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RESEARCH PAPER

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Mutation breeding approach to breed drought tolerant maize hybrids

Saif-u-Malook¹, Saeed Ahmad Qaisarani², Muhammad Khalid Shabaz⁴, Hafiz Ghulam Muhu-Din Ahmed¹, Muhammad Arslan Nawaz¹, Muhammad Sarfaraz⁵, Ghulam Mustafa⁵, Qurban Ali^{1,3}

Department of Plant Breeding and Genetics, University of Agriculture Faisalabad, Pakistan

²Department of Environmental Sciences, COMSATS Institute of Information Technology, Vehari, Punjab, Pakistan

³Center of Excellence of Molecular Biology, University of the Punjab, Lahore, Pakistan

*Department of Agronomy, Bahauddin Zakariya University, Multan, Pakistan

Department of Agronomy, University of Agriculture, Faisalabad, Pakistan

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Abstract

Crop production worldwide is focus to increasing environmental limitations, particularly to drought due to its high degree of impact and wide distribution. Conventional breeding approaches are trying to improve abiotic stress tolerance in crop plants, which have had some improvement, but are restricted by the quantitative nature of the traits. Maize is an important cereal crop. It is used as raw material in food, medicine and textile industries for the manufacturing of corn oil, corn flakes, dextrose, textile dyes livestock and poultry. Drought seriously affects plant growth from seedling to maturity. Mutation also proved to be tremendously important for evaluating functions for individual drought tolerant genes due to the availability of knock-out mutants and its docility for genetic transformation. To produce hybrids it is needed to collect information about germplasm diversity, combining ability and heterotic pattern that is essential in maximizing the effectiveness of the breeding programs. It will be helpful in selection of genotypes with improved yield. When genes for resistance to a particular disease or stress cannot be found in the available gene pool then mutation induction is the best alternative. In this review, the response of plant to drought and mutation are described, the regulatory routes which allow plants to manage with drought are presented, and how the present information can be useful to obtain stress tolerant plants.

^{*}Corresponding Author: Saif-u-Malook 🖂 saifulmalookpbg@gmail.com

Introduction

Maize (Zea mays L.) is the third most important grain crop of the world and covers 4.8% of the total cropped area (Saleem et al., 2008). Maize is used as food for humans and feed for livestock and poultry. It is used as raw material in food, medicine and textile industries for the manufacturing of corn oil, corn flakes, dextrose and textile dyes (Ali et al., 2011 and Saif-Malook et al., 2014e). Its grain constitutes about 9.7396 % grain protein, 4.85% grain oil, 9.4392% grain crude fibre, 71.966% grain starch, 11.77% embryo while fodder contains 22.988% acid detergent fibre, 51.696% neutral detergent fibre, 28.797% fodder cellulose, 40.178% fodder dry matter, 26.845% fodder crude fibre, 10.353% fodder crude protein and 9.095% fodder moisture (Ali et al., 2014abc). Maize accounts for 2.20 % of value of agriculture and 0.5% to GDP. Its area of production in Pakistan is 1085 thousand hectares with annual production of 4631 thousand tons and yield per hectare is 4268 per hectare. Production increased 6.8 % over last year due to use of hybrid varieties at large area (Anonymous, 2013). Maize is mainly affected by many biotic and abiotic factors. Drought badly affects plant growth from seedling to maturity (Areous et al., 2005 and Saif-ul-Malook et al., 2014abcde). Drought is a second factor after soil infertility that causes reduction in maize grain yield. Maize is more susceptible to drought as compared to other cereals except barley (Banziger and Araus, 2007). Genetic variation of useful traits is required for crop improvement. When a certain variation is lacking, mutagenic agents such as radiations and certain chemicals can be used to induce mutations and generate genetic variation from which desired mutants can be selected. When genes for resistance to a particular disease or tolerance or stress cannot be found in the available gene pool then mutation induction is the best alternative (Novak and Brunner, 1992). Selection of genotypes for improvement can be enhanced by studying correlation of yield and yield related components (Panwar et al., 2013). Direct or indirect effect caused by component traits on grain yield can be calculated through path analysis (Dogan, 2009). To produce hybrids it is need to collect information about germplasm diversity, combining ability and heterotic pattern that is essential in maximizing the effectiveness of the breeding programs. Combining ability and heritability analysis are useful tools to select better inbred lines for the development of desirable hybrids. There are highly significant differences among yield and yield related traits like ear length, kernel rows per ear, weight of 100 grains and grain yield observed by parents and their crosses. General combining ability as well as specific combining ability were also observed by using Line x tester analysis (Rahman *et al. 2013*; Ali *et al.* 2013; *et al.* 2014c).

Combining Ability analysis

Camacho and Caraballo (1994) reported significant differences in varietal means for all the traits like leaf area per plant, plant height, weight of dry root and weight of dry shoot under drought conditions in maize. The root dry weight was considered as a best indicator for drought tolerance in maize. Aguiar et al. (2003) conducted an experiment using different inbred lines to calculate general combining ability (GCA) as well as specific combining ability to evaluate hybrids of inbred lines in different environments. The interaction between environment and genotype and additive effects of gene action were significant for plant height, grain yield and ear height, while nonadditive gene effects were also significant for above mentioned traits. Abdelmula and Sabiel (2007) found that the effects of drought stress was significant for traits like plant height, time to 25% silking and grain yield. Plant height showed genetic advance and heritability. Plant height, leaf area and leaves per plant were correlated in a positive manner with grain yield. They found that leaves per plant and stem diameter might best selection criteria for improvement in maize. Dadheech and joshi (2007) concluded that dominant genes have more importance for inheritance of traits in maize like protein contents, starch contents and grain yield. Iqbal et al. (2007) showed significant specific

lqbal et al. (2007) showed significant specific combining ability and also found that non additive genes were more important for traits as number of kernels per ear, 1000 grain weight and number of

kernels per row in maize. Aliu *et al.* (2008) reported that leaf area was under the control of additive and non-additive genes and also concluded that if GCA of inbred lines was higher than SCA of these inbred lines might be higher or might not be higher.

Akbar et al. (2009) reported that the inheritance of some traits was controlled by non-additive genes like grain yield. Additive genes were responsible for days to 50% maturity. Kanagarasu et al. (2010) reported that GCA variances were less than SCA variances for the traits like cob girth, plant height, 100 grain weight, plant yield, cob length, rows per ear and leaf length in maize. This showed that dominance was predominant for the traits. Khalil et al. (2010) performed an experiment to evaluate single cross hybrids of maize for the production of double cross hybrids in maize and concluded that grain yield depends on both general combining ability as well as specific combining ability, So it shows both additive as well as non-additive effects of gene. Best parent selection criteria for hybridization is higher GCA but for the development of double cross hybrids is higher SCA. Wali et al. (2010) revealed that variance due to single crosses were significant for yield and yield contributing traits like kernels per row, weight of 100 grains, number of kernel rows per ear and grain yield. GCA effects was less than the SCA effects for kennels per row and weight of 100 grains and reported predominant non additive gene effects for inheritance of these traits in maize.

Ahsan et al. (2011) reported that shoot length and weight of root were significantly and positively correlated with weight of shoot. Stomatal frequency and epidermal cell size were also significantly and negatively correlated with fresh shoot weight, therefore shoot length, weight of root and epidermal cell size could be used as best selection criterion for high yield of shoot weight under water deficit conditions. Mhike et al. (2011) revealed significant results for GCA effects for yield contributing traits except plant height and stem lodging. They also showed that non additive genes were predominant for all traits such as plant height, anthesis dates and

grain yield. Pavan et al. (2011) showed that SCA variances were higher than GCA variances for specific traits as plant height, days to 50% silking, ear girth, ear length, number of kernel in ear, grain yield per plant, 100 grain weight, which revealed predominant relationship of non-additive gene effects in maize. Abuali et al. (2012) showed that yield and its component traits had high SCA than GCA for yield and yield contributing traits except number of kernels per row, cob girth, number of rows per ear and harvest index for maize. Afshar and Heidari. (2012) explained the SCA for some traits as plant height, ear length and grain yield and showed the predominance non-additive gene action and epistatic gene effects for the traits in maize. Haddadi et al. (2012) revealed that number of rows per ear, kernel weight, days to silking and cob to ear ratio had more GCA variance than SCA variances. It indicated that additive gene effects have more importance for these traits in maize. There was significant positive correlation between kernel yield and other yield components as kernel length, kernels per ear row and weight of kernel. Therefore these are suitable selection criteria for improvement in yield. Maseka and Ishaaq (2012) reported high GCA effects for some traits like cob length and grain yield in maize. Abrha et al. (2013) conducted an experiment and evaluated parents and crosses by line x tester analysis and reported that different genotypes showed significant differences in mean squares for different traits like kernels per row, number of rows per ear, ears per plant, plant height and grain yield and also mentioned that general combining ability of lines showed significant differences in mean squares for all traits but general combining ability of testers showed significant differences in mean squares only for ear height and grain yield.

Stress breeding

Chen *et al.* (1996) reported genetic association of leaf rolling rate, stomatal conductance, anthesis-silking interval, number of leaves, plant height, harvest index and leaf angles with drought resistance. Heritability estimates under drought conditions were low as compared to normal conditions. Bruce *et al.* (2002) evaluated that average yield was increased due to

better nitrogen application, tolerance to water deficient environment, continual sensitivity to input supply and better weed control. Pollination and early grain filling was much sensitive and highly effected due to drought. It was cleared that tropical germplasm exposed less number of spikelets, early vigorous silking and decreased anthesis-silking interval (ASI) after selection to drought tolerant superior maize genotypes. New molecular marker and ample gene expression profiling method gave the chances for continuous crop breeding that give positive results under variable environments. Qayyum et al. (2003) described that water stress is an environmental factor which decreases growth and ultimately yield of crop. It was suggested that significant genetic variation, heritability and better performance of various quantitative traits under water stress may be helpful to improve grain yield. It was also concluded that hybrids H₃, H₈, H₁₁, H₁₅, H19, H27 and H29 gave higher yield under drought. Khan et al. (2004) evaluated maize genotypes for seedling traits under normal and drought conditions. Higher genotypic coefficients of variance were observed for dry shoot weight, dry root weight, emergence percentage, fresh shoot weight and fresh seedling weight. Under drought conditions genetic variation and heritability estimates were high than normal. Overall best performance was observed for genotypes P2-55, P2-284, P2-286, P2-290 and P2-309 under normal and drought conditions.

Abdelmula *et al.* (2007) evaluated fifteen maize genotypes to determine the genotypic and differential responses of growth and yield at three different drought conditions. Grain yield was decreased due to water stress at reproductive stage while there was no effect on vegetative traits. G-3 genotype performed better than others and PR-1 was susceptible to drought at D3 level. A positive and significant correlation was observed between leaf area index and leaves per plant while positive but non-significant correlation was recorded between grain yield per plant with plant height and stem diameter. Ahsan *et al.* (2011) evaluated twenty five genotypes for the determination of physio-genetic behavior of maize

under drought conditions. Higher significant differences were found among genotypes 20P2-1, L5-1, 150P2-1, 70NO2-2, 150P1 and L7-2 than others. Fresh shoot length and fresh root weight were directly associated with fresh shoot weight while positively correlated with fresh shoot weight. It was suggested that increased fresh shoot length, fresh root weight and decreased stomata frequency and epidermal cell size may be useful criteria for selection under drought conditions.

Ali et al. (2011a) evaluated maize genotypes for seedling traits under moisture stress condition. It was concluded that highest heritability for chlorophyll content, positive and direct association of fresh root length with other seedling traits indicated that selection may be helpful in developing higher grain yielding maize genotypes. Ahsan (2013) found higher heritability and genetic advance for 100-seed weight, grains per plant and grain yield per plant in maize. Ali et al. (2011b) performed an experiment on forty maize genotypes under 60% moisture contents. It was found that chlorophyll content, fresh and dry root and shoot weight showed higher heritability, genetic advance and strong genotypic correlation was also found. High and positive direct effects on shoot length were recorded for chlorophyll contents, fresh root and shoot weight. It was concluded that selection on the basis of shoot length, fresh root and shoot weight and chlorophyll contents may be helpful to improve grain yield of maize.

Farhad *et al.* (2011) evaluated maize hybrid for drought tolerance. FH 421, FH 810, Pioneer 32-F-10, Pioneer 32-W-86, Monsanto 919, Monsanto 6525, NK 8441 and SS 5050 maize hybrid for evaluation of drought tolerance were used. Monsanto 919 provided better performance for plant height, leaf area, water potential, osmotic potential, turgor potential and minimum relative saturation deficit. FH 810 was drought sensitive. Leaf water content was correlated with osmotic potential under drought condition. Correlation studies showed that plant height and leaf area directly and indirectly related with grain yield under drought conditions. Saleem *et al.* (2011)

reported significant differences among maize genotypes under drought. W-64-TMS and PB-7-1 inbred lines showed high genetic variation and heritability so these can be used for breeding program under drought conditions. Ali *et al.* (2012) evaluated the growth related seedling traits of maize accessions. It was reported that high values of heritability and genetic advance for fresh root and fresh shoot length and fresh root-to-shoot length ratio indicated that selection can be mad on the basis of these traits for higher yielding maize genotypes under drought conditions.

Chohan et al. (2012) reported partial dominance effect for cell membrane thermo-stability and net photosynthetic rate at seedling stage under drought conditions. Khodarahmpour, (2012) evaluated four germination traits of maize hybrids under four levels of osmotic potential. It was reported that germination and growth was reduced due to water shortage. The mean germination time become high with decreased in osmotic potential. Hybrid Simon gave better performance under drought. Moradi et al. (2012) evaluated eight hybrids for yield performance on the basis of grain yield, stress tolerant index, stress susceptibility index, tolerance index and mean productivity. The results showed that H6 gave better performance in irrigated conditions while H8 gave good result in drought conditions. It was concluded that H8 and KSC704 performed best under drought conditions. Ahsan et al. (2013) reported additive gene action for all yield related traits except stomata frequency and stomata size for which complete and over dominance effects were found.

Mutations Breeding

Alvi et al. (2003) studied eighteen maize hybrids to determine interrelationship between yield and its components using genotypic correlation and path coefficient analysis. Grain yield was found to be positively and significantly associated with all parameters. Number of kernels per row had maximum positive direct effect on grain yield followed by 1000-kernel weight, ear length and number of row per ear. Ear height had negative direct

effect on grain yield. The main yield components were 1000-kernel weight, ear length, number of rows per ear and kernel per row. Gao et al. (2004) evaluated effect of enhanced UV-B radiation on growth, yield and seed qualities of maize under field conditions. Maize yield, dry matter accumulation, chlorophyll a and b, protein content, sugar and starch were reduced. Hameed et al. (2008) evaluated effect of gamma rays on desi and kabuli chickpea seeds for traits such as germination, growth, seedling fresh weight, root and shoot length and their ratio. Irradiation doses were inversely related with germination percentage and growth rate of sprouts. Bello et al. (2010) studied correlation and path coefficient analysis of 10 maize varieties and their 45 F₁ hybrids. For grain yield with plant height, number of grains per ear, ear weight, days to 50% tasseling with plant and ear height, phenotypic and genotypic correlations were positive and significant. For grain yield with plant height, ear height and ear weight environmental correlation was positive significant. Highest direct effect of days to 50% silking, ear weight and number of grains per ear was revealed through path analysis. For improving maize varieties and hybrids for high grain yield, days to flowering, plant height, ear height, number of grains per ear and ear weight cold be important selection criteria. Khawar et al. (2010) studied effect of different levels of gamma rays on seeds of maize, wheat, chick pea and black eye beans. With the increase in dose level, shoot and root lengths were decreased in all samples. Seeds with doses of more than 2 kGy were not germinated.

Majeed *et al.* (2010) studied mean germination time, germination percentage, survival percentage, shoot and root length, number of branches, leaves per plant and fresh and dry weight of *Lepidium sativum* L. under different levels of gamma radiations. At higher doses of gamma rays, mean germination time was significantly affected and delayed while seed germination percentage was not affected significantly. At higher doses of gamma irradiation, other growth parameters showed declining tendency. Rafiq *et al.* (2010) reported correlation and path analysis studies

in a cross between ten inbred lines and three testers. Presence of large variability for all traits was revealed through analysis of variability parameters. Genotypic coefficient of variation with high heritability was found in grain yield, ear length, ear height, 100-seed weight and ear diameter. Significant correlation of ear diameter, 100-grain weight, ear length, rows per ear and grains per row with grain yield was revealed through genotypic correlation coefficient. Through path analysis, it was found that 100-grain weight exhibited highest direct effect on grain yield followed by grains per row, kernel rows per ear, ear length and ear diameter. Rahimi and Bahrani (2011) studied effect of gamma irradiation on agronomic characteristics and quality of fatty acids of two brassica cultivar. Measured traits were decreased with increasing irradiation level up to 300 Gy. At 100 Gy level, highest seed yield, plant height and1000-seed weight were obtained. Minimum amounts of measured traits were obtained at 500 Gy. Hedimbi et al. (2012) reported effect of ultraviolet-A and ultraviolet-B radiations on maize seedling under controlled conditions. There was reduction in concentration of chlorophyll a and b with the increase in exposure duration. By increasing time of exposure to ultraviolet-A and ultraviolet-B, number of leaves, stem diameter, plant height and seedling height were also reduced. In plants exposed to ultraviolet-B, there was more reduction in chlorophyll a and b.

Zarei et al. (2012) evaluated interrelationship among grain yield and related characters in 11 corn hybrids. Plant height, ear length, days to maturity, kernel length, number of kernels per ear and 100-grain weight had positive correlation with yield. Plant height and ear length showed highest correlation followed by 100-grain weight and kernels per row. Positive direct effect on grain yield was shown by ear length, kernel length, 100-grain weight and total number of kernels per ear. Number of total kernels had highest direct effect while 100-grain weight had highest indirect effect. Emrani et al. (2013) evaluated two cultivars of maize for different levels of gamma radiations. The traits studied were germination percentage, germination rate, root length, shoot

length and shoot diameter. Root and shoot growth of both cultivars was highest on third and fourth day at 400 and 600 cGy level of irradiation. At 800 cGy, traits showed significant reduction. Girija and Dhanavel (2013) studied effect of different levels of gamma rays on different quntitative traits of cowpea such as plant height, leaves per plant, branches per plant, days to first flower, clusters per plant, pods per plant, seeds per plant, hundred seed weight and yield per plant. There was more reduction for all characters at higher doses as compared to lower doses in M1 generation. The results revealed that different doses of gamma rays can be used effectively for creating variability for various quantitative traits. Marcu et al. (2013) studied effect of gamma radiation on dry maize seeds. With the increase in irradiation dose, germination percentage germination index, root length and shoot length were decreased. Plants obtained from seeds exposed to higher doses, survived for less than 10 days. Yadav and Singh (2013) evaluated two maize genotypes under 13 levels of gamma radiations for different characters such as water activity, seedling height, seedling fresh and dry weights, fresh and dry weights of roots and shoots and enzymatic activity. Seedling height increased at lower gamma irradiation level and reduced beyond 0.2kGy dose for both genotypes. Germination percentage was significantly changed in one genotype while other genotype was unaffected.

Conclusions

It was concluded that different doses of gamma rays can be used effectively for creating variability for various quantitative traits. High dose of gamma irradiation, other growth parameters showed declining tendency to breed drought tolerant genotypes. It was also determined that selection on the basis of shoot length, fresh root and shoot weight and chlorophyll contents may be helpful to improve grain yield of maize.

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