

**GENE ACTION AND GENETIC MARKERS AS
ASSISTED SELECTION FOR DROUGHT
TOLERANCE IN GRAIN SORGHUM**

By

HEBA MOHAMED HAFEZ ESSA

B.Sc. Agric. Sc. (Agronomy), Ain Shams University, 2002

M.Sc. Agric. Sc. (Agronomy), Ain Shams University, 2010

**A thesis submitted in partial fulfillment
Of**

The requirements for the degree of

**DOCTOR OF PHILOSOPHY
in
Agricultural Science
(Crop Breeding)**

**Department of Agronomy
Faculty of Agriculture
Ain Shams University**

2016

ABSTRACT

Heba Mohamed Hafez Essa: Gene Action and Genetic Markers as Assisted Selection for Drought Tolerance in Grain Sorghum. Unpublished Ph. D. Thesis, Department of Agronomy, Faculty of Agriculture, Ain Shams University, 2016.

This investigation was conducted at Giza and Shandaweel Research Stations of the Field Crops Research Institute (FCRI), Agricultural Research Center (ARC), Egypt, in the seasons from 2011 to 2013. The study aimed to estimate some genetic parameters for grain yield and some related yield traits, using the six populations, *i.e.* P₁, P₂, F₁, F₂, BC₁ and BC₂ of two grain sorghum (*Sorghum bicolor* L. Moench) crosses, *viz.* ICSB-88005 × MR-812 (Cross 1) and ICSB-37 × ICSR-93002 (Cross 2). P₁, P₂, F₁, F₂, BC₁ and BC₂ of the two crosses were evaluated under two levels of watering [100 and 50% ET] in two separate experiments. Data regarding backcrosses and F₂'s under normal watering were not recorded due to the dissimilarity of its genetic make-up with those under drought. The investigation also aimed to study the relationships between the results of ISSR-PCR, RAPD-PCR and field results in an attempt to develop molecular markers for drought tolerance.

Results of analysis of variance indicated significant differences among the studied generations of each cross for studied traits. The genetic variances within F₂ population were also found to be significant for all the studied traits in the two crosses, therefore genetic parameters were estimated.

Some of the scaling test values of A, B, C and D were significant for different studied traits, suggesting the presence of non-allelic interaction. F₂ deviation (E1) and backcross deviation (E2) were significant, with few exceptions, retiring the contribution of epistatic gene action in the inheritance of the studied traits.

Results of generation mean analysis showed that the dominance \times dominance (dd) type of epistatic gene action, followed by the additive (a) gene effects and the additive \times dominance (ad) type of epistatic gene action contributed with the large part of genetic component controlling the inheritance of the studied traits compared to and the dominance gene effect (d) and the additive \times additive (aa) type of epistatic gene effect. Heritability estimates in the broad sense under drought conditions ranged from 68.95% for no. of green leaves/plant in cross I to 86.09% for days to heading in cross II. Heritability estimates in the narrow sense ranged from 19.14% for plant height to 59.21% for panicle width in cross II.

The expected genetic advances as percentage of the F₂ mean (Δg %) for the studied traits under drought conditions ranged from 3.03% for days to heading to 28.72% for panicle width in cross II. Results of path coefficient analysis revealed that 1000-grain weight, panicle weight and panicle length proved to be the major grain yield contributors.

Concerning genetic markers studies, the primers A-11, B-20, C-01, HB-11, HB-12 and HB-13 showed close relationship for drought tolerance segregates in the F₂ generation and thus they could be used as marker assisted selection for drought tolerance. Therefore, the ISSR-PCR and RAPD-PCR analysis could be considered as reliable molecular markers associated with drought tolerance in grain sorghum that can be utilized during breeding programs *via* marker-assisted selection.

Key words: Grain sorghum (*Sorghum bicolor* (L). Moench), Six populations, Drought tolerance, Scaling test, Gene action, Heritability, Genetic advance, Correlation, Path coefficient, ISSR-PCR and RAPD-PCR.

CONTENTS

| | Page |
|--|-------------|
| LIST OF TABLES | III |
| LIST OF FIGURES | VI |
| INTRODUCTION | 1 |
| REVIEW OF LITERATURE | 3 |
| 1. Effect of water deficit on growth and yield of grain sorghum. | 3 |
| 2. Heterosis, inbreeding depression and potence ratio. | 9 |
| 3. Genetic components of variance. | 15 |
| 4. Phenotypic and genotypic coefficients of variability (PCV and GCV), heritability and genetic advance. | 17 |
| 5. Correlation and path coefficient analysis. | 22 |
| 6. Molecular markers. | 27 |
| MATERIALS AND METHODS | 30 |
| A. Field experimental work | 30 |
| I. Genetic materials. | 30 |
| II. Experimental work. | 30 |
| III. Statistical and genetical analysis. | 34 |
| 1. Analysis of variance. | 34 |
| 2. Phenotypic and genotypic coefficient of variability. | 35 |
| 3. Genetic parameters estimated. | 35 |
| B. Molecular genetic markers. | 44 |
| RESULTS AND DISCUSSION | 49 |
| 1. Analysis of variance. | 49 |
| 2. Mean performance. | 53 |
| 3. Heterosis, inbreeding depression and potence ratio. | 60 |

II

| | Page |
|--|------|
| 4. Scaling test, F_2 -deviation (E1), backcross-deviation (E2) and types of gene action. | 67 |
| 5. Heritability estimates and expected genetic advance from selection. | 75 |
| 6. Correlation and path coefficient analysis studies | 81 |
| 7. Molecular genetic markers. | 88 |
| SUMMARY | 100 |
| REFERENCES | 109 |
| ARABIC SUMMARY | |

LIST OF TABLES

| No. | Pages |
|---|--------------|
| 1. Names, origin and drought stress response of the four grain sorghum pure lines used as parents in the study. | 30 |
| 2. Amount of irrigation water (m ³) applied for each irrigation treatment based on ET% in 2013 season. | 32 |
| 3. Analysis of variance for each experiment and expectations of mean squares (EMS). | 34 |
| 4. List of the primer names and their nucleotide sequences used in the study for ISSR and RAPD procedure. | 48 |
| 5. Mean squares of parents and F ₁ 's for studied traits under normal conditions for the two crosses. | 51 |
| 6. Mean squares of the six populations for studied traits under drought stress conditions for the two crosses. | 52 |
| 7. Means (X) and variances (S ²) of the six populations under drought conditions and parents and F ₁ 's under both normal (N) and drought (D) conditions and drought susceptibility index (DSI) for the studied traits in grain sorghum crosses. | 58 |
| 8. Mid- and better parent in heterosis (%) and inbreeding depressing (I.D. %) for studied traits, using six populations data in the two grain sorghum crosses under drought stress condition. | 65 |
| 9. Potence ratio (P) and degree of dominance for studied traits, using six populations data in the two grain sorghum crosses under drought stress condition. | 66 |
| 10 Estimates of scaling tests, F ₂ deviation (E1) and backcross deviation (E2) for studied traits, using six population means in the two grain sorghum crosses under drought condition. | 69 |

- 11** T-test of differences between parents, F-test for significant of genetic variance among F_2 plants and types of gene effects for studied traits, using six populations means in the two grain sorghum crosses under drought stress condition. **74**
- 12** Variances, heritability estimates in broad (h_{bs}) and narrow (h_{ns}) sense and genetic advance after one generation of selection for the best 5 % of the F_2 population (Δg) and in percentage of F_2 mean ($\Delta g \%$) for studied traits, using six populations data in the two grain sorghum crosses under drought stress condition. **80**
- 13** Phenotypic correlation coefficients between grain yield per plant and six contributing traits in F_2 populations data of the grain sorghum cross (ICSB-88005 \times MR-812) under drought stress conditions. **83**
- 14** Phenotypic correlation coefficients between grain yield per plant and six contributing traits in F_2 populations data of the grain sorghum cross (ICSB-37 \times ICSR-93002) under drought stress conditions. **84**
- 15** Phenotypic path coefficient of grain yield per plant and its contributing characters, using the six populations data in the two grain sorghum crosses under drought stress conditions. **87**
- 16** Phenotypic components (direct and joint effects) in percent of grain yield per plant variation using the six populations data in grain sorghum crosses under drought stress conditions. **87**
- 17** Grouping of F_2 plants into groups according to grain yield per plant in the two grain sorghum crosses. **92**

- 18** RAPD-PCR and ISSR-PCR fragments of the six tested primers with sensitive ICSB-88005 (1) and moderately tolerant MR-812 parental lines (2), relatively tolerant F₁ (3), the most low yield of F₂ (4) and the most high yield of F₂ plants (5) for grain sorghum cross (ICSB-88005 × MR-812). **93**
- 19** RAPD-PCR and ISSR-PCR fragments of the six tested primers with sensitive ICSB-37 (6) and moderately tolerant ICSR-93002 parental lines (7), relatively tolerant F₁ (8), the most low yield of F₂ (9) and the most high yield of F₂ plants (10) for grain sorghum cross (ICSB-37 × ICSR-93002). **96**

LIST OF FIGURES

| No. | | Pages |
|------------|--|--------------|
| 1. | RAPD-PCR and ISSR-PCR profile of sensitive (1) and moderately tolerant parental lines (2), relatively tolerant F ₁ (3), the most low yield of F ₂ (4) and the most high yield of F ₂ plants (5) for the grain sorghum cross (ICSB-88005 × MR-812) and ISSR-PCR , RAPD-PCR profile of sensitive (6) and moderately tolerant parental lines (7), relatively tolerant F ₁ (8), the most low yield of F ₂ (9) and the most high yield of F ₂ plants (10) for the grain sorghum cross (ICSB-37 × ICSR-93002) with primers A-11, B-20, C-01(RAPD), HB-11, HB-12 and HB-13(ISSR). (M=marker and ISSR= Randomly amplified polymorphic). | 99 |