicuas et al., 2014). The need for less biased data requires less individual-centered methods for assessing for the trait as raised by Borrell et al. (2008). Chlorophyll fluorescence-based methods have since been developed to overcome this problem such as those provided by the Minolta chlorophyll meter SPAD-502 (Inoue et al., 1988) and more recently, photosynQ (Kuhlert et al., 2016). These approaches, alongside providing un-biased information on visual leaf pigmentation, provide in-depth information about how physiologically functional the pigmentation is.

This is by assessing for rates of photosynthesis-related physiological processes such as non-photochemical quenching and linear electron flow under various environments, that way providing more useful information to the breeding programs during selection of germplasm. The parameters measured for by these chlorophyll fluorescence approaches have, however, not been studied for the gene actions controlling their inheritance, especially in tropical maize germplasm.

The objective of this study, therefore, was to explore the gene action that controlled the individual modes of inheritance of these parameters, in order to provide a better understanding of their heterosis, and of the contribution of individual parental lines to functional stay green expression in maize hybrids.

MATERIALS AND METHODS

Site of the study

The study was carried out at the National Crops Resources Research Institute (NaCRRI), Namulonge in Uganda, under a rain-out shelter. Namulonge is located in the Lake Victoria crescent area at 0.5288° N, 32.6123° E, at an altitude of 1204 m (Otum et al., 2018). The area had an average temperature of 21.1°C, with mean daily maximum and minimum temperatures of 27° and 17°C, respectively during the period of study (Uganda National Meteorological Authority, 2020).

Constitution of F1 hybrids

Five high performing inbred lines (CELO15027, CELO15034, CELO15031, CELO15018, CELO15046), were planted and mated with two low performing inbred lines (CELO15043, CELO15045), in regard to stay green physiological determinants as assessed by Epaku et al. (2021). These inbred lines were sourced from the International Maize and Wheat Improvement Center (CIMMYT) breeding programs in Kenya and Ethiopia. A North-Carolina II mating design was used producing 10 F1 hybrids. This was done under field conditions with each entry having one row constituting eight hills spaced at 75 x 30 cm. Three seeds were planted per hill and later thinned to one. N.P.K fertilizer (17:17:17) was applied at a rate of 12 g per hill twice; first at planting and when the plants were 65 cm high by side dressing.

Evaluation of resultant F1 hybrids

The resultant F1 hybrids were evaluated using a completely randomized design, with two replications and four plants for each hybrid under each replicate. The experiment comprised a well-watered (WW) treatment and two water-stress treatments that were applied starting at six weeks after planting (6WAP) and at eight weeks after planting (8WAP). The potting mixture for the WW treatment was maintained at a water potential between 0.003-0.006 MPa; while that for the water-stress treatments was maintained at a water potential between 0.015 and 0.02 MPa using a tensiometer (Zaidi et al., 2015). The procedure involved measuring the soil moisture at 10, 20 and 30 cm three times every seven days to determine the amount of water to be added for maintenance of the required water potential. N.P.K fertilizer (17:17:17) was applied at a rate of 12 g per pot twice; first at planting and when the plants were 65 cm high, by side dressing within the pot. The photosynthetically active radiation under the rain-out shelter was 450 to 570 nm supplied by sunlight with a 12 h light period. Ambient temperatures in the shelter were 27 to 32°C and 24 to 56% relative atmospheric humidity as measured with a multi-spectrophotometer (Kuhlert et al., 2016).

Data collection

Data were collected on maximum quantum yield of photosystem II photochemistry (ΦII), linear electron flow (LEF), non-photochemical quenching (NPQ) and relative chlorophyll (RC) using a multi-spectrophotometer operating on photosynQ version v.10 (Kuhlert et al., 2016). This was done at the base of the leaf lamina of the second and fourth leaves from the top of the plant, for three plants per entry, and their respective averages computed. For leaf area
under green (LAUG), one sided green leaf area of each genotype was measured by multiplying the length with the widest width of the longest green leaf per genotype, followed by dividing this result with the ground area occupied by each genotype (Wang et al., 2012). The ratio of RC to LAUG was then computed to obtain RC: LAUG. Data for all parameters were collected first at six weeks after emergence of the crop before commencement of the water stress treatments. NPQ was assessed again at 60 days after flowering (DAF) under all treatments; RC: LAUG was assessed again at 20, 40 and 50 DAF under WW, 8WAP and 6WAP treatments, respectively; ΦII was assessed again at 40 DAF under all treatments; while LEF was assessed again at 20, 40 and 50 DAF under WW, 8WAP and 6WAP treatments, respectively as suggested by Epaku et al. (2021).

Data processing and analysis

Data collected were analyzed using the linear model:

\[
Y_{ijk} = \mu + Mi + Fj + MFi + MEi + FEj + FMEije + Ee + R(E)kl + \epsilon_{ijk}
\]

(Comstock and Robinson, 1948).

where \(Y_{ijk}\) = value of F1 cross of the \(i^{th}\) female and the \(j^{th}\) male in the \(k^{th}\) replication as regards stay green; \(\mu\) = population mean; \(i = 1, 2, \ldots, n\) (\(n = 5\) males); \(j = 1, 2, \ldots, B\) (\(B = 4\) plants per entry); \(M_i\) = effect of the \(i^{th}\) male; \(F_j\) = effect of the \(j^{th}\) female; \(MFi\) = effect of interaction between \(i^{th}\) male and \(j^{th}\) female; \(\epsilon_{ijk}\) = error effect of the \(ijk^{th}\) observation.

General combining abilities (GCA) were computed as the difference between the general performance of a line across all crosses (\(y\)) and the population mean (\(y\)); (GCA\(_i\) = \(y_i\) - \(\bar{y}\)). The significance of the GCA effects were tested using a t-test where \(t = \frac{GCA_i}{SE_{GCA}}\). Specific combining ability (SCA) was computed as \((SCA_{ij} = GCA_i - g_i - g_j)\); where \(g_i\) = GCA of the \(i^{th}\) female; \(g_j\) = GCA of the \(j^{th}\) male.

Heritability was estimated in form of broad-sense coefficient of genetic determination (BS.CGD) which is the total genetic variation, and narrow- sense coefficient of genetic determination (NS.CGD) which is the proportion which can be explained by additive effects (Dabholkar, 1992).

Heritability estimates across treatments were calculated as:

BS.CGD = \(\frac{(2\sigma^2 gca) + \sigma^2 sca}{(2\sigma^2 gca) + \sigma^2 sca + (\sigma^2 gca \times site + \sigma^2 sca \times site)/3 + (error/6)}\)

NS.CGD = \(\frac{2\sigma^2 gca}{(2\sigma^2 gca) + \sigma^2 sca + (\sigma^2 gca \times site + \sigma^2 sca \times site)/3 + (error/6)}\)

Heritability estimates within environments were calculated as:

BS.CGD = \(\frac{(2\sigma^2 gca) + \sigma^2 sca}{(2\sigma^2 gca) + \sigma^2 sca + error}\)

NS.CGD = \(\frac{2\sigma^2 gca}{(2\sigma^2 gca) + \sigma^2 sca + error}\)

Baker’s ratio (BR) was calculated using variance components due to GCA and SCA as:

BR = \(\frac{(2\sigma^2 gca)}{(2\sigma^2 gca) + \sigma^2 sca}\)

RESULTS

The results of the ANOVA for all the parameters are presented in Table 1. There was no significant variation for ΦII and RC: LAUG among genotypes under all treatments. LEF significantly varied among males, females and male*female interactions (P≤0.05) under the 6WAP treatment while under the 8WAP treatment it varied between females and male*female interactions (P≤0.05). NPQ showed significant variation (P<0.001) among males under the 6WAP and 8WAP treatments.

The results of the combining ability analysis and variance components for RC: LAUG are shown in Table 2. All the genotypes had insignificant GCA for RC: LAUG under all treatments. The cross CELQ15045×CELQ15046 had positive significant SCA (P≤0.05) for RC: LAUG under the 8WAP treatment. The trait generally had a low heritability across treatments but with a higher heritability under the well-watered treatment than in the water stress treatments.

The results of the combining ability analysis and variance components for NPQ are shown in Table 3. All the genotypes had insignificant GCA and SCA for NPQ under all treatments. GCA for NPQ was higher under the water-stress treatments. CELQ15018 and CELQ15043 showed positive GCA in all the treatments. CELQ15018 and CELQ15043 had positive GCA for NPQ under all treatments. Heritability for NPQ was higher under the water stress treatments than in the WW treatment.

The results of combining ability analysis and variance components for ΦII are shown in Table 4. All the genotypes had insignificant GCA and SCA for ΦII under all treatments. GCA for ΦII was generally higher under the WW treatment. CELQ15031, CELQ15046 and CELQ15045 had positive GCA for ΦII under all treatments. Cross CELQ15045×CELQ15027 showed the highest positive SCA for ΦII under all treatments. The trait had no detectable narrow sense heritability under all treatments but a moderate broad sense heritability under the water-stress treatments.

The results of combining ability analysis and variance components for LEF are shown in Table 5. All the genotypes had insignificant GCA for LEF under all treatments. GCA and SCA for LEF were generally higher under the water stress treatments than in the WW treatment. CELQ15031 had the highest positive GCA in all the environments.

CROSSES CELQ15045×CELQ15046, CELQ15043×CELQ15031 and CELQ15045×CELQ15031 had positive significant SCA (P≤0.05) for LEF under the WW, 6WAP and 8WAP treatments, respectively. The trait also had a higher broad sense heritability especially under the water stress treatments.

The results for the correlation studies between mean performance of genotypes for particular traits and their GCA for those respective traits are shown in Table 6. Mean ΦII had a significant (P<0.001) positive correlation with GCA for RC: LAUG. Mean LEF had a significant (P≤0.05) positive correlation with GCA for NPQ. Mean LEF had a positive insignificant correlation with GCA for ΦII. The means of all parameters apart from ΦII had a positive significant (P<0.001) correlation with the GCAs of the genotypes for those respective traits.
Table 1. Mean square analysis of variance among F1 hybrids for stay green physiological determinants inclusive of maximum quantum yield of photosystem II photochemistry (ΦII), linear electron flow (LEF), non-photochemical quenching (NPQ) and relative chlorophyll to leaf area under green ratio (RC: LAUG).

| Parameter | Source of variation | d.f | WW | 6WAP | 8WAP |
|-----------|---------------------|-----|-----|------|------|
| RC: LAUG  | Genotype            | 9   | 6,591 | 1,107,496,840 | 504,783 |
|           | Male                | 4   | 1,542 | 1,128,714,760 | 684,471 |
|           | Female              | 1   | 1    | 2,077,656,520 | 1,099,112 |
|           | Male: Female        | 4   | 13,287 | 843,739,001 | 176,513 |
|           | Residual            | 9   | 7,973 | 1,004,993,631 | 693,548 |
| NPQ       | Genotype            | 9   | 0.042 | 0.119* | 0.119* |
|           | Male                | 4   | 0.047 | 0.219** | 0.219** |
|           | Female              | 1   | 0.00046 | 0.0823 | 0.0823 |
|           | Male: Female        | 4   | 0.048 | 0.029   | 0.029  |
|           | Residual            | 9   | 0.047 | 0.034   | 0.034  |
| ΦII       | Genotype            | 9   | 0.011 | 0.023   | 0.054  |
|           | Male                | 4   | 0.0006 | 0.027   | 0.041  |
|           | Female              | 1   | 0.045 | 0.002   | 0.019  |
|           | Male: Female        | 4   | 0.013 | 0.024   | 0.075  |
|           | Residual            | 9   | 0.052 | 0.071   | 0.049  |
|           | Genotype            | 9   | 2.474 | 8,319*  | 5,142*  |
|           | Male                | 4   | 3.666 | 10,219* | 4,420  |
|           | Female              | 1   | 1.414 | 11,940* | 6,796*  |
|           | Male: Female        | 4   | 1.546 | 5,513*  | 5,451*  |
|           | Residual            | 9   | 3,502 | 1,995   | 1,926  |

**Significant at 1%, *significant at 5%, d.f= degree of freedom; WW= well-watered; 6WAP= six weeks after planting water-stress; 8WAP= eight weeks after planting water-stress; RC:LAUG=relative chlorophyll per unit leaf area under green; NPQ= non-photochemical quenching; ΦII= maximum quantum yield of photosystem II photochemistry; LEF= linear electron flow.

Source: Authors

Table 2. Combining ability and variance components for RC: LAUG under the three treatments.

| Variable | Genotype | WW | 6WAP | 8WAP | 6WAP + 8WAP | Across treatments |
|----------|----------|----|------|------|-------------|------------------|
| CELQ15027 | 4.18     | 17965 | -458 | 8753 | 5836        |
| CELQ15034 | -1.81    | -12215 | 524  | -5845 | -3897       |
| CELQ15031 | 30.49    | -12403 | 330  | -6036 | -4014       |
| CELQ15018 | -11.3    | -12177 | -271 | -6224 | -4153       |
| CELQ15046 | -21.56   | 18831  | -125 | 9352  | 6228        |
| CELQ15043 | 0.23     | -10192 | -234 | -5213 | -3475       |
| CELQ15045 | -0.23    | 10192  | 234  | 5213  | 3475        |
| CELQ15043×CELQ15027 | 55.67 | -12381 | 33.62 | -6174.01 | -4097.44 |
| CELQ15045×CELQ15027 | -55.67 | 12381  | -33.62 | 6174.01 | 4097.44 |
| CELQ15043×CELQ15034 | -77.01 | 10907  | 183.15 | 5545.09 | 3671.06 |
| CELQ15045×CELQ15034 | 77.01   | -10907 | -183.15 | -5545.09 | -3671.06 |
| CELQ15043×CELQ15031 | 47.92   | 9939   | -2.48 | -4968.27 | 3328.15 |
| CELQ15045×CELQ15031 | -47.92  | -9939  | 2.48  | -4968.27 | -3328.15 |
| CELQ15043×CELQ15018 | -41.63  | 10547  | 136.63 | 5341.94 | 3547.41 |
| CELQ15045×CELQ15018 | 41.63   | -10547 | -136.63 | -5341.94 | -3547.41 |
| CELQ15043×CELQ15046 | 15.04   | -19011 | -350.94* | -9681.29 | -6449.18 |
| CELQ15045×CELQ15046 | -15.04  | 19011  | 350.94* | 9681.29  | 6449.18 |
Table 2. Contd.

| Variance components and heritability | Genotype variance | Male variance | Female variance | Male×Female variance | GCA variance | Bakers' ratio | Additive variance | Dominance variance | Environmental variance |
|-------------------------------------|-------------------|--------------|----------------|---------------------|-------------|---------------|------------------|-------------------|-----------------------|
|                                     | 0.00              | 0.00         | 0.00           | 2656.99             | 0.00        | 0.00          | 0.00             | 0.00              | 3986.87               |
|                                    | 51251605.14       | 71243939.9   | 12391751.9     | 0                   | 9100790.67  | 0             | 365203162.7     | 10628              | 502496815.3           |
|                                    | 0                 | 126898.54    | 92259.89       | 0                   | 113631.99   | 1             | 454527.96       | 0                 | 346774.49             |
|                                    | 13213031.33       | 14408619.95  | 32776849.07    | 1                   | 21473323.46 | 1             | 85893293.82     | 0                 | 150442057             |
|                                    | 5756348.22        | 6473807.36   | 14606319.83    | 0                   | 9601696.77  | 1             | 38406787.09     | 0                 | 67167781.78           |

*Significant at 5%, WW= well-watered; 6WAP= six weeks after planting water-stress; 8WAP= eight weeks after planting water-stress.
Source: Authors

Table 3. Combining ability and variance components for NPQ under the three treatments.

| Variable       | Genotype | WW     | 6WAP   | 8WAP   | 6WAP + 8WAP | Across treatments |
|----------------|----------|--------|--------|--------|-------------|------------------|
| GCA CELQ15027  | 0.00     | -0.18  | -0.18  | -0.18  | -0.12       |                  |
| CELQ15034      | -0.16    | -0.02  | -0.02  | -0.02  | -0.07       |                  |
| CELQ15031      | -0.01    | 0.35   | 0.35   | 0.35   | 0.23        |                  |
| CELQ15018      | 0.01     | 0.09   | 0.09   | 0.09   | 0.06        |                  |
| CELQ15046      | 0.15     | -0.24  | -0.24  | -0.24  | -0.11       |                  |
| CELQ15043      | 0.00     | 0.06   | 0.06   | 0.06   | 0.04        |                  |
| CELQ15045      | 0.00     | -0.06  | -0.06  | -0.06  | -0.04       |                  |
| CELQ15043×CELQ15027 | 0.04 | -0.12  | -0.12  | -0.12  | -0.07       |                  |
| CELQ15045×CELQ15027 | -0.04 | 0.12   | 0.12   | 0.12   | 0.07        |                  |
| CELQ15043×CELQ15034 | -0.17 | -0.02  | -0.02  | -0.02  | -0.07       |                  |
| CELQ15045×CELQ15034 | 0.17  | 0.02   | 0.02   | 0.02   | 0.07        |                  |
| CELQ15043×CELQ15031 | 0.04  | 0.06   | 0.06   | 0.06   | 0.05        |                  |
| CELQ15045×CELQ15031 | -0.04 | -0.06  | -0.06  | -0.06  | -0.05       |                  |
| CELQ15043×CELQ15018 | -0.02 | 0.11   | 0.12   | 0.11   | 0.06        |                  |
| CELQ15045×CELQ15018 | 0.02   | -0.11  | -0.12  | -0.11  | -0.06       |                  |
| CELQ15043×CELQ15046 | 0.12   | -0.03  | -0.03  | -0.03  | 0.02        |                  |
| CELQ15045×CELQ15046 | -0.12 | 0.03   | 0.03   | 0.03   | -0.02       |                  |
| SCA CELQ15027  | 0.00     | 0.04   | 0.04   | 0.05   | 0.02        |                  |
| CELQ15034      | 0.00     | 0.05   | 0.05   | 0.05   | 0.02        |                  |
| CELQ15031      | 0.00     | 0.01   | 0.01   | 0.01   | 0.00        |                  |
| CELQ15018      | 0.00     | 0.00   | 0.00   | 0.01   | 0.00        |                  |
| CELQ15046      | 0.00     | 0.03   | 0.03   | 0.03   | 0.01        |                  |
| CELQ15043      | 0.00     | 1.00   | 1.00   | 0.00   | 0.00        |                  |
| CELQ15018      | 0.00     | 0.13   | 0.13   | 0.13   | 0.05        |                  |
| CELQ15046      | 0.00     | 0.00   | 0.00   | 0.03   | 0.01        |                  |
| CELQ15043      | 0.02     | 0.02   | 0.02   | 0.01   | 0.02        |                  |
| CELQ15046      | 0.03     | 0.88   | 0.88   | 0.95   | 0.78        |                  |
| CELQ15043      | 0.00     | 0.88   | 0.88   | 0.79   | 0.68        |                  |

WW= well-watered; 6WAP= six weeks after planting water-stress; 8WAP= eight weeks after planting water-stress.
Source: Authors

DISCUSSION

Maximum quantum yield of photosystem II photochemistry (ΦII) inheritance was predominantly under the control of the environment with a small percentage controlled by inter-allelic interactions. This suggested that where-as
Table 4. Combining ability and variance components for ΦII under the three treatments.

| Variable | Genotype | WW   | 6WAP | 8WAP | 6WAP + 8WAP | Across treatments |
|----------|----------|------|------|------|-------------|------------------|
|          |          |      |      |      |             |                  |
| GCA      | CELQ15027| -0.02| 0.05 | 0.08 | 0.07        | 0.04             |
|          | CELQ15034| 0.01 | -0.08| -0.13| -0.10       | -0.07            |
|          | CELQ15031| 0.01 | 0.08 | 0.08 | 0.08        | 0.06             |
|          | CELQ15018| 0.01 | -0.10| -0.09| -0.09       | -0.06            |
|          | CELQ15046| 0.00 | 0.05 | 0.05 | 0.05        | 0.03             |
|          | CELQ15043| -0.05| -0.01| -0.03| -0.02       | -0.03            |
|          | CELQ15045| 0.05 | 0.01 | 0.03 | 0.02        | 0.03             |
|          | CELQ15043×CELQ15027 | -0.08 | -0.11 | -0.15 | -0.13   | -0.11 |
|          | CELQ15045×CELQ15027 | 0.08 | 0.11 | 0.15 | 0.13   | 0.11 |
|          | CELQ15043×CELQ15034 | 0.01 | -0.03 | -0.13 | -0.08   | -0.05 |
|          | CELQ15045×CELQ15034 | -0.01 | 0.03 | 0.13 | 0.08   | 0.05 |
|          | CELQ15043×CELQ15031 | -0.01 | 0.01 | 0.03 | 0.02   | 0.01 |
|          | CELQ15045×CELQ15031 | 0.01 | -0.01 | -0.03 | -0.02   | -0.01 |
|          | CELQ15043×CELQ15018 | 0.00 | 0.10 | 0.15 | 0.13   | 0.09 |
|          | CELQ15045×CELQ15018 | 0.00 | -0.10 | -0.15 | -0.13   | -0.09 |
|          | CELQ15043×CELQ15046 | 0.08 | 0.03 | 0.10 | 0.06   | 0.07 |
|          | CELQ15045×CELQ15046 | -0.08 | -0.03 | -0.10 | -0.06   | -0.07 |

Variance components and heritability

|          | Genotype variance | Male variance | Female variance | Male×Female variance | GCA variance | Baker's ratio | Additive variance | Dominance variance | Environmental variance | Broad sense heritability | Narrow sense heritability |
|----------|-------------------|---------------|-----------------|----------------------|-------------|---------------|-------------------|---------------------|-------------------------|---------------------------|--------------------------|
|          | 0.00              | 0.00          | 0.00            | 0.00                 | 0.00        | 0.00          | 0.00              | 0.00                | 0.03                    | 0.67                      | 0.00                     |

WW= Well-watered; 6WAP= six weeks after planting water-stress; 8WAP= eight weeks after planting water-stress.

Source: Authors

Table 5. Combining ability and variance components for LEF under the three treatments.

| Variable | Genotype | WW   | 6WAP | 8WAP | 6WAP + 8WAP | Across treatments |
|----------|----------|------|------|------|-------------|------------------|
|          |          |      |      |      |             |                  |
| GCA      | CELQ15027| -25.36| -45.12| -32.12| -38.62      | -34.2            |
|          | CELQ15034| -37.66| -15.68| -2.68 | -9.18       | -18.67           |
|          | CELQ15031| 31.63 | 81.16 | 54.16 | 67.66       | 55.65            |
|          | CELQ15018| 8.23  | 13.75 | 1.75  | 7.75        | 7.91             |
|          | CELQ15046| 23.16 | -34.1 | -21.1 | -27.6       | -10.68           |
|          | CELQ15043| -8.41 | 24.43 | 18.43 | 21.43       | 11.49            |
|          | CELQ15045| 8.41  | -24.43| -18.43| -21.43      | -11.49           |
|          | CELQ15043×CELQ15027 | -14.7 | -30.04| -24.04| -27.04      | -22.93          |
|          | CELQ15045×CELQ15027 | 14.7  | 30.04 | 24.04 | 27.04       | 22.93            |
| SCA      | CELQ15043×CELQ15034 | 0.06  | -5.55 | 0.45  | -2.55       | -1.68            |
|          | CELQ15045×CELQ15034 | -0.06 | 5.55  | -0.45 | 2.55        | 1.68             |
|          | CELQ15043×CELQ15031 | -1.61 | 62.76*| 63.76 | 63.26*      | 41.63*           |
heterosis could be exploited to improve germplasm for this trait as reported by Zhang et al. (2000), a great deal of the performance of the resultant genotypes had to do with how favorable the environment they were exposed to was in terms of photosynthetic efficiency. Malik et al. (1999) however, reported that inheritance for net-photosynthesis was mainly under the control of additive gene effects, probably owing to the different methods of measurement for the trait used in their study. The significant positive correlation between mean NPQ and GCA for RC: LAUG suggested that these two traits could be selected for simultaneously to hasten the breeding process (Banziger et al., 2000).

Generally, the variation for NPQ and LEF among the F1 hybrids was more under the 6WAP treatment followed by the 8WAP typically owing to the relative length of periods of exposure to water-stress (Efeoğlu et al., 2009). The variation for NPQ only under the water-stress treatments implied that the trait could best be selected under this treatment. Additionally, variation for this trait was mainly due to additive effects implying that it could effectively be improved by re-current selection among germplasm (Badu-Apraku et al., 2011). Selection for high mean NPQ among lines could imply selection for lines with a high GCA for LEF (Banziger et al., 2000). NPQ had a moderate heritability across environments with higher heritability under the water-stress treatments affirming the need to select for it under water-stress treatments (Badu-Apraku et al., 2013). Crosses CEL15043×CEL15031, CEL15045×CEL15034 and CEL15045×CEL15046 showed positive though insignificant SCA for this trait suggesting that these crosses could be studied in different environments to further understand and exploit their heterosis (Bekavac et al., 2007). This heterosis suggested that where-as inheritance for NPQ was mainly additive, a significant percentage of it was under the control of inter-allelic interactions. Similar findings were reported by Holá et al. (2010) that non-additive gene effects were just as important as additive gene effects in the inheritance of photosynthetic parameters.

The F1 hybrids showed significant variation for LEF only under the water stress treatments, with the highest heritability recorded under the same suggesting that

Table 5. Contd.

| Variance components and heritability | Variance | Heritability |
|-------------------------------------|----------|-------------|
| Genotype variance                   | 3161.92  | 0.52        |
| Male variance                       | 1176.52  | 0.52        |
| Female variance                     | 642.7    | 0.52        |
| Male×Female variance                | 1759.07  | 0.52        |
| GCA variance                        | 971.21   | 0.52        |
| Baker’s ratio                       | 0.52     | 0.52        |
| Additive variance                   | 3884.82  | 0.52        |
| Dominance variance                  | 7036.27  | 0.52        |
| Environmental variance              | 997.82   | 0.52        |
| Broad sense heritability            | 0.92     | 0.52        |
| Narrow sense heritability           | 0.33     | 0.52        |

*Significant at 5%, WW=well-watered; 6WAP=six weeks after planting water-stress; 8WAP=eight weeks after planting water-stress.
Source: Authors

Table 6. Correlation between mean trait performance of genotypes and their general combining abilities.

| Correlation | GCA RC: LAUG | GCA NPQ | GCA ΦII | GCA LEF |
|-------------|--------------|---------|---------|---------|
| RC: LAUG    | 1**          | 1**     | 0.58**  | 0.963** |
| NPQ         | -0.81        | 0.963** | 1       | 0.055   |
| ΦII         | 0.58**       | 0.036   | 1       |         |
| L EF        | -0.76        | 0.963** | 0.055   | 1**     |

**Significant at 1%.
Source: Authors
inheritance for this trait would be better studied under similar environmental conditions. Heritability for this trait was largely due to dominance variance implying that it was mainly under the control of inter-allelic interactions and would be better exploited through heterosis rather than re-current selection (Rojas and Sprague, 1952). Particularly, the cross CELQ15043XCELQ15031 showed positive significant SCA for this trait under the water stress treatments suggesting that it was the best performing for this trait among the germplasm screened (Cairns et al., 2013). Crosses inclusive of CELQ15045XCELQ15027, CELQ15045XCELQ15046 and CELQ15045XCELQ15034 also showed positive though insignificant SCAs for this trait suggesting the need to screen them in even more environments for further exploration of their heterosis (Cukadar-Olmedo and Miller, 1997). Inbred lines CELQ15031, CELQ15018 and CELQ15043 also showed positive though insignificant GCA for LEF implying the need to cross them in even more diverse populations to further exploit their genetic ability to improve germplasm for this trait (Veeresha et al., 2015).

RC: LAUG was moderately under the control of additive effects under the water-stress treatments used in this study, a finding that was similar to those by Cukadar-Olmedo and Miller (1997), Joshi et al. (2007), Belicuas et al. (2014), Sayed and Bedawy (2016), and Annor and Badu-Apraku (2016) who reported that retention of leaf pigmentation was mainly under the control of additive gene effects. The significant SCAs for RC: LAUG in crosses CELQ15043XCELQ15046 and CELQ15045XCELQ15046 were in agreement with the findings by Mhike et al. (2012) and suggested that these crosses could be studied further for heterosis in regards to this trait. The high heritability for this trait under the well-watered treatment was also in agreement to the findings by Joshi et al. (2007) who reported heritability for leaf pigmentation between 0.73 and 0.80; but disagreed with the findings by Mhike et al. (2012), who reported that leaf pigmentation retention had a higher heritability under water-stress conditions, leaving the conflicting results unresolved.

The higher GCAs in water-stress treatments for all traits were an indication that these inbred lines had a higher breeding value for these traits under water-stress (Jiang et al., 2004). Broad sense heritability estimates for PHI, LEF and NPQ were higher than 0.5 suggesting the need for further selection for each trait (Sayed and Bedawy, 2016).

Conclusion

The study therefore, established that non-additive gene effects were just as important as additive gene effects as regards inheritance of the physiological determinants for the stay green trait, and that the effect the environment had on the inheritance of these traits should be taken into account. The crosses CELQ15045XCELQ15046 and CELQ15043XCELQ15031 exhibited positive and highly significant SCA effects and could be considered as good specific combiners for leaf pigmentation (RC: LAUG) and LEF across treatments.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

ACKNOWLEDGEMENT

This work was accomplished with financial support from the Regional Universities Forum for Capacity in Agriculture (RUFORUM) together with the World Bank through the African Centre of Excellence in Materials, Product Development and Nanotechnology (MAPRONANO ACE II).

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