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Estimating of genetic components by using line x tester for earliness in some rice genotypes under water stress condition

By

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Abstract

In the majority of agricultural ecosystems across the globe, water stress is one of the leading causes of crop production decline. Breeding for drought tolerant genotypes is an important strategy to overcome this constraint. The degree of tolerance developed to confront water stress were examined through analysis variance, combining ability, heterosis, genetic parameter and heritability for all 22 selected trails in two environments. Also, the behavior of the selected rice genotypes was determined through biological, anatomical, and molecular responses under water stress. According to the results of the field during seasons of study, tremendous genetic variability among genotypes under normal and water stress conditions for various characters were observed. The additive genetic variance played the most important role in the inheritance of the traits and the selection might be practiced successfully in early generations. Based on the positive response of the biochemical analyses, genotypes Sakha 107× Sakha super 300 and Sakha 107 × M206 were considered tolerant genotypes for water stress. The promising crosses had higher values of several root-, stem-, and leaf- anatomical parameters than their parents when they were stressed by a water deficit. Molecular analyses were done by 10 ISSR primers, the two crosses recorded the highest similarity value and are also grouped in a distinct cluster. Also, under drought stress, the relative expression of four genes were significantly elevated in the most two drought-tolerant rice genotypes.

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Summary

The present investigation was carried out at the experimental of Rice Research and Training Center (RRTC), Sakha Agricultural Research Station during 2020-2021. Tolerance of developed rice genotypes to confront water stress was evaluated through analysis of variance, combining ability, heterosis, genetic parameters and heritability for 22 selected traits at two environment (normal and drought) conditions. The experimental materials, included six genotypes *i.e.* Giza 177, Sakha 105, Sakha 106, Sakha 107, Sakha 108, GZ 10101-5 and three testers *i.e.* Hispagran, Sakha super 300 and M 206. The six lines were crossed with the three testers to produce 18 F_1 crosses obtained by mating in L x T design during 2020 growing season. The experiment was conducted in a randomized block design with three replications. The evaluation of the parents and their F_1 crosses were done in 2021 summer season. F_1s, developed by three best crosses and their parents, were taken into account for further studies (biochemical, anatomical and molecular analysis). The results obtained have been summarized as follows:

1- Through the analysis of variances for various characters, tremendous genetic variability was observed among genotypes. The mean squares due to parents for the characters were significant. The mean squares due to hybrids were also highly significant for all characters under study. This indicated to the existence of considerable amount of genetic variability among parents and crosses for all traits.The parents *vs*. crosses comparison was significant for all characters indicating substantial amount of heterosis in crosses.

2- None of the parents or crosses showed consistent high performance for all characters. The parents Giza177, Hispagran and Sakha 107

along with cross combinations; Giza 177 x Hispagran, Sakha 107 x Hispagran, Sakha 107 x M 206 and Sakha 108 x M 206 were the earliest genotypes. While, Sakha 105, Sakha 108, Sakha107 x Hispagran and GZ10101-5 x Sakha super 300 gave the lowest values for plant height under normal and water stress conditions. Giza 177, Sakha Super 300, Giza 177 x Hispagran, Sakha 107 x Sakha super 300 occupied first position for milling percentage, head rice, grain thickness trails. Likewise, the parents Sakha 107, Sakha 108, and M 206 along with crosses, Sakha 107 x M 206, Sakha 107 x Sakha super 300, and Sakha 108 x M 206 recorded the best values in remaining studied trails with high differences compared with all other, lines, testers and crosses, respectively.

3- Highly significant positive values of GCA effects would be of interest in most traits under study, except for days to Days to 50% flowering, days to maturity and plant height whereas the highly significant negative values would be useful from the breeder's point of view. Only Giza 177 and Hispagran and GZ10101-5 possessed negative (desirable) GCA effects for the above mentioned trails.

4- Among all parents, Sakha107, Sakha108, M206 and Sakha super 300 exhibited high GCA effects for all the remaining trails and were observed as good general combiners for yield and most of its contributing characters.

5- None of the crosses was consistently superior for all the traits in estimates of specific combining ability (SCA). Four crosses; GZ10101-5 x Sakha super 300, Sakha 107 x M 206, Sakha 107 x Hispagran and Sakha 108 x Hispagran recorded negative, significant and desired SCA effects for days to 50 % heading, days to maturity and plant height traits. While, Sakha107 x Sakha super 300, Sakha 107 x M 206, Sakha 108 x Sakha super 300, Sakha 108

x M 206 manifested significant, desired SCA effects and were the best specific combination for most studied traits.

6- Among the crosses, three crosses *i.e.* Sakha 107 x M 206 (good x good), Sakha 107 x Sakha super 300 and Sakha 108 x M206 (average x good) had significant desired SCA effects and significant desired heterotic response over mid parent in desirable direction for most important characters.

7- When studying combining ability analysis, it was observed that both additive and non- additive type of gene action played an important role in the expression of all the traits except grain filling rate. Also, high values were determined in broad sense for all traits. On the other hand, high narrow sense heritability values were estimated for these traits.

8- High estimates of additive component of variance in comparison with the dominance genetic variance with flag leaf area, flag leaf angle, relative water content, number of panicle per plant, panicle length, panicle weight, number of spikelets per panicle, grain yield per plant, harvest index, grain thickness and grain shape traits. On the other hand, high estimates of dominance component of variance in comparison with the additive genetic variance were recorded for days to 50% heading, days to maturity, plant height, number of tillers, spikelets fertility, 1000 grain weight, hulling, milling, head rice and amylose content trails.

9- The additive genetic variance as a portion of the total genetic variance was larger than the dominance genetic variance. So, the selection for most trails might be practiced successfully in early generations using the pedigree method to improve most important traits under study and develop the new promising rice crosses.

10- Evaluation of cross for heterosis breeding based on mean performance, mid-parent heterosis and combining ability effects indicated that, out of 18 cross rice combinations, three cross combinations namely, Sakha 107 x M 206, Sakha 107 x Sakha super 300 and Sakha 108 x M 206 recorded significantly positive values and were most promising for grain yield and most of desirable traits and were selected to further studies.

11- Biochemical changes were assessed in the selected promising crosses; Sakha 107 x M 206, Sakha 107 x Sakha super 300 and Sakha108 x M206 leaves under normal and drought conditions to determine the drought effects on the biochemical processes of rice plants. Activities of ascorbate peroxidase (APX), superoxide dismutase (SOD), catalase (CAT) and total phenol contents were significantly increased with varied extents under drought stress compared to normal irrigation. The highest activities were recorded on the crosses Sakha107 x M 206 and Sakha 107 x Sakha super 300, superior to the parents in all values.

12- Significant enhancements in proline accumulation were noted with Sakha 107 x M 206 and Sakha 107 x Sakha super 300 compared to the remaining crosses and parents. In all parents and crosses under drought stress, total chlorophyll concentration more decreased than the normal conditions. However, in Sakha 107 x M 206 and Sakha 107 x Sakha super 300 crosses, total chlorophyll concentration increased significantly as compared to the parents under drought conditions.

13- The differential response of the promising rice crosses compared to their parents were studied anatomically to describe the effect of drought stress on some anatomical changes in structure of roots, stem and leaf of these crosses. Although a sign of shrinkage has been seen in all root samples under drought stress, the crosses Sakha 107 x M 206 and Sakha 107 x Sakha super 300 crosses

showed larger root diameter compared to their parents. Anatomical parameters such as thickness of epidermis (μ), thickness of cortex (μ) and vascular cylinder (μ) increased also in the promising crosses compared to their parents. However, xylem vessel numbers increased in the parents; Sakha 107 and M 206. Xylem vessel diameters (μ) in promising crosses were markedly increased and better than the parents.

14- The stem cross-sectional, pith and xylem vessel diameter also increased in the crosses Sakha 107 x M 206 and Sakha 107 x Sakha super 300 crosses as compared with the parents. In addition, the epidermis thickness, cortex thickness increased significantly in the crosses compared to the parents. The vascular bundle number in the stems of crosses stem were unchanged significantly compared to the parents. However, a significant induction in the vascular bundle diameter was observed in the promising crosses stems.

15- The leaf thickness and mesophyll thickness increased significantly in the crosses Sakha107 x M 206 and Sakha 107 x Sakha super 300 crosses compared to the parents. For the vascular elements, a significant induction of the xylem and vascular bundle diameter of the two crosses compared to the parents were found. For the mid-vein parameters, a significant induction in the mid-vein leaf thickness of crosses were also found. Also, the bundle sheath thickness was significantly induced in the leaf mid-vein of the promising crosses compared to the parents.

16- Genetic diversity were analyzed in this study to genetically select the most promising drought-tolerant genotypes (parents and crosses). The total number of alleles, produced by the ten primers, was 82 including 44 polymorphic markers and 38 monomorphic markers. A high polymorphism of 92.30% was scored for primer 3. No polymorphism of was scored for primers 6, 7, 9 and 10. The other primers produced polymorphism ranging between 42.85%

and 77.77%. The total percentage of polymorphic markers for all primers in the selected seven genotypes is 53.33%, which indicates to low levels of genetic variation among the selected genotypes. The primer ISSR-5 amplified the highest number of ISSR loci (14 bands) but primers ISSR-3 gave the highest percentage 92.30 % of polymorphism.

17- Cluster analysis separated the selected genotypes into two main clusters. Sakha108 occupied the first main cluster. Meanwhile, second main cluster of genotypes contained two sub clusters. First sub- cluster consisted of Sakha 108 x M 206. The second sub-cluster was divided into two groups, Sakha 107 x M 206, Sakha 107 x Sakha super 300 and Sakha107 grouped together in first group and two parents Sakha super 300, M 206 grouped together in second group. The similarity coefficient ranged between 88.2 % and 78.1 %. Maximum similarity was observed between crosses Sakha107 x M 206 and Sakha 107 x Sakha super 300 (88.2%). The second highest similarity 88.10 % was between the cross Sakha 107 x M 206 and their parent Sakha107 and the third highest similarity (88.0%) was between Sakha 107 x Sakha super 300 and Sakha 107. Also, large distances were found among between other genotypes, these results were substantiated by the fact that these genotypes have different origin with high level of genetic variation.

18- The relative expression of osACS2 (encodes1-aminocyclopropane-1-carboxylic acid synthase, ethylene key biosynthesis enzyme) was investigated in the three promising crosses Sakha 107 x M 206, Sakha 107 x Sakha super 300 and Sakha 108 x M 206 along with a drought- sensitive as a check cross Giza 177 x Hispagran under normal and drought conditions. It was found that OsACS2 was highly responsive to drought stress compared to the normal condition with Sakha 107 x M 206 and Sakha 107 x Sakha super 300 cross. For the gene OsCML31 which encodes the calcium

signaling protein calmodulin-related sensor protein. The two rice crosses Sakha 107 x M206 and Sakha 107 x Sakha super 300 were able of elevating the expression of CML31 significantly more than control conditions.

19- The expression of OsCYP94C2a gene (Cytochrome P450 of the subfamily CYP94 subclade C member 2a) was quantified as well. Both Sakha 107 x M 206 and Sakha 107 x Sakha super 300 rice crosses were able of sharpening the expression of OsCYP94C2a in response to water stress compared to normal conditions. In addition, the expression of OsCYP94C2a in Sakha 107 x M 206 and Sakha 107 x Sakha super 300 was the highest under drought stress in these two crosses compared to Sakha 108 x M2 06 and Giza 177 x Hispagran. The expression of OsSRO1c (encodes Similar to Radical-induced cell death One protein) was also quantified and found to be significantly up-regulated in all investigated rice crosses in response to drought stress. The expression of OsSRO1c was extremely higher in drought-stressed Sakha 107 x M 206 and Sakha 107 x Sakha super 300 plants rather than Sakha108×M206 and Giza 177×Hispagran crosses relative to corresponding control plants.

Conclusion

As a results of this study, it cold be concluded that:

1- Tremendous genetic variability among genotypes under normal and water stress conditions for various characters were observed through the analysis of variances.

2- The Additive genetic variance played the most important role in the inheritance of the traits under study. So, the selection for most trails might be practiced successfully in early generations.

3- Sakha 107 x M 206 and Sakha 107 x Sakha super 300 diagnosed as drought tolerant crosses, by using statistical analysis parameter, biochemical and anatomical parameters as well as genotypic levels and may be exploited in practical plant breeding program. The same may be evaluated critically to judge its superiority in performance with stability across the years and soil type for its utility on commercial scale.