



Review

A study on marker aided variety and crop management for salt tolerance

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Accepted 09 January, 2017

Salinity is one of the major abiotic constraints for crop management in irrigated as well as rainfed areas and results to poor harvests. The yield reduction in salt affected soils can be overcome by soil reclamation or by improving salt tolerance in crops. The saline soils can be reclaimed by leaching the salt and cultivation of salt tolerance crops. Rice is known as a good reclamation crop. Salt tolerance is a complex polygenic trait. Insufficient knowledge about mechanism of its inheritance results to slow progress in its introgression into target crops. The role of traditional as well as marker assisted breeding approaches has been discussed for introgression of salt tolerance in crops in order to obtain better harvests.

Key word: Abiotic constraints, salinity, crop management, introgression, traditional breeding, marker assisted selection.

INTRODUCTION

Salt tolerance is generally a sustained growth of the plant in the soil environment impregnated with NaCl or other salt combinations. The available three categories of alkali, saline and saline-sodic soils are problem soils and are generally also termed as salt affected soils. The area affected by salinity in the world covers about 400 million hectares (Flowers et al., 1977), of which 54 millions are found in south and south east Asia (Akbar and Ponnampuruma, 1982). The alkali soils contain excess sodium cations on exchange complex and concentrated carbonates/bicarbonates of sodium with high pH ranging from 8.5 to 10.7 with a high sodium adsorption ratio and

poor soil structure. These soils are found in semi arid and moist climate with more than 550 mm rainfall. Rain water and irrigated water log on surface soil. Saline soils contain mainly SO_4^- and Cl^- of sodium, calcium, magnesium and carbonate of Ca^{++} and Mg^{++} with electrical conductivity (EC) $>4 \text{ dSm}^{-1}$, $<15\%$ exchangeable percentage (ESP) and friable surface in dry condition. The pH value of these soils are much lower (< 8.2) than that of sodic soils. In addition, saline-sodic soil have high EC ($>4 \text{ dSm}^{-1}$), pH (>8.5) and more than 15% ESP. Such soils turn compact after irrigation and uptake

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of nutrient is reduced, resulting to reduced growth of plants. Accumulation of some compatible solutes like proline has been observed under salt stress conditions and has been suggested to be part of the mechanism(s) that controls salt tolerance in plants. It has been reported that over-production of proline has improved salt tolerance in rice, however, its relative importance for tolerance and precise protective function during stress require further investigations (Bohnert and Jensen, 1996).

The available reviewed data indicated that among the technical constraints, abiotic constraints (salinity, drought) are more prominent and yield limiting in most of the crops than biotic constraints (Dey and Upadhyaya, 1996; Hossain, 1995). The problems of salinity has reached serious proportions in irrigated rice production in semi-arid and arid climates (Asch et al., 1997) in general, and in the Basmati rice belt (north-western regions of India and adjoining regions of Pakistan) in particular and is further increasing (Abrol and Sehgal, 1994). Even with good water management, seasonal increases in topsoil salinity can hardly be avoided (Raes et al., 1995).

Despite the fact that this large area is either barren or with very low productivity, there are only few instances where salt tolerant cultivars have been developed. These soils are generally found in arid and semiarid climate zone with rainfall less than 550 mm. Water through rain and irrigation is absorbed soon in these soils. Saline soils are usually developed under arid and semi arid climatic zones and also not uncommon in water logged soils. Other crops generally do not grow in these areas except rice, oat, barley, barseem and sugar beet. Among these, rice is one of the most suitable crops for saline soils in water logged conditions, although, it is usually considered to be moderately sensitive to salinity (Akbar et al., 1972; Maas and Hoffman, 1977; Mori and Kinoshita, 1987).

BREEDING APPROACHES FOR CROP IMPROVEMENT

The yield reduction in salt soils can be overcome by soil reclamation or by improving salt tolerance in target crops. Therefore, need for improvement in salt tolerance in rice is well understood (Nabors, 1990; Flowers and Yeo, 1995). Breeding for salt tolerance in crops is difficult and very slow due to complexity of this polygenic trait, insufficient knowledge about mechanism of inheritance of genes controlling the character, low genetic variance, conspicuous correlations among tolerance at different developmental, physiological stages and lack of efficient selection criteria (Akita and Cabuslay, 1988; Yeo et al., 1990). The crop's germplasm does have sources for salt tolerance trait. The traditionally conserved land races are considerably adapted to grow in saline environments. Such characteristics perhaps, have evolved during the course of evolution to fit in the stress environment for survival. For example, rice land races such as Pokka,

Nona-Bokra, Damodar, Dasal, Getu, etc are well adapted to grow in saline environments (Akbar et al., 1985).

Traditional breeding efforts made for introgression of complex traits like salinity tolerance have met with limited success and only a few salt-tolerant varieties (<40 cultivars in different crops) have been developed and released for commercial cultivation. Likewise, introgression of desirable traits into Basmati rice (aromatic rice with superfine grain/cooking qualities) is difficult by conventional breeding methods due to complex nature of Basmati rice grain quality traits and salt tolerance, and its poor combining ability with the other rice genotypes (Singh et al., 2000; Glaszmann, 1987). There are so many other reasons for slow progress of development of efficient salt tolerant varieties. For example, only two salt tolerant chickpea varieties WCG-1 (Sadbhawana), WCG-3 (Vallabh Kallar Chana-1) were developed at SVP University of Agriculture and Technology, Meerut, Uttar Pradesh and Karnal chana-1 was developed at CSSRI, Karnal, Haryana, India in the last decade. In Basmati rice, only one salt tolerant variety CSR-30 was developed at CSSRI, Karnal, Haryana, India during the same period.

In the last ten years, a rapid progress has been made towards the development of molecular marker technologies and their application in linkage mapping, molecular dissection of the complex agronomical traits and marker assisted breeding (Caetano-Anollós et al., 1998; Flowers et al., 2000; Jain and Selvaraj, 1997; Staub et al., 1996). Application of molecular marker technology can greatly enhance the efficiency and accuracy of breeding process (Singh, 2009). Molecular markers have proven to be useful in both basic and applied research such as DNA fingerprinting, varietal identification, diversity analysis, phylogenetic analysis, and marker assisted breeding and map based cloning of genes in rice (McCouch et al., 2001). Different types of DNA markers including microsatellite DNA markers (simple sequence repeats, SSRs), AFLPs and ISSRs have been successfully used for genotype identification, diversity analysis and gene/QTL analysis in rice including Basmati rice (McCouch et al., 2001; Nagaraju et al., 2002). DNA markers are of great importance in plant breeding, especially for the selection of polygenic traits because they have several advantages like no G x E effect, no epistatic effect, desired homozygous plants can be easily picked up, and greater reliability to distinguish the homozygous lines from others at an early stage/generation. Focusing on specific crop and problem, transgenic production and/or marker-assisted selection approaches may be developed and used to evolve stress-tolerant crop varieties. For important crops like rice, wheat, brassica, chickpea, etc. a map-based marker assisted *in situ* technology should be developed for precision breeding, as well as gene identification through molecular hybridization. The molecular techniques should be standardized for (i) exploitation of hybrid vigor using specific markers; (ii) search for and use of still unex-

ploited, trait related gene or gene blocks (QTLs); (iii) engineering of required biosynthetic pathways. Marker associated gene blocks relating to salt tolerance shall be identified using QTL techniques in landraces and progenitor species of crop plants. By development and deployment of map-based and marker-assisted technologies for precision breeding for improvement in major crops like rice and wheat for the trait, genes/QTLs have been mapped for several important agronomical traits such as tolerance against abiotic stresses including drought and salinity (Flowers et al., 2000; Forster et al., 2000; Kearsey and Farquhar, 1998; Lilley et al., 1996; Zhang et al., 1999). Among the abiotic stresses, maximum progress has been made towards the drought related traits and there have been only a few studies to map QTLs for salinity tolerance (Ali et al., 2000; Flowers et al., 2000; Koyama et al., 2001; Lang et al., 2001). Molecular markers such as RFLP (Ali et al., 2000; Zhang et al., 1995), RAPD (Foolad and Chen, 1998; Xie et al., 2000) and AFLP (Ali et al., 2000), have been used for QTL mapping for salinity tolerance in rice. Recently, a new class of PCR based markers, inter simple sequence repeat (ISSR) markers, has been widely used to determine intra- and inter-genomic diversity (Singh et al., 2008a, b, 2009), since they reveal variation within unique regions of the genome at several loci simultaneously (Blair et al., 1999; Zietkiewicz et al., 1994). ISSRs targets a subset of simple sequence repeats (SSRs) and amplifies the region between two closely spaced and oppositely oriented SSRs. Application of molecular marker technology can greatly enhance the efficiency and accuracy of breeding process. Molecular marker technologies have revolutionized the genetic analysis of crop plants and its application has been suggested for the molecular dissection of complex physiological traits such as salt tolerance (Flowers et al., 2000). In several crops, comprehensive molecular marker/linkage maps with a variety of DNA markers have been developed. Most of the maps have been developed using mapping populations, which include the recombinant inbred lines (RILs), double-haploid lines (DHLs), and backcross/F₂/F₃ families (Lang et al., 2001).

Genes/QTLs have been mapped for several important agronomical traits such as yield, quality and tolerance against abiotic stresses including drought and salinity (Flowers et al., 2000; Forster et al., 2000; Kearsey and Farquhar, 1998; Lilley et al., 1996; Zhang et al., 1999). Among the abiotic stresses, maximum progress has been made towards the drought related traits and there have been only a few studies to map QTLs for salinity tolerance (Ali et al., 2000; Flowers et al., 2000; Koyama et al., 2001; Lang et al., 2001; Xie et al., 2000; Zhang et al., 1995). Molecular markers such as RFLP (Ali et al., 2000; Zhang et al., 1995), RAPD (Foolad and Chen, 1998; Xie et al., 2000) and AFLP (Ali et al., 2000), have been used for QTL mapping for salinity tolerance in rice. In all, it is required (1) to construct the fine-maps of

important QTLs like *Saltol* and validation of candidate genes in the QTL region; (2) based on converging positional and functional data (2) to find out the physiological bases of salinity tolerance in different crops and (3) for the development of a precision marker-aided backcrossing (MAB) system to efficiently transfer the *Saltol* allele into popular varieties of different crops. In addition to other applications, genetic diversity is generally applied in crop improvement for development of synthetic and composite varieties of cross pollinated crops and development of hybrid varieties in both cross pollinated and self pollinated crops for the exploitation of heterosis to get higher levels of production and enhance productivity of crops in order to ensure food security to the fast growing human population. However, there is no evidence to indicate any relationship between molecular markers based genetic diversity and heterosis. The estimation of molecular marker based genetic diversity is accounted for only by base pair sequence, representing a very small part of DNA, and thus have no direct significant bearing upon expression of agronomical traits/component characters contributing towards the target character. On the other hand, estimation of genetic diversity is generally based on agronomical characters (Mahalanobis, 1936; Singh, 1985). Such agronomical traits compulsorily bear plus/minus effects towards target character directly or indirectly. Such agronomical traits jointly could represent most of the functional genomic DNA. Therefore, estimates of diversity based on these characters may provide genetically diverse parents which in nick combinations may result to significant exploitable heterotic effects (Singh, 2010). However, such specific combinations involving nick parents may be identified by using related molecular markers. Such nick cross combinations may well be exploited to obtain heterosis for salt tolerance. Thus, molecular marker technologies cannot be a substitute of traditional breeding techniques.

Variations in levels of alkalinity/sodicity are largely observed even in small soil patches. Such variations enhance the cost of local control and diminish the precision of the experiments required to be designed to serve the useful purposes in the present context. Therefore, designing elegant experiments required artificially developed soil plots. Maintenance of such artificial soils requires lot of resources, skill and time. Sometimes such facilities may go beyond the approach of common Indian scientist. The findings developed under such artificial conditions when transferred to field may sometimes lose their effectiveness and precision as was observed in laboratory conditions.

Participatory breeding (Singh, 2010) and shuttle breeding (Mallik et al., 2002) seems to be competent technologies to provide broader genetic base to such complex characters due to the fact that such genotypes are developed, subjecting the segregating populations to more actual environments for selection. In these technologies, F₃ and more fixed selections made from

segregating populations will be supplied to different selection zones. Yield is a complex character and its Heritability of yield is not significantly far from zero (Allard, 1960). Therefore, yield cannot be taken as criteria for selection of salt tolerance like characters whose expression is drastically retarded and therefore, special care is required for synthesis of selection criterion based on suitable component characters. However, traditional breeding efforts could perhaps be adequately made to exploit heterotic effects in order to improve the complex traits like salinity tolerance by involving the associated traits using the improved criteria. Nicked single cross combinations for negative heterotic effects for such associated traits, if obtained, could be of value. Such cross combinations may possibly be able to express consistent salt tolerance and be used for development of salt-tolerant varieties of a crop (Singh, 1985). The agricultural scientists may feel that traditional methods will only be opted to feed the fast growing population at least for the next two decades. Therefore, traditional technology for crop improvement requires attention for its improvement, refinement and revalidation.

In addition to breeding technology, crop management techniques under resource conservation merit special considerations. The underneath process of bio-tilling in zero-tilling in wheat and direct seeding techniques in aerobic rice cultivation, respectively prevents compactness of such problem soils after irrigation. It allows proper growth of the crops in problem soils resulting to better harvests (Singh, 2010). Participatory plant breeding also seems to have potential to make selections in the soils having saline patches at the actual sites (Singh, 2010). With one more crop management technique, the seed priming in wheat and rice promotes utilization of such soils by allowing the germination at faster rate before such soils turn compact.

Conclusion

Excessive salt stress is a major problem in vast areas of the world. Salinity in coastal areas fluctuates within the year, being high during the dry season because of tidal inundation and intrusion from underground water. Salinization also results from mismanagement of water for irrigation and drainage and is currently becoming a looming problem in inland areas worldwide, steadily leading to soil deterioration and eventually forcing poor farmers off their lands. About 10 million ha of agricultural lands in the world are believed to be lost annually to salinization (Pessarakli and Szabolcs, 1999). Such problems could be overcome primarily by water and crop management including promoting cultivation salt tolerant crops. During reproductive development, salt tolerant genotypes tend to exclude salt from flag leaves and developing panicles (Yeo and Flowers, 1986; Khatun et al., 1995). Although, these traits are essentially independent, none of the known salt tolerant landraces had

more than a few of them and considerable variation in their extent of expression exists among different in their extent of expression exists among different genotypes, suggesting the likelihood of identifying even better donors and alleles of useful genes. Pyramiding of contributing traits at seedling and reproductive stages is needed for developing resilient salt-tolerant cultivars. This could help to improve salinity tolerance beyond the present phenotypic range if selection is carried out independently for individual contributing traits or ultimately by tagging major QTLs and genes underlying these traits (Ismail et al., 2007).

Note: The paper was presented in Zonal seminar on “Physiological and molecular interventions for yield and quality improvement in crop plants” held on September 17-18, 2010 organized by Indian Society for Plant Physiology, IARI, New Delhi, India & Centre of Excellence in Agriculture Biotechnology, SVP University of Agriculture & Technology, Meerut, UP, India.

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