Approaches to Increasing Salt Tolerance in Crop Plants

4

Ratna Karan and Prasanta K. Subudhi

Abstract

Soil salinity is widely recognized as a major threat to global food security. Salinity and other abiotic stresses, which are expected to be more frequent in future due to disturbances in global climate, pose a serious challenge for plant scientists to ensure food supply for the growing world population. Several approaches have been advocated to address the salinity problem, but the most logical solution to maintain crop productivity in salt-affected areas is to enhance salt tolerance of crop plants. Due to the genetic and physiological complexity associated with salt tolerance, efforts to breed salt-tolerant plants have met with limited success. Although progress has been made in deciphering the genetic basis of salt tolerance, sustained efforts are needed to systematically dissect and utilize the natural variability in the available germplasm for improving crop adaptation in saline environments using modern genomics tools. Wide range of variability for salt tolerance in wild relatives, cultivars of major field crops, and halophytes offers bright prospect for discovery of superior salt-tolerant alleles for crop improvement. With an enhanced understanding of molecular mechanisms and the associated genes for component traits of salt tolerance, it would be possible to breed salt-tolerant plants using an integrated approach involving conventional breeding, physiological analysis, marker-assisted selection, and transgenic technology.

Keywords

Abiotic stress tolerance • Conventional breeding • Genomics • Marker-assisted selection • Mutagenesis • Quantitative trait loci • Transgenics

---

R. Karan • P.K. Subudhi (✉)
School of Plant, Environmental, and Soil Sciences,
Louisiana State University Agricultural Center,
Baton Rouge, LA 70803, USA
e-mail: psubudhi@agctr.lsu.edu

P. Ahmad and M.N.V. Prasad (eds.), *Abiotic Stress Responses in Plants: Metabolism, Productivity and Sustainability*, DOI 10.1007/978-1-4614-0634-1_4, © Springer Science+Business Media, LLC 2012

1 Introduction

Abiotic stresses such as drought, salinity, submergence, extreme temperatures, mineral toxicities, and deficiencies impair crop growth and productivity and threaten global food security (Gao et al. 2007; Witcombe et al. 2008). Among these stresses, salinity is impacting more farm lands worldwide due to poor quality irrigation water, inadequate drainage, salt water flooding of coastal land, and salt accumulation in dry areas (Kijne 2006). Around 21% of world's irrigated land is estimated to be affected by salinity (Ghassemi et al. 1995) and it continues to be a major problem in the arid and semi-arid regions. The negative impact of climate change on food crops is well recognized. Global temperatures are estimated to rise between 1.1°C and 6.4°C during the next century (IPCC 2007). The increased temperatures will disrupt weather patterns, leading to regular occurrence of floods, drought, and salinity. Melting ice caps and glaciers are expected to cause a rise in sea level (Wassmann et al. 2004; Melloul and Collin 2006), which may seriously affect crop productivity in coastal areas due to increased soil salinity.

World population is increasing at an alarming rate and it is expected to grow from 6 billion today to nearly 8.3 billion by 2030 (FAO 2010). With no prospect of expanding arable land due to urbanization, rapid industrialization, and water scarcity in many populous developing countries of the world (Rengasamy 2010), providing food security for the world population will require at least 57% increase in food grain production by 2050 (Wild 2003). Although most major crops have witnessed increased productivity in the past, productivity has been stagnant in recent years and replicating the success of the past to increase food production further may not be easy. Increasing salinity tolerance of the world's major food crops is an important goal of plant scientists as the world's population is increasing more quickly than the area of agricultural land to support it (FAO 2010). To alleviate the negative impact of salinity on food production, use of halophytic species, improved water management, and enhancing salinity tolerance in major field crops have been advocated. But improving adaptation of crop plants in saline environments remains a challenging task due to complex genetic basis of salt tolerance mechanisms. In this chapter, we discuss about different approaches for the development of salinity-tolerant crop plants to boost food grain production.

2 Effect of Salinity on Crop Plants

Salinity inhibits seed germination and alters the physiology and anatomy of the plants resulting in reduced crop yield. Salt accumulates in soil due to movement of salty water from adjacent areas during flooding or poor quality irrigation water. When water recedes after flooding, salt from lower soil profiles comes to the soil surface due to the capillary movement. Salinity is a soil condition characterized by a high concentration of soluble salts, mostly chloride and sulfates of sodium in the soil. Soils are classified as saline when the electrical conductivity (EC) is 4 dS/m or more, which is equivalent to approximately 40 mM NaCl and generates an osmotic pressure of approximately 0.2 MPa (USDA-ARS 2008). Both chloride and sodic salts cause damage to the root system of crops. The chloride-triggered injury is identifiable by the extensive leaf blade scorching symptoms whereas the accumulation of sodic salts results in leaf mottling and leaf necrosis. Soil salinity creates both osmotic and ionic stresses in plants. Presence of salts in the soil solution reduces the ability of the plant to take up water, and this leads to reduction in the growth rate, referred as the osmotic or water-deficit effect of salinity. If an excessive amount of salt enters the plant in the transpiration stream, it causes injury to cells in the transpiring leaves, resulting in further reduction in plant growth. This is called the salt-specific or ionic effect of salinity (Greenway and Munns 1980). Salinity stress is finally quantified as a decrease in water potential. Plants resist low water potential in different ways by accumulating osmolytes and modifying the properties of cell walls through production of protective proteins (Verslues et al. 2006). Plants differ greatly in their tolerance to salinity, as reflected in their different growth responses. Most of the commonly used food crops are sensitive to salinity (Flowers and Colmer 2008). Among cereals, rice (*Oryza sativa*) is the most sensitive and barley (*Hordeum vulgare*) is the most tolerant. Bread wheat (*Triticum aestivum*) is moderately tolerant than durum wheat (*Triticum turgidum* ssp. durum). The variation in salinity tolerance in dicotyledonous species is even greater than in monocotyledonous species. Rice, wheat, and barley are the most extensively studied cereal crop plants for understanding the physiological and molecular basis of salt tolerance. The comprehensive survey of salt tolerance for crops and pasture species published by the US Salinity Laboratory (Maas and Hoffman 1977; USDA-ARS 2005), presents a threshold salinity below which there is no reduction in yield, and then a linear reduction in yield with increasing salinity. Salinity reduces the rate of leaf expansion, and closes stomata and thereby reduces photosynthesis, through the soil water deficit caused by the osmotic stress (Rahnama et al. 2010). Upon exposure to soil salinity, plants accumulate toxic concentrations of Na^+ in leaves, which impose an additional limitation to growth by reducing the longevity of photosynthetic tissues (Munns 2002). The control of Na^+ transport and its effective exclusion from the mesophyll cells of leaves is therefore an important requirement for salinity tolerance. Na^+ exclusion from leaves is associated with salt tolerance in cereal crops including rice (Asch et al. 2000; Haq et al. 2010), durum wheat (Munns and James 2003), bread wheat (Cuin et al. 2009, 2010), barley (Shavrukov et al. 2010), and its wild relatives (Garthwaite et al. 2005), tall wheatgrass (Colmer et al. 2006), and *Triticum tauschi*i (Schachtman et al. 1991). The major components that govern salt tolerance are reduced salt uptake or salt exclusion, enhanced K^+/Na^+ ratio, tissue tolerance, closure of stomata, upregulation of antioxidant system for protection against reactive oxygen species (ROS), synthesis of osmolytes, water use efficiency (WUE), early flowering, and vigorous growth to dilute the salt concentration in plant tissue (Colmer et al. 2005; Ismail et al. 2007).

3 Approaches for Developing Salt-Tolerant Crop Plants

3.1 Conventional Approach

3.1.1 Germplasm Screening and Classical Breeding

The success of the crop-breeding program largely depends on the availability of natural genetic variation among the germplasm resources. Large number of cultivated and wild germplasm in major crops, preserved in the Consultative Group on International Agricultural Research (CGIAR) institutions and national centers, provide unique resources for systematic screening for discovery of novel variability to improve adaptation of crop plants in saline environments. Particularly, the wild relatives, land races, and traditional cultivars are the potential reservoirs of novel alleles to improve abiotic stress tolerance. Accurate phenotyping procedures are critical for identifying useful germplasm for crop improvement program as well as for deciphering the genetic basis of the mechanisms associated with salinity tolerance. Several parameters for salinity tolerance are studied by growing the germplasm in a variety of culture techniques such as hydroponics, pot culture, and field screening. Plant materials are evaluated from germinating seeds through seedlings up to mature plants. Salinity causes not only ion toxicity and imbalance, but also reduces photosynthesis in plants. Classical screening methods are based on assessment of yield responses to salt stress. Although screening based on yield represents the combined genetic and environmental effects on plant growth and includes integration of the physiological mechanisms conferring salinity tolerance at the whole plant level, it is more convenient and practical if indirect indicators of salt tolerance can be employed at the whole plant, tissue, or cellular levels (Ashraf and Harris 2004). Faster screening methods can be employed for identification of potential parents in a breeding program through selection for high leaf K^+/Na^+ ratios in the presence of salinity, and high K^+/Na^+ discrimination that has been described as a physiological index for salinity tolerance in bread wheat (Dvorak et al. 1994), durum wheat (Munns et al. 2000), and rice (Asch et al. 2000).

Gregorio et al. (1997) used Standard Evaluation System (SES) for rice seedling salinity screening based on the percent of leaf damage, which in turn is used to assign the SES scores. These SES scores measure overall survival and/or vigor of the plant and are therefore good indicators of performance of the plant under stress. SES scores as well as low Na^+ uptake, K^+ uptake and low Na^+/K^+ ratio have been reported to be tightly linked to seedling salinity tolerance (Lee et al. 2003a, Lisa et al. 2004). Analysis of Na^+ uptake, K^+ uptake and Na^+/K^+ ratio of rice seedlings under salt stress, however, are difficult to measure in the large populations associated with breeding programs. Therefore, screening for salinity tolerance at seedling level using SES method is ideal.

Artificial salinized soil in pots or irrigation with saline water under field condition has been used in rice (Aslam et al. 1993). Absolute shoot fresh and dry weights along with percent mortality at seedling stage in rice under salinity stress revealed an efficient, reproducible, reliable, and simple method for assessing relative salinity tolerance in breeding program (Aslam et al. 1993). Physiological characters such as leaf area index (LAI), measurement of Na^+, K^+, and Ca^+ along with amount of photosynthetically active radiation (PAR) absorbed by plants under salinity stress have been used to evaluate salt tolerance in rice (Zeng et al. 2003b). Leaf area index (LAI) was found to be significantly contributing to the yield of grain than other physiological parameters under salt stress. Salt tolerance as defined by the grain yield and amount of PAR absorbed by a plant was found to be strongly related to LAI (Zeng et al. 2003a). Nutrient solution popularly known as Yoshida medium (Yoshida et al. 1976), supplemented with different concentrations of NaCl, is commonly used for salt tolerance screening in rice. A number of cultivars, landraces, and advanced breeding lines such as Pokkali, Nonabokra, SR26B, Damodar, Cheriviruppu, CSR11, Getu, FL378, FL 478, IR 51500-AC17, IR 51500-AC11-1, IR 4595-4-1-13, IR 51491-AC10 have been identified as useful sources for salt tolerance in rice (Dwivedi et al. 2010). Using these lines as donors, few salt-tolerant lines have been released (Gregorio et al. 2002; Ismail et al. 2007). Large number of introgression lines with enhanced abiotic stress tolerance have been developed in a massive backcross breeding program using three recurrent parents and 203 donor lines with tolerance to several abiotic stresses in rice (Ali et al. 2006).

Genetic variation in two key physiological traits, leaf Na^+ and K^+/Na^+ discrimination ratio, among the genotypes of barley and wheat, indicates the possibility of genetic improvement of salt tolerance. Bread wheat, which maintains a lower shoot Na^+ concentration than durum wheat, restricts Na^+ transport to leaf tissues through Na^+ exclusion and maintains high selectivity of K^+ over Na^+, while barley is not so efficient with respect to these traits. However, the adverse effects of Na^+ within leaves of barley are minimized by its compartmentalization into vacuoles (with Cl^-) in leaves by a mechanism known as tissue tolerance (Munns and James 2003; Colmer et al. 2005) and the production of organic solutes to osmotically balance the cytosol (Garthwaite et al. 2005). Khan et al. (2006) compared performance of 16 wheat genotypes under saline condition using gravel culture technique in lysimeters with four salinity levels, i.e., control (1.5 dS m^{-1}), low saline (6.0 dS m^{-1}), medium saline (9.0 dS m^{-1}), and highly saline (12.0 dS m^{-1}). On the basis of less than 50% reduction in yield and yield components, they found five genotypes viz. LU-26 s, HT-45, ESW-9525, V-8319, Sarsabz were tolerant, whereas Bhittai, Marvi, Chakwal-86, DS-17, Sussi (SD-66), Zardana were medium tolerant, SD1200/51, Khirman, V-7012 medium sensitive, and RWM-9313, SH-43 sensitive. Tolerant wheat genotypes were successful in maintaining low Na^+ and high K^+ uptake and high K^+/Na^+ ratio. A durum wheat line 149 has low Na^+ concentrations and high K^+/Na^+ ratios in the leaf blade (Munns et al. 2000) due to presence of two genes, *Nax1* and *Nax2*, which are responsible for exclusion of salts from leaves and roots, respectively (Munns and James 2003). However, the concentration of Na^+ in shoots of line 149 as a whole was not as low as bread wheat (Husain et al. 2004), suggesting retention of Na^+

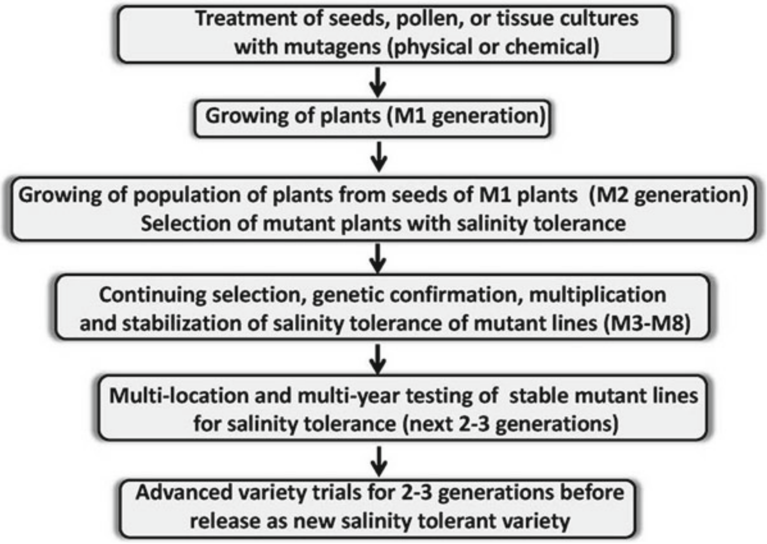

Fig. 4.1 A generalized approach for developing salinity-tolerant crop plants using mutation breeding

in the leaf sheath. The physiological traits associated with Na$^+$ accumulation include the rate of transfer of Na$^+$ from the root to the shoot (net root xylem loading) and this was much lower in Line 149 than the durum cv Tamaroi (Davenport et al. 2005). The genotypes did not differ in unidirectional root uptake of Na$^+$. The major differences in Na$^+$ transport between the genotypes were in the rate of transfer to the shoot and the preferential accumulation of Na$^+$ in the leaf sheath versus the leaf blade (Davenport et al. 2005). The K$^+$/Na$^+$ ratio at early seedling stage has been shown to be a convenient and reliable indicator of salt tolerance in wheat and barley (Tajbakhsh et al. 2006; Thalji and Shalaldeh 2007). Stomatal conductance and chlorophyll content in leaves can be measured by a non-destructive, rapid, and simple technique using a porometer and SPAD meter, respectively. El-Hendawy et al. (2007) used net photosynthetic rate, stomatal conductance, and SPAD values to screen wheat genotypes for salinity tolerance and reported genetic variation for these traits. Based on this study, SPAD reading could be used as a potential tool for large-scale screening for salt tolerance.

Wild relatives of wheat and barley exhibit genetic variation for Na$^+$ and Cl$^-$ exclusion (Colmer et al. 2005) which provides opportunity to improve salt tolerance through interspecific hybridization. In case of wheat, the diploid progenitors, *Aegilops tauschii* (DD) and *Triticum urartu* (AA), and synthetic hexaploid wheat involving *Triticum urartu*, were superior in K$^+$/Na$^+$ discrimination. Similarly, wild *Hordeum* species including *Hordeum vulgare* subsp. *spontaneum* showed high leaf K$^+$ concentration and enhanced ability to exclude Na$^+$ and Cl$^-$. Wild relatives of wheat, Tall wheatgrass (*Agropyron elongatum*) and *Lophopyrum elongatum* provide a source of novel genes for improvement of salt tolerance of bread wheat. A comprehensive list of salt-tolerant germplasm in important cereal crops is reported by Dwivedi et al. (2010).

3.1.2 Mutation Breeding

Creation of novel and useful genetic variation in important agronomic traits is the most important prerequisite for a crop-breeding program. Mutagenic agents, such as X-rays, gamma rays, fast neutrons, and chemical mutagens such as ethyl methane sulfonate have been used to induce mutations in seeds to generate genetic variation for crop improvement (Fig. 4.1). It offers the possibility of inducing desired characters that either cannot be found in nature or have been lost during evolution. The mutagen treatment breaks the nuclear DNA and during the process of DNA repair, new mutations are induced randomly. The mutants with abiotic stress tolerance can be selected by plant breeders to develop salinity-tolerant crop plants. The purpose of induced mutations is to enhance the mutation frequency rate in order to select appropriate variants for salinity tolerance. The FAO/IAEA Mutant Variety Database (MVD) collects information on plant mutant varieties (cultivars) released officially or commercially worldwide. However, with international collaborative effort coordinated by IAEA and FAO, more than 2,700 mutant varieties with one or more useful traits from induced mutations (mainly from X-rays and γ-rays) have been released in 170 different plant species all over the world. Diamant variety in barley, created by irradiation of dormant seeds with X-rays, has very high yield, short stem, very good grain, and malting quality as well as lodging resistance. Calrose 76 was the first semi-dwarf table rice variety released in the US produced by irradiation with γ-rays. Some outstanding examples of mutant rice cultivars are VND 95-20 and VND 99-3 which have long grains with excellent grain quality and wide adaptation to acid sulphate soil and salinity. These mutants not only increased biodiversity, but also provided valuable breeding material for crop improvement. The database of mutant varieties can be found at the web http://mvgs.iaea.org/AboutMutantVarieties.aspx, maintained by Plant Breeding and Genetics section of joint IAEA/FAO program.

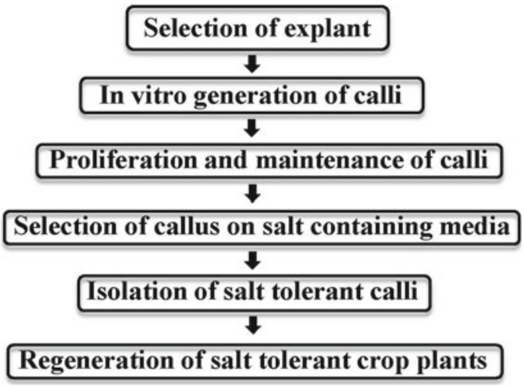

Fig. 4.2 An in vitro procedure for regeneration of salinity tolerant crop plants

3.2 Tissue Culture Approach

Plant tissue culture techniques provide a promising and feasible approach to develop salt-tolerant crop plants. Haploid culture, double haploidy, somaclonal variation, and in vitro-induced mutagenesis has been used to create variability to improve salinity tolerance in crop plants. Cell and tissue culture techniques have been used to obtain salt-tolerant plants through in vitro culture approaches: selection of mutant cell lines from cultured cells and subsequent plant regeneration (Zair et al. 2003; Gandonou et al. 2006; Lu et al. 2007; Queirs et al. 2007) and in vitro screening of plant germplasm for salt tolerance (Arzani and Mirodjagh 1999; Dziadczyk et al. 2003; Lee et al. 2003b; Wheatley et al. 2003; Dasgupta et al. 2008). Cereal tissue culture is of great economic importance for selection of improved cell lines under in vitro condition (Fig. 4.2). Tissue culture has been used as a breeding tool for rapid screening of genetic materials for salt tolerance in wheat (Arzani and Mirodjagh 1999). Houshmand et al. (2005) evaluated salt-tolerant genotypes of durum wheat derived from the in vitro culture in field experiments under both saline and non-saline field conditions as well as under greenhouse condition using salinized solution culture. In spite of the smaller range of genotypes used from the in vitro method, tolerant genotypes performed comparably with those of the field-derived tolerant genotypes for grain yield under saline field conditions. Overall, in vitro selected tolerant genotypes showed significantly better performance for biomass production under high salinity condition than field-derived tolerant genotypes.

3.2.1 Somaclonal Variants

Somaclonal variation refers to the variation seen in plants that have been originated through plant tissue culture. It is particularly common in plants regenerated from callus. The amount of variation that can be expected under in vitro condition may vary with the clone, age of the clone, and use of selection pressure applied to single cells for

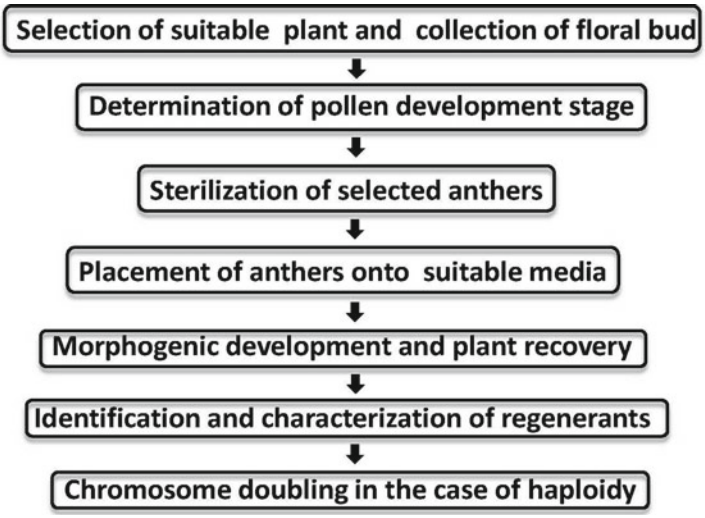

Fig. 4.3 A common procedure for haploid and dihaploid production in crop plants

salinity stress. Somaclonal variations are stable and occur at high frequencies. Novel gene mutations may result during the tissue culture process. It can be performed in vegetatively or sexually or asexually propagated plants. This approach reduces the time required for the release of new variety compared to mutation breeding and has been useful in breeding programs (Zhu et al. 2000).

3.2.2 Double Haploids

A major limitation of the conventional breeding is the long-time frame in developing varieties. Double haploid breeding enables breeders to produce genetically uniform lines within one generation. This effectively bypasses the lengthy process of self-pollination and selection normally required to produce true breeding genotypes. Double Haploids (DH) are plants that have undergone chromosome duplication from haploid plants. The production of haploids and DHs through gametic embryogenesis is the most effective way for the development of complete homozygous lines from heterozygous parents in comparison with the conventional breeding methods that employ several generations of selfing for getting homozygous plants. DH technique is well established in a range of economically important crop species, including major cereals (Wedzony et al. 2009). Various methods such as chromosome elimination subsequent to wide hybridization, pollination with irradiated pollen, selection of twin seedlings, in vivo or in vitro pollination with pollen from a triploid plant, gynogenesis and pollen embryogenesis through in vitro anther or isolated microspore culture were used to obtain DHs (Forster and Thomas 2005). Anther culture and DH production is influenced by various internal and external factors. A generalized method for haploid and DH production is shown in Fig. 4.3. A list of haploid-derived varieties of asparagus, barley, brassica, eggplant, melon, pepper, rapeseed, rice, tobacco, triticale, and wheat are available at http://www.scri.sari.ac.uk/assoc/COST851/Default.htm. The development of protocols to produce haploid and DH plants has significant impact on agricultural systems. Mostly in vitro-derived anther or isolated microspore culture method are preferably used to obtain haploids and DHs from diploid plants (Germanà 2010). Recently, a simple method for synthesizing DHs (SynDH) especially for allopolyploid species has been reported by utilizing meiotic restitution genes (Zhang et al. 2011a). This method involved three steps: hybridization to induce recombination, interspecific hybridization to extract haploids, and spontaneous chromosome doubling by selfing the interspecific F_1s. Zhang et al. (2011a) used *Triticum turgidum* L. and *Aegilops tauschii* Coss, the two ancestral species of common wheat (*Triticum aestivum* L.) to demonstrate the SynDH method using molecular markers. DHs produced in this way contain recombinant chromosomes in the genome(s) of interest in a homogeneous background. This method does not require special equipment or treatments involved in the DH production, and it can be easily applied in any breeding program. Lee et al. (2003b) produced salt-tolerant DHs rice using anther culture techniques with different genotypes in six F_1 hybrids obtained by backcross or three-way cross between *indica* and *japonica* differing in salt tolerance. It was found that the efficiency of callus induction and plant regeneration was decreased by NaCl concentration and salt tolerance of donor variety, whereas induction in *japonicas* was higher than those in indicas. The percentages of callus induction in Gyehwa 5 (*japonica*, tolerant) and IR61633-B-2-2-1 (*japonica*, sensitive) were 21.1% and 13.5% on agar medium containing 0.3% NaCl, respectively. In four F_1 hybrids, the frequencies of high salt-tolerant DHs were 21.4% and 8.9% in 0.3% NaCl medium and the control, respectively. Therefore, the high frequency of salt-tolerant DHs could be selected in the callus induction medium (0.3% NaCl) and in the combinations crossed with salt-tolerant *japonica* as the third parent. F_1 anther culture has become an effective tool to attain homozygosity of recombinants within the shortest possible time. The technique also offers the opportunity to screen haploid materials at the early stage of tissue culture. This allows recessive mutants to be identified under a variety of selection pressures. This approach was used to develop salt-tolerant homozygous recombinants from diverse cross combinations, which led to the identification of the promising rice varieties IR51500-AC-17, IR51485-AC-1, IR51500-AC11-1, and AC6534-4 for salinity, AC6533-3 for sodicity, and AC6534-1 for dual tolerance (Singh et al. 1992; Singh and Mishra 1995; Senadhira et al. 2002). Rahman et al. (2010) developed DH lines from the crosses involving salt-tolerant IRRI-derived lines using anther culture and in a field study one line AC-1 was promising for cultivation in saline areas of Bangladesh.

3.3 Genomics Approach

Genome-mapping techniques are accelerating identification of exact position and function of individual genes controlling agronomic traits including tolerance to salinity. Striking similarities among the genomes of different crop species has been helpful in expanding the genetic variability of important traits for crop improvement. The scope and precision of current breeding programs are enhanced due to use of linked markers for selection of desirable alleles for the target traits.

3.3.1 QTLs for Components of Salinity Tolerance

Plant adaptation to unfavorable environments is governed by morphological, physiological, and unique genetic architecture. By integrating physiological and genetic strategies, we can obtain a better understanding of the molecular basis of crop adaptation thus paving the way toward a more targeted breeding approach for enhancing abiotic stress tolerance in crop plants. QTL mapping is revealing genetic components of salt tolerance for genetic improvement of existing varieties. QTLs controlling salinity tolerance related traits have been mapped in several mapping populations in major field crops: rice (Table 4.1), wheat (Table 4.2), and barley (Table 4.3). To date, there are over 10,000 mapped QTLs reported for rice and maize in the Gramene database (http://www.gramene.org).

Bonilla et al. (2002) identified a major QTL, designated *Saltol*, on chromosome 1, using an RIL population between the highly tolerant landrace Pokkali and sensitive IR29. The QTL accounts for about 45% of the variation for seedling and shoot Na^+/K^+ ratio. Ismail et al. (2007) mapped the *Saltol* QTL within 1.2 Mb, which is currently being introgressed into several popular salt-sensitive rice cultivars. But multiple alleles were identified at the *Saltol* locus when several Pokkali accessions and near-isogenic lines were analyzed (Thomson et al. 2010). Using an RIL population developed from the cross Co39 X Moroberekan, Haq et al. (2010) identified a major effect on QTL for leaf Na^+ concentration and K^+:Na^+ ratio on chromosome 1 which may harbor

Table 4.1 Molecular markers associated with quantitative trait loci for salt tolerance in rice

Molecular markers	QTLs	Cross	References
	Chr 1		
RM8094, RM10793, SKC1, RM493	Standard Evaluation System (SES) score	Pokkali X IR29	Alam et al. (2011)
C813-C86	Survival days of seedling	Nona Bokra X Koshihikari	Lin et al. (2004)
C1211-S2139	Shoot K^+ concentration	Nona Bokra X Koshihikari	
RM562-RM543, RM8086-RM8231	Na^+ uptake	Taromnahelli X Khazar	Sabouri and Sabouri (2008)
RM562-RM9	Shoot K^+ concentration	IR64 X Binam	Zang et al. (2008)
E12M51_1 to E12M37_1	Na^+ uptake, K^+ Conc., Na^+:K^+ ratio	IR4630 X IR15324	Koyama et al. (2001)
Est_2-RZ569A	Seedling salinity tolerance	Milyang 23 X Gihobyeo	Lee et al. (2007)
R117–R2417	Shoot length	Niponbare X Kasalath	Takehisa et al. (2004)
	Chr 2		
OSR17-RM211, RM530-RM250	Survival days of seedling	IR64 X Binam	Zang et al. (2008)
R418-R3393	Tiller number	Niponbare X Kasalath	Takehisa et al. (2004)
	Chr 3		
RM5639,RM5626, RM3867	Standard Evaluation System (SES) score Survival days of seedling	Pokkali X IR29	Alam et al. (2011)
RM231-RM175	Shoot K^+ concentration	IR64 X Binam	Zang et al. (2008)
RM81B-RM22	Shoot Na^+ concentration	IR64 X Binam	
RM231-RM175	Na^+ uptake	IR64 X Binam	
RM416-RM5626	K^+ uptake	Taromnahelli X Khazar	Sabouri and Sabouri (2008)
RM1022-RM6283	Na^+:K^+ uptake ratio	Taromnahelli X Khazar	
RM6832-RM7389	Shoot length	Taromnahelli X Khazar	
R1927-R250	Seedling salinity tolerance	Niponbare X Kasalath	Takehisa et al. (2004)
RG179-RZ596		Milyang 23 X Gihobyeo	Lee et al. (2007)
	Chr 4		
RM6659	Standard Evaluation System score (SES)	Pokkali X IR29	Alam et al. (2011)
RM261-E12M79_6	Na^+ conc., K^+ uptake, Na^+:K^+ ratio	IR4630 X IR15324	Koyama et al. (2001)
C891-C513	Root K^+ concentration	Nona Bokra X Koshihikari	Lin et al. (2004)
C1016	Shoot length	Niponbare X Kasalath	Takehisa et al. (2004)
	Chr5		
RM163	Standard Evaluation System (SES) score	Pokkali X IR29	Alam et al. (2011)
RM421-RM440	Seedling root dry weight	Taromnahelli X Khazar	Sabouri and Sabouri (2008)
C624-C1268	Shoot fresh weight	Niponbare X Kasalath	Takehisa et al. (2004)
	Chr 6		
RM20224	Standard Evaluation System (SES) score	Pokkali X IR29	Alam et al. (2011)
RM50-RM539	Shoot K^+ concentration	IR64 X Binam	Zang et al. (2008)
RM527-RM3	Shoot Na^+ concentration	IR64 X Binam	
RM3827-RM340	Na^+:K^+ uptake ratio	Taromnahelli X Khazar	Sabouri and Sabouri (2008)
C214-R2549	Survival days of seedling	Nona Bokra X Koshihikari	Lin et al. (2004)
R1167-R1608	Shoot fresh weight	Niponbare X Kasalath	Takehisa et al. (2004)
G1314-Rz413b	Na^+ conc., K^+ uptake	IR4630 X IR15324	Koyama et al. (2001)
	Chr 8		
RM4955-RM152	K^+ uptake	Taromnahelli X Khazar	Sabouri and Sabouri (2008)

(continued)

Table 4.1 (continued)

Molecular markers	QTLs	Cross	References
RM38-RM25	Survival days of seedling	IR64 X Binam	Zang et al. (2008)
	Chr 9		
RM1553-RM5702	Na^+ uptake	Taromnahelli X Khazar	Sabouri and Sabouri (2008)
R1751-R2638	Root Na^+ concentration	Nona Bokra X Koshihikari	Lin et al. (2004)
C506-C1263	Tiller number	Niponbare X Kasalath	Takehisa et al. (2004)
RM205, E12M31_1	K^+ uptake	IR4630 X IR15324	Koyama et al. (2001)
	Chr 10		
RM222	Standard Evaluation System (SES)score	Pokkali X IR29	Alam et al. (2011)
RM7545-RM4455	Na^+ uptake	Taromnahelli X Khazar	Sabouri and Sabouri (2008)
	Chr 11		
RM26063, RM224	Standard Evaluation System (SES) score	Pokkali X IR29	Alam et al. (2011)
RM120-RM181	Shoot K^+ concentration	IR64 X Binam	Zang et al. (2008)

Table 4.2 Molecular markers associated with quantitative trait loci for salt tolerance in wheat

Molecular markers	QTLs	Cross	References
	Chr 1		
Xm71p78.5	Grain yield	Chinese spring X SQ1	Quarrie et al. (2005)
XksuD1.2-Xcdo426	Seedling shoot dry weight	Opata85 X W7984	Ma et al. (2007)
Gbm1153-barc028a	Leaf symptoms	Berkut X Krichauff	Genc et al. (2010)
Barc028a-gwm164	Tiller number	Berkut X Krichauff	
wPt-4647-wmc147	Shoot K^+ conc.	Berkut X Krichauff	
	Chr 2		
XksuD22, XksuE16,Xgwm312	Shoot Na^+ conc.	Tamaroi X Line 149	Lindsay et al. (2004)
Xm86p65.1	Grain yield	Chinese spring X SQ1	Quarrie et al. (2005)
Xfba70.1-Xcdo447	Seedling dry weight	Opata85 X W7984	Ma et al. (2007)
Xcdo1281-Xfba106	Seedling shoot fresh weight	Opata85 X W7984	
Gwm102-wmc027	Leaf symptoms	Berkut X Krichauff	Genc et al. (2010)
Gwm095-cfa2263	Seedling biomass	Berkut X Krichauff	
wPt-3114-wmc170	Shoot Na^+ conc.	Berkut X Krichauff	
wmc272-barc349	Shoot Na^+ conc.	Berkut X Krichauff	
	Chr 3		
Xbarc042-Xgwm383	Grain yield	Chinese spring X SQ1	Quarrie et al. (2005)
Xglk683-Xtam61	Germination salt tolerance	Opata85 X W7984	Ma et al. (2007)
Xglk683-Xtam61	Germination radical dry weight	Opata85 X W7984	
Xfbb168-Xbcd147	Germination radical dry weight	Opata85 X W7984	
Xtam47-Xcdo460	Seedling salt injury	Opata85 X W7984	
Xfbb168-Xbcd147	Seedling shoot dry weight	Opata85 X W7984	
Xfbb168-Xbcd147	Seedling shoot fresh weight	Opata85 X W7984	
Xfbb117-Xfbb156	Seedling dry weight	Opata85 X W7984	
Xfbb156-Xfba220	Seedling fresh weight	Opata85 X W7984	
Xcdo1406-Xbcd288226	Chlorophyll content	Opata85 X W7984	
Gwm299-gwm247	Shoot K^+ conc.	Berkut X Krichauff	Genc et al. (2010)
Cfd223-cfd152	Shoot K^+ conc.	Berkut X Krichauff	
	Chr 4		
Xm92p78.8-Xpsr490.2Ss1	Grain yield	Chinese spring X SQ1	Quarrie et al. (2005)
Xgwm165.2	Grain yield	Chinese spring X SQ1	

(continued)

Table 4.2 (continued)

Molecular markers	QTLs	Cross	References
Xcdo1081-Xfbb226	Germination salt tolerance	Opata85 X W7984	Ma et al. (2007)
Xcdo1081-Xfbb226	Germination radical fresh weight	Opata85 X W7984	
Xbcd588-Xbcd129	Seedling fresh weight	Opata85 X W7984	
Xbcd588-Xbcd129	Seedling shoot fresh weight	Opata85 X W7984	
Xbcd588-Xbcd129	Seedling root fresh weight	Opata85 X W7984	
wPt-7062-gwm6	Tiller number	Berkut X Krichauff	Genc et al. (2010)
wPt-7062-gwm6	Seedling biomass	Berkut X Krichauff	
wPt-7919-wPt-0150	Shoot K^+ conc.	Berkut X Krichauff	
	Chr 5		
Xwg232.2-Xbarc074	Grain yield	Chinese spring X SQ1	Quarrie et al. (2005)
Xbarc044	Grain yield	Chinese spring X SQ1	
Xbcd1871-Xcdo749	Germination stage salt tolerance	Opata85 X W7984	Ma et al. (2007)
Xfbb12.2-Xfba127	Seedling salt injury	Opata85 X W7984	
Gwm304-gwm186	Leaf symptoms	Berkut X Krichauff	Genc et al. (2010)
wPt-1370-Vrn1A	Tiller number	Berkut X Krichauff	
wPt-1370-Vrn1A	Seedling biomass	Berkut X Krichauff	
wPt-1370-Vrn1A	Chlorophyll content	Berkut X Krichauff	
wPt-1370-Vrn1A	Shoot K^+ conc.	Berkut X Krichauff	
	Chr 6		
Xm87p78.5a	Grain yield	Chinese spring X SQ1	Quarrie et al. (2005)
Xfba345-Xglk479	Seedling salt injury	Opata85 X W7984	Ma et al. (2007)
Xfbb231.1-Xpsr106	Seedling root fresh weight	Opata85 X W7984	
Xfbb231.1-Xpsr106	Seedling root dry weight	Opata85 X W7984	
cfd287-cfd076a	Leaf symptoms	Berkut X Krichauff	Genc et al. (2010)
cfd287-cfd076a	Seedling biomass	Berkut X Krichauff	
cfd080-barc171	Shoot Na^+ conc.	Berkut X Krichauff	
	Chr 7		
Xpsp3094.1Xm68p78.6	Grain yield	Chinese spring X SQ1	Quarrie et al. (2005)
Xfba72-Xfba127	Germination stage radical dry weight	Opata85 X W7984	Ma et al. (2007)
Xfba311-Xbcd178	Seedling fresh weight	Opata85 X W7984	
Xwg380-XksuD2	Chlorophyll content	Opata85 X W7984	
wPt-5153-ksm019	Leaf symptoms	Berkut X Krichauff	Genc et al. (2010)
wPt-5153-ksm019	Shoot K^+ conc.	Berkut X Krichauff	
gwm282-wPt-0961	Seedling biomass	Berkut X Krichauff	
wPt-4744-gwm282	Shoot Na^+ conc.	Berkut X Krichauff	

multiple genes for salt tolerance whose impact varies with stress duration, leaf age, and leaf type. Lang et al. (2001) reported a micro-satellite marker, RM223 on rice chromosome 8, associated with salt tolerance at both vegetative and reproductive stages. Recently, Alam et al. (2011) identified salinity-related QTLs at seedling stage using SES scores and identified *Saltol* as well as non-*Saltol* related QTLs at seedling-stage in Pokkali, which may assist in QTL pyramiding and marker-assisted breeding programs. A study by Lin et al. (2004) employing the tolerant *indica* landrace Nonabokra with the susceptible *japonica* Koshihikari, identified several large-effect QTLs, including the *SKC1* QTL and a QTL for shoot Na^+ concentration. While the salt-tolerant landraces Pokkali and Nonabokra were routinely used in the past for breeding, the level of tolerance attained by new lines is always lower than traditional donors (Gregorio et al. 2002), and the existing tolerant varieties seem to be superior in only few traits associated with tolerance. The QTL *SKC1*, originally detected by its effect on K^+ concentration, was cloned by map-based cloning and was

Table 4.3 Molecular markers associated with quantitative trait loci for salt tolerance in barley

Molecular markers	QTLs	Cross	References
	Chr 1H		
GIb1-ABC160	Seedling salt tolerance	Steptoe X Morex	Mano and Takeda (1997)
ABC160-His4A	Seedling salt tolerance	Steptoe X Morex	
WG789B-ABR337	Seedling salt tolerance	Steptoe X Morex	
Drun8-ABC261	Germination salt tolerance	Harrington X TR306	
bPb-2240-bPb-0631	Spikes per plant	CM72 X Gairdner	Xue et al. (2009)
	Chr 2H		
ABG459-Pox	Seedling salt tolerance	Steptoe X Morex	Mano and Takeda (1997)
ABC152D-Rrn5S1	Seedling salt tolerance	Steptoe X Morex	
His3C-ABC152D	Seedling salt tolerance	Steptoe X Morex	
P21M12d	Shoot dry weight	Derkado X B83-12/21/5	
bPb-6088-bPb-4377	Dry weight per plant	CM72 X Gairdner	Ellis et al. (2002)
Bmag0381-bPb-0827	Grain number per plant	CM72 X Gairdner	Xue et al. (2009)
bPb-3536-bPb-1103	Shoot Na^+ Conc.	CM72 X Gairdner	
	Chr 3H		
bPb-0049-bPb-4564	Plant height	CM72 X Gairdner	Xue et al. (2009)
bPb-7989-bPb-4660	Spikes per plant	CM72 X Gairdner	
	Chr 4H		
MWG634-WG622	Germination salt tolerance	Steptoe X Morex	Mano and Takeda (1997)
P17M62f	Shoot dry weight	Derkado X B83-12/21/5	
bPb-1278-bPb-3512	Tiller number	CM72 X Gairdner	Ellis et al. (2002)
bPb-1278-bPb-3512	Spikes per line	CM72 X Gairdner	Xue et al. (2009)
bPb-0130-bPb-8437	Spikes per plant	CM72 X Gairdner	
	Chr 5H		
WG889-ABC324	Germination salt tolerance	Steptoe X Morex	Mano and Takeda (1997)
ABC309-MWG632	Germination salt tolerance	Harrington X TR306	
iEst9-WG908	Seedling salt tolerance	Steptoe X Morex	
WG364-MWG514B	Seedling salt tolerance	Steptoe X Morex	
CDO504-ABG712	Seedling salt tolerance	Harrington X TR306	
TubA3-MWG740	Seedling salt tolerance	Harrington X TR306	
Bmag337	Shoot dry weight	Derkado X B83-12/21/5	Ellis et al. (2002)
	Chr 6H		
ABG387B-ABG458	Germination salt tolerance	Steptoe X Morex	Mano and Takeda (1997)
BCD340E-ksuD17	Seedling salt tolerance	Steptoe X Morex	
bPb-6421-bPb-3921	Spikes per line	CM72 X Gairdner	
bPb-7323-bPb-2751	Grain yield	CM72 X Gairdner	Xue et al. (2009)
bPb-8889-bPb-7323	Na^+: K^+ ratio	CM72 X Gairdner	
	Chr 7H		
bPb-1209-bPb-6821	Spikes per line	CM72 X Gairdner	Xue et al. (2009)
P40M38b	Tiller number	Derkado X B83-12/21/5	Ellis et al. (2002)

identified as the sodium transporter *OsHKT8* (Ren et al. 2005). Combining superior alleles underlying the component traits could potentially result in higher levels of tolerance. Based on QTL-linked marker profile, Manneh et al. (2007) identified superior salt-tolerant genotypes of rice to improve selection efficiency while selecting for yield in stress environments. Wang et al. (2011) mapped 16 QTLs for salt tolerance of rice at the seed germination stage and four QTLs with major effect could be useful to improve stand establishment under saline areas. QTLs associated with tolerance at various developmental stages will be needed for more stable performance in salt affected areas.

Improving salt tolerance and productivity wheat is a major challenge in breeding program. In studies involving hexaploid, tetraploid, and diploid types, it was suggested that the D genome of wheat carries gene *Kna1* that controls the relative concentration of K^+ and Na^+ in the shoots of plants grown in saline hydroponic culture (Shah et al. 1987; Gorham et al. 1987, 1997; Gorham 1994). This gene was mapped on to the chromosome 4DL (Dvorak et al. 1994) and then fine mapped as a single gene (Dubcovsky et al. 1996). Ma et al. (2007) used a wheat RIL population derived from the cross Opata85 × W7984 (international mapping population) and identified 47 QTLs based on salt tolerance index, salt injury index, biomass, shoot length, root length, chlorophyll content, and proline content on all wheat chromosomes except 1B, 1D, 4B, 5D, and 7D. Ten of these QTLs were effective during germination stage and 37 QTLs were important at the seedling stage. In another study, genetic analysis of wheat Line 149 identified two major genes *Nax1* and *Nax2* for Na^+ exclusion (Munns et al. 2003). *Nax1* was located on chromosome 2A by QTL analysis (Lindsay et al. 2004) and has been fine mapped as an Na^+ transporter of the HKT gene family, *HKT7* (Huang et al. 2006). *Nax2* located on chromosome 5A was identified as *HKT8* (Byrt et al. 2007). *Nax1* removes Na^+ from the xylem in roots and the lower parts of leaves, the leaf sheaths, while *Nax2* removes Na^+ from the xylem only in the roots (James et al. 2006). *Nax2* has the same phenotype as *Kna1*, the QTL for Na^+ exclusion and enhanced K^+/Na^+ selectivity in bread wheat, *T. aestivum* (Dvorak et al. 1994). *Nax2* was shown to be homologous to *Kna1* in *T. aestivum*, namely *TaHKT8* (Byrt et al. 2007). The *HKT* gene family encodes transporters in the plasma membrane that mediate the uptake of Na^+ or K^+ from the apoplast (Hauser and Horie 2010). They are important for cellular Na^+ and K^+ homeostasis, and their expression in the stele, particularly in the xylem parenchyma cells lining the xylem vessels, helps in retrieving Na^+ from the transpiration stream and thus contributing to Na^+ exclusion from leaves (Munns and Tester 2008; Hauser and Horie 2010). James et al. (2011) analyzed a population derived from crossing of a tetraploid durum wheat (*Triticum turgidum* ssp. durum) and hexaploid bread wheat (*Triticum aestivum*) by marker-assisted selection (MAS) for hexaploid plants containing either *Nax1*, *Nax2*, or both *Nax1* and *Nax2*. *Nax1* line decreased the leaf blade Na^+ concentration by 50%, whereas *Nax2* line decreased it by 30%, and both genes together decreased it by 60%. High Na^+ sheath:blade ratio in *Nax1* lines conferred extra advantage under waterlogged and saline conditions. The effect of *Nax2* on lowering the Na^+ concentration in bread wheat was surprising as this gene is already present in bread wheat, putatively at the *Kna1* locus. The results indicate that both *Nax* genes have the potential to improve the salt tolerance of bread wheat. A list of salinity-related QTLs for wheat and barley is given in Tables 4.2 and 4.3, respectively.

3.3.2 Marker-Assisted Selection

The application of QTL mapping provided the means to genetically dissect tolerance traits into discrete QTLs that can then be pyramided into high-yielding varieties using marker-assisted selection (MAS). Marker-assisted selection is the use of molecular markers linked to useful traits to select individuals with desirable genetic makeup during the variety development process. It provides a dramatic improvement in the efficiency with which breeders can select plants with desirable combination of genes. DNA markers should enhance the recovery rate of the isogenic recurrent genome after hybridization and facilitate the introgression of quantitative trait loci necessary to increase stress tolerance (Fig. 4.4). Molecular marker techniques were used successfully to transfer alleles of interest from wild relatives into commercial cultivars (Tanksley and McCouch 1997). Use of permanent mapping populations, such as recombinant inbred lines (RILs) or chromosomal segment substitution lines (CSSLs) enables testing stress-tolerant traits in replicated experiments across different environments, which can help in differentiating the QTLs based on their effectiveness at different stress levels. Once important QTL targets are identified, it can be used as single introgressions in a set of near-isogenic lines (NILs), which will help in identifying the complexity of different traits by limiting the variation between lines to focus only on the locus of interest.

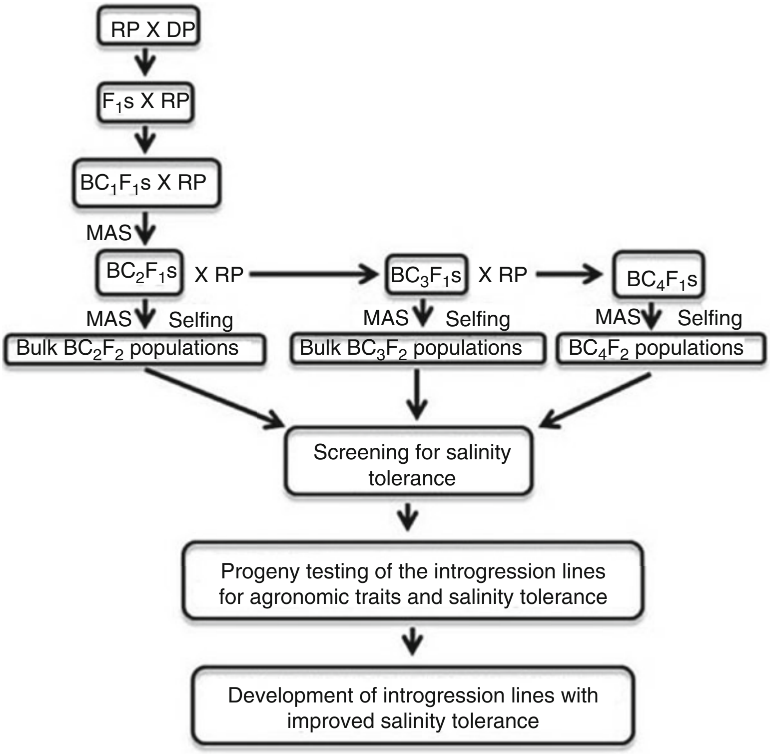

Fig. 4.4 A schematic representation of marker-assisted backcross breeding procedure for development of introgression lines with enhanced salt tolerance. *RP* recurrent parent, *DP* donor parent, *MAS* Marker assisted selection

The recent advances in genomics have paved the way for clear and reliable methods for MAS in plants starting from QTL identification, NIL development, and fine-mapping to transferring the QTL into popular varieties using a precise marker-assisted backcrossing (MABC) strategy (Collard et al. 2005, 2008 Collard and Mackill 2008;). MABC involves the manipulation of genomic regions involved in the expression of particular traits of interest through DNA markers, and combines the power of a conventional backcrossing program with the ability to differentiate parental chromosomal segments (Fig. 4.4).

3.4 Transgenic Approach

Progress in genomics is instrumental in discovery and characterization of large number of salt stress-related candidate genes offering unique opportunity for exploiting transgenic technology to enhance salinity tolerance in crop plants. Development of transgenic plants with improved tolerance to abiotic stresses has brought some hope for sustainable agriculture under harsh environmental conditions. Genetic engineering is an attractive option when genes of interest are present in cross barrier species, distant relatives, or non-plant sources (Bhatnagar-Mathur et al. 2008). It is also faster to introduce beneficial genes than the conventional or molecular breeding. Due to the complexity associated with salt tolerance mechanism, effort should be made to introduce and fine tune multiple genes with synergistic effect under suitable stress inducible promoters for controlling their expression at a specific time, in a specific organ, or under specific conditions of stress.

3.4.1 Discovery of Candidate Genes

When a plant is subjected to abiotic stresses, expressions of a number of genes are changed, resulting in altered levels of several proteins and metabolites inside the cell. Altered expression of these genes may be responsible for conferring protection or susceptibility to abiotic stresses. Candidate genes for salinity stress tolerance have been identified from prokaryotic extremophiles, lower eukaryotes, tolerant wild relatives, landraces, cultivars, and T-DNA mutant plants using high throughput techniques such as differential display polymerase chain reaction (DD-PCR), cDNA-amplified fragment length polymorphism (cDNA-AFLP), suppression subtractive hybridization (SSH), serial analysis of gene expression (SAGE), DNA microarray, and two-dimensional gel electrophoresis.

Differentially expressed genes under osmotic stress have been identified by DDRT-PCR in barley, sunflower, and pak-choi (Wei et al. 2001; Liu and Baird 2003; Qiu et al. 2009). Wei et al. (2001) identified a gene encoding the barley vacuolar ATPase subunit B (BSVAP) from salt-sensitive barley cultivar Maythorpe. In barley roots, this enzyme may be involved in the sequestration of Na^+ and Ca^{2+} ions into the vacuole, since the proton gradient produced by the ATPase is used by Na^+/H^+ and Ca^{2+}/H^+ antiporters to drive the uptake of Na^+ and of Ca^{2+} (Taiz 1992). Liu and Baird (2003) isolated 17 cDNA clones that are differentially expressed in sunflower under drought or salinity and 13 of these cDNAs were confirmed by quantitative RT-PCR to be expressed differentially in response to osmotic stress. Qiu et al. (2009) generated 101 cDNA fragments differentially expressed under salt stress in Pak-choi (*Brassica campestris* L. ssp. chinensis (L.) Makino var. communis Tsen et Lee), and found seven cDNA sequences highly homologous to some known expression genes or the genes related to the signaling pathways in plants under different abiotic stress.

Kumari et al. (2009) applied SSH technique in a salt-sensitive rice cultivar IR64 and a salt-tolerant landrace Pokkali and isolated 1,194 salinity-regulated cDNAs. Gene expression analysis of selected genes using macroarrays and Northern blots indicated that salinity tolerance of Pokkali may be due to constitutive overexpression of many salt responsive genes which are stress inducible in IR64. Few clones mapped near *Saltol* locus on chromosome 1. Using global gene expression analyses, Walia et al. (2005, 2007) observed gene expression changes in large number of genes under salinity stress in salt-sensitive genotypes compared with the tolerant lines. A total of 164 ESTs were upregulated in the tolerant line (FL478) under salinity stress during vegetative stage (Walia et al. 2005), whereas during panicle initiation stage, 292 genes were upregulated and 346 genes were downregulated in salt-sensitive *japonica* M103. Interestingly, in the salt-tolerant *japonica* only 54 were upregulated and 54 downregulated (Walia et al. 2007). Cotsaftis et al. (2011) carried out root-specific transcriptome studies in contrasting genotypes of rice (FL478, IR29, Pokkali and IR63731) under salinity stress and identified several gene families with known links to salinity tolerance.

Chen et al. (2003) used cDNA-AFLP to analyze differentially expressed genes in wheat under salinity stress and found a large number of gene fragments related to salt stress. One of the cDNA encoding glycogen synthase kinase-shaggy kinase (*TaGSK1*) was induced by NaCl stress as a part of the signal transduction component. This technique has also been used to isolate differentially expressed ESTs under salinity stress in soybean (Akoi et al. 2005) and *Spartina alterniflora* (Baisakh et al. 2008). An expressed sequence tag (EST) analysis in a grass halophyte *Spartina alterniflora* at early stages of salt stress (Baisakh et al. 2008) produced 1,227 quality ESTs of which 27% of ESTs represented genes for stress response. Transcript abundance analysis of eight known genes of various metabolic pathways and nine transcription factor genes showed temporal and tissue-dependent variation in expression under salinity stress.

A combination of genetic mapping and bulked transcriptome profiling was used by Pandit et al. (2010) resulting in identification of two genes, an integral transmembrane protein DUF6 and a chloride cotransporter, which colocalized within the QTL interval. Walia et al. (2006) performed expression analysis of genes in barley (*Hordeum vulgare* L.) during salinity stress at the seedling stage of barley using microarray. Genes, differentially regulated by salinity, were also associated with various abiotic stresses such as low temperature, heat stress, and drought stress. Expression level of genes related to jasmonic acid (JA) biosynthesis and jasmonic acid-responsive genes (JRGs) was found to be differentially regulated. Jasmonic acid may function as a "master switch" for stress-induced signaling pathway leading to changes in gene expression (Wasternack et al. 1998). It has been reported that JA and ABA affect gene expression in a synergistic manner through more or less independent signaling pathways (Ortel et al. 1999). This indicates that ABA signaling pathway is also activated along with JA pathway in response to salinity stress in barley. Ozturk et al. (2002) analyzed drought and salinity-induced responses in barley (*Hordeum vulgare* L. cv. Tokak) transcriptome using cDNA microarray and reported alteration in 5% of the genes under salinity stress compared to 15% under drought stress. Upregulation under both drought and salt stress was restricted to ESTs for metallothionein-like and LEA proteins, while increases in ubiquitin-related transcripts characterized salt stress.

Yan et al. (2005) investigated the salt stress-responsive proteins in the root of rice (*Oryza sativa* L. cv. Nipponbare) in proteomics approach using 3-week-old seedlings treated with 150 mM NaCl for 24, 48 and 72 h. Two-dimensional gel electrophoresis showed 34 upregulated and 20 downregulated proteins. Mass spectrometry analysis could identify 12 spots representing 10 different proteins involved in regulation of carbohydrate, nitrogen, and energy metabolism, reactive oxygen species scavenging, mRNA and protein processing, and cytoskeleton stability. Comparative proteomic analysis in two contrasting hybrid rice variety by Ruan et al. (2011) led to identification of new components involved in salt-stress signaling. One protein that was upregulated during salt stress was homologous to cyclophilin 2 (*OsCYP2*). Chitteti and Peng (2007) reported the differential expression of phosphoproteome under salinity stress in the root of rice. They identified 17 differentially upregulated and 11 differentially downregulated putative phosphoproteins. Witzel et al. (2009) conducted two-dimensional gel electrophoresis using a series of hydroponics-based salinity stress experiments in contrasting genetic mapping parents of barley cvs Steptoe and Morex. The proteome analysis of roots from both genotypes revealed cultivar-specific and salt stress-responsive protein expression. Twenty-six proteins could be identified by mass spectrometry. Among those, two proteins involved in the glutathione-based detoxification of reactive oxygen species (ROS) were more abundant in the tolerant genotype.

The identification of various abiotic stress-specific changes in gene expression has been achieved by comparing gene expression in non-induced and salinity stress-induced tissues or by comparing contrasting cultivars (Sahi et al. 2003; Baisakh et al. 2008; Karan et al. 2009; Kumari et al. 2009). In one such study, it was noted that less number of genes were induced under salt stress in *Thellungiella halophila* (a salt-tolerant relative of *Arabidopsis*) in comparison to *Arabidopsis* (Inan et al. 2004). This indicated that the stress tolerance of *Thellungiella halophila* may be due to constitutive overexpression of a few salt tolerance related genes which are stress inducible in *Arabidopsis* (Taji et al. 2004). Sengupta and Majumder (2009) analyzed changes in leaf protein expression under salt stress in the wild halophytic rice *Porteresia coarctata* and salt-sensitive *Oryza sativa* and identified 16 proteins involved in osmolyte synthesis, photosystem functioning, RubisCO activation, cell wall synthesis, and chaperone functions. These differentially regulated genes in different plant species may serve as candidates to improve salinity tolerance in crop plants using transgenic approach.

3.4.2 Transformation in Crop Plants

Most common genes used for genetic engineering of stress-tolerant plants include transcription factors, signal transduction genes, water channel proteins, ion transporters, detoxifying genes, molecular chaperones, dehydrins, and osmoprotectants (Table 4.4).

Calcium acts as a secondary messenger in various signal transduction pathways in plants. Xu et al. (2011) isolated a calcium-binding multi-stress-responsive gene *OsMSR2* from rice which

Table 4.4 Examples of transgenic intervention in crop plants to improve salt tolerance

Gene	Gene product	Gene source	Target plant	Effect of the transgene	Reference
HvCBF4	CBF transcription factor	*Hordeum vulgare*	Rice	Enhanced salinity, drought, and cold tolerance	Oh et al. (2007)
OSNAC5	Transcription factor	*Oryza sativa*	Rice	Enhanced salt tolerance	Takasaki et al. (2010)
ABP9	Transcription factor	*Zea mays*	Arabidopsis	Enhanced drought, high salt, freezing temperature, and oxidative stresses tolerance	Zhang et al. (2011b)
TaMYB2A	Transcription factor	*Triticum aestivum*	*Arabidopsis*	Enhanced salt, drought, and freezing tolerance	Mao et al. (2011)
TaSRG	Transcription factor	*Triticum aestivum*	Arabidopsis	Improved salt tolerance	He et al. (2011)
CgNHX1	Vacuolar Na$^+$/H$^+$ exchanger,	*Chenopodium glaucum*	Rice	Improved salt tolerance	Li et al. (2008)
PgNHX1	Vacuolar Na$^+$/H$^+$ antiporter	*Pennisetum glaucum*	Rice	Enhanced salt tolerance	Verma et al. (2007)
AtNHX1	Na$^+$/H$^+$ antiporter	*Arabidopsis thaliana*	Maize	Improved salt tolerance	Li et al. (2010b)
ThNHX1	Na$^+$/H$^+$ antiporter	*Thellungiella halophila*	Arabidopsis	Improved salt tolerance	Wu et al. (2009)
SsNHX2	Na$^+$/H$^+$ antiporter	*Suaeda salsa*	Arabidopsis	Improved salt tolerance	Li et al. (2009)
AtNHX1	Vacuolar Na$^+$/H$^+$ antiporter	*Arabidopsis*	Groundnut	Improved salt and drought tolerance	Asif et al. (2011)
OsKAT1	Shaker family K$^+$ channel	*Oryza sativa*	Rice	Enhanced salt tolerance	Obata et al. (2007)
PutHKT2;1	K$^+$ transporter	*Puccinellia tenuiflora*	Arabidopsis	Improved salt tolerance	Ardie et al. (2009)
ThIPK2	Inositol poly-phosphate kinase	*Thellungiella halophila*	*Brassica napus*	Improved abiotic stress tolerance	Zhu et al. (2009)
ZmSIMK1	Mitogen activated protein kinase	*Zea Mays*	Arabidopsis	Increased tolerance to salt stress	Gu et al. (2010)
TaSnRK2.4	Sucrose non-fermenting 1 type serine/threonine protein kinase	*Triticum aestivum*	Arabidopsis	Enhanced tolerance to drought, salt, and freezing stresses	Mao et al. (2010)
ZmMKK4	Mitogen activated protein kinase kinase kinase	*Zea mays*	*Arabidopsis*	Salt and cold tolerance	Kong et al. (2011)
ZmALDH2.2A1	Aldehyde dehydrogenase	*Zea mays*	Tobacco	Improved tolerance to salt and drought	Huang et al. (2008)
OsBADH	Betaine aldehyde dehydrogenase	*Oryza sativa* (*Indica*)	Rice (*Japonica*)	Improved salt tolerance	Hasthanasombut et al. (2011)
BADH	Betaine aldehyde dehydrogenase	*Suaeda liaotungensis*	Maize	Improved salt tolerance	Wu et al. (2008)
$H^+-PPase$	H(+)-pyrophosphatase	*Thellungiella halophila*	Cotton	Improved salt tolerance	Lv et al. (2008)
PcINO1	Myo-inositol 1-phosphate synthase	*Porteresia coarctata*	Brassica, Rice	Improved salt tolerance	Das-Chatterjee et al. (2006)
MIPS	Myo-inositol 1-phosphate synthase	*Spartina alterniflora*	Tobacco, Rice	Improved salt tolerance	Baisakh et al. (2009)
SOD	Cu/Zn superoxide dismutase	*Avicennia marina*	Rice	Improved salt tolerance	Prashanth et al. (2008)
ThCBL9	Calcineurin B-like protein	*Thellungiella halophila*	Arabidopsis	Improved salt and osmotic stress tolerance	Sun et al. (2008)
GST	Glutathione S-transferase	*Suaeda salsa*	Rice	Abiotic stress resistance	Zhao and Zhang (2006)
SbGSTU	Glutathione S-transferase	*Salicornia brachiata*	Tobacco	Enhanced salt tolerance	Jha, Sharma and Misra (2010)
GLYII	Glyoxalase II	*Oryza sativa*	Rice	Higher salt tolerance	Singla-Pareek et al. (2008)
PcSrp	Serine-rich-protein	*Porteresia coarctata*	Finger millet	Improved salt tolerance	Mahalakshmi et al. (2006)

is strongly upregulated by a wide spectrum of stresses, including cold, drought, and heat in different tissues at different developmental stages of rice. Expression of *OsMSR2* gene into *Arabidopsis* conferred enhanced tolerance to high salt and drought through ABA mediated pathway. Calcium-dependent protein kinases (CDPKs) regulate downstream components in calcium signaling pathway. Rice has 29 genes for CDPKs and constitutes a large multigene family. Asano et al. (2011) used mini-scale full-length CDNA overexpressor (FOX) gene hunting system and generated 250 independent transgenic rice plants overexpressing individual rice CDPKs. Transgenic rice plants over expressing *OsCDPK21-FOX* had more survival rate under salinity stress than wild type plants and also found to be involved in the positive regulation of ABA and salt stress signaling pathways.

The members of NAM, ATAF, and CUC (NAC) transcription factor family has been used to improve abiotic stress tolerance through genetic engineering. Transgenic rice plants constitutively overexpressing *OsNAC6* showed tolerance to dehydration, salinity, and blast disease (Nakashima et al. 2007). However, the plants exhibited growth retardation and low reproductive yields. By utilizing stress-inducible promoters, such as the native *OsNAC6* promoter, it may be possible to reduce negative impact on plant growth and reproduction. *SNAC1* (Stress responsive NAC1), which is expressed in guard cells under drought stress, improved both salt and drought tolerance in transgenic rice plants with upregulation of many stress-responsive genes (Hu et al. 2006). *OsNAC5* is induced by abiotic stresses such as salinity, drought, cold, abscisic acid, and methyl jasmonic acid and interacts with *OsNAC5, OsNAC6,* and *SNAC1*. Transgenic plants overexpressing *OsNAC5* had improved tolerance to high salinity compared to control plants (Takasaki et al. 2010).

Li et al. (2010a) isolated and characterized a basic helix-loop-helix (bHLH) protein gene *OrbHLH001* from wild rice (*Oryza rufipogon*), which encodes an ICE1-like protein containing multiple homopeptide repeats and is induced by salt stress in the shoots. Transgenic *Arabidopsis* expressing *OrbHLH001* enhanced salt and freezing tolerance independent of a CBF/DREB1 cold-response pathway. Oh et al. (2005) reported that *DREB1A* and *ABF3* rice transgenic lines did not show any stunted growth despite constitutive expression. However, transgenic plants exhibited drought and salinity stress tolerance but low level of freezing stress tolerance. DREB homologs have been isolated from rice: *OsDREB1A* and *OsDREB1B* are cold inducible, *OsDREB2A* is induced by dehydration and salinity stress (Dubouzet et al. 2003). Over-expression of the *OsDREB1* gene in rice and *Arabidopsis* correlated with improved salt, drought, and low temperature tolerance (Ito et al. 2006). *OsDREB1F* is one of the most upregulated genes isolated from upland rice. The transgenic plants harboring *OsDREB1F* gene led to enhanced tolerance to salt, drought, and low temperature in both rice and *Arabidopsis* (Wang et al. 2008). The HARDY (HRD) gene, an AP2/ERF-like transcription factor from *Arabidopsis* enhanced drought resistance and salt tolerance, accompanied by an enhancement in the expression of abiotic stress associated genes (Karaba et al. 2007).

The Na^+/H^+ antiporters play an important role in the maintenance of cellular ion homoeostasis, cytoplasmic pH regulation, and cell turgor leading to salt tolerance in plants (Horie and Schroeder 2004). Ohta et al. (2002) used a *NHX1* gene from a halophyte *Atriplex gmelini* to generate transgenic rice lines which showed eight-fold higher activity of this gene compared to the wild rice. These transgenic lines survived in 300 mM NaCl for 3 days but the wild types failed to survive. Over-expression of vacuolar Na^+/H^+ antiporter gene *OsNHX1* enhanced salt tolerance in rice (Fukuda et al. 2004), whereas over-expression of the same gene from wheat together with H^+-pyrophosphatase (TVP1) improved both salt and drought tolerance in *Arabidopsis* (Brini et al. 2007). A field trial of wheat transgenic lines expressing *AtNHX1* gene exhibited higher grain yield, heavier and larger grains in saline conditions and there was less Na^+ and more K^+ accumulation in leaves in transgenic lines compared with the non-transgenic lines (Xue et al. 2004).

Garg et al. (2002) overexpressed *E. coli* trehalose bisynthetic genes (*otsA* and *otsB*) under the control of either tissue-specific or stress responsive promoter in transgenic rice plants which accumulated 3–10 times more trehalose and showed better growth, less photo-oxidative damage and favorable mineral balance under salt, drought, and low temperature stress conditions. Transgenic rice overexpressing *OsCYP2* encoding a cyclophilin 2 protein showed increased salinity tolerance with lower levels of lipid peroxidation products and higher activities of antioxidant enzymes than wild type seedlings (Ruan et al. 2011).

Halophytes represent an ideal target for understanding the genetic and molecular basis of their adaptation in saline environments (Subudhi and Baisakh 2011). Several types of genes belonging to different metabolic functions have been identified and used for overexpression into glycophytic plants to enhance salinity stress tolerance. Ohta et al. (2002) reported enhanced salinity tolerance in transgenic rice using the vacuolar Na^+/H^+ antiporter (*AgNHX1*) from *Atriplex gmelini*. Similarly, transgene expression of Na^+/H^+ antiporter (*SsNHX1*), *SsNHX2* (an alternative splicing variant of *SsNHX1*) from *Suaeda salsa* in *Arabidopsis* resulted into higher salt tolerance as well as vigorous growth of transgenics along with higher fresh, dry weights, and Na^+ and K^+ accumulation compared with the wild type plants (Li et al. 2009). *Tau* class glutathione transferases genes are specific to plant and important for protecting plants against oxidative damage. Expression of *SbGSTU* from a halophyte *Salicornia brachiata* in tobacco led to suppressed growth of transgenic seedlings at higher salt, but these transgenic seedlings had continuous growth significantly better than WT seedlings below 300 mM NaCl (Jha et al. 2010). Jithesh et al. (2006) isolated a cDNA (*Sod1*) encoding a cytosolic Cu/ZnSOD that accumulated during oxidative stress in the mangrove species, *Avicennia marina*. Transgenic Pusa Basmati-1 rice plants with *Sod1* were more tolerant to oxidative, salinity (150 mM NaCl) and drought stresses indicating mangrove as a potential source for other abiotic stress tolerance candidate genes (Prashanth et al. 2008). Majee et al. (2004) isolated and characterized a novel salt-tolerant gene L-myo-inositol 1-phosphate synthase from halophytic rice, *Porteresia coarctata*. Incorporating this gene into rice and other plant species increased synthesis of inositol under salinity stress and enhanced tolerance to salinity stress (Das-Chatterjee et al. 2006). Jacobs et al. (2011) identified a sodium pumping ATPase gene *PpENA1* from the moss *Physcomitrella patens*, which is not found in higher plants. Expression of *PpENA1* gene increased the salinity tolerance of transgenic rice.

4 Conclusions and Future Perspective

Salinity stress causes extensive crop losses in many parts of the world due to lack of salt tolerance in major field crops. Enhancing tolerance to salinity in crops will be an important goal of plant breeders in future to ensure food supply for the growing world population (Flowers 2004). Wide range of variation in level of salt tolerance found in halophytes, wild species, and crop germplasm clearly demonstrates the genetic basis of salt tolerance. Although it is widely recognized that the genetic and physiological basis of salt tolerance in plants is inherently complex due to involvement of multigene controlled traits or mechanisms, lack of thorough understanding of these mechanisms and their contribution toward salt tolerance is a major limitation to develop salt-tolerant plants. Additionally, lack of proper screening methods, low heritability, and low selection efficiency of component traits, and limited knowledge of the interactions among the genotype, plant growth stage, and the timing of stress also contributed to slow progress in breeding salinity-tolerant crops (Flowers and Flowers 2005; Ismail et al. 2007).

Conventional breeding technologies coupled with molecular genetic analysis particularly the QTL mapping studies are revealing important genetic components to enhance salt tolerance in field crops. Salt-tolerant germplasm have been identified and developed by breeding, mutation, and tissue culture techniques. QTL mapping and marker-assisted breeding will be increasingly employed with the effort for systematic dissection and utilization of natural variation in the available germplasm for improving crop performance in saline environments (Collins et al. 2008). Particularly, the wild relatives of crops and landraces hold enormous promise to mine the superior alleles for enhancing crop adaptation under salinity (Tanksley and McCouch 1997; Feuillet et al. 2008).

A large number of candidate genes involved in different salt tolerance mechanisms from diverse sources have been identified. It is expected that modern genomics tools will continue to be employed in model organisms such as Arabidopsis and rice, salt-tolerant crop accessions and wild relatives, and halophytes to provide genetic clues for plant adaptation to salinity through identification and verification of candidate genes (Collins et al. 2008; Subudhi and Baisakh 2011). Genomic technology is expected to contribute significantly toward discovery of useful candidate genes for various component traits which can be targeted to improve elite cultivars using transgenic pyramiding (Takeda and Matsuoka 2008). The utility of transgenic technology can be further enhanced through discovery and exploitation of stress inducible promoters which could enhance salt tolerance with minimum undesirable pleiotropic effect on crop growth and yield. It is conceivable that designing crop genotypes with improved salinity tolerance may be difficult but not impossible. However, collaboration of geneticists, molecular breeders, physiologists, and genomicists will be needed to implement an integrated approach to discover, test, and introgress the superior alleles to enhance salt tolerance in major food crops.

Acknowledgments The financial support for this study from the United States Department of Agriculture-CSREES is gratefully acknowledged. This manuscript is approved for publication by the Director of Louisiana Agricultural Experiment Station, USA as manuscript number 2011-306-5892.

References

Akoi A, Kanegami A, Mihara M, Kojima T, Shiraiwa M, Takahara H (2005) Molecular cloning and characterization of a novel soybean gene encoding a leucine-zipper-like protein induced to salt stress. Gene 356:135–145

Alam R, Sazzadur Rahman M, Seraj ZI, Thomson MJ, Ismail AM, Tumimbang-Raiz E, Gregorio GB (2011) Investigation of seedling-stage salinity tolerance QTLs using backcross lines derived from *Oryza sativa* L. Pokkali. Plant Breed. doi:10.1111/j.1439-0523.2010.01837.x

Ali AJ, Xu JL, Ismail AM, Fu BY, Vijayakumar CHM, Gao YM, Domingo J, Maghirang R, Yu SB, Gregorio G, Yanaghihara S, Cohen M, Carmen B, Mackill D, Li ZK (2006) Hidden diversity for abiotic and biotic stress tolerances in the primary gene pool of rice revealed by a large backcross breeding program. Field Crops Res 97:66–76

Ardie SW, Xie LN, Takahashi R, Liu SK, Takano T (2009) Cloning of a high-affinity K^+ transporter gene *PutHKT2, 1* from *Puccinellia tenuiflora* and its functional comparison with *OsHKT2, 1* from rice in yeast and Arabidopsis. J Exp Bot 60:3491–3502

Arzani A, Mirodjagh SS (1999) Response of durum wheat cultivars to immature embryo culture, callus induction and in vitro salt stress. Plant Cell Tiss Org Cult 58:67–72

Asano T, Hakata M, Nakamura H, Aoki N, Komatsu S, Ichikawa H, Hirochika H, Ohsugi R (2011) Functional characterisation of *OsCPK21*, a calcium-dependent protein kinase that confers salt tolerance in rice. Plant Mol Biol 75:179–191

Asch F, Dingkuhn M, Dörffling K, Miezan K (2000) Leaf K^+/Na^+ ratio predicts salinity induced yield loss in irrigated rice. Euphytica 113:109–118

Ashraf M, Harris PJC (2004) Potential biochemical indicators of salinity tolerance in plants. Plant Sci 166:3–16

Asif MA, Zafar Y, Iqbal J, Iqbal MM, Rashid U, Ali GM, Arif A, Nazir F (2011) Enhanced expression of *AtNHX1* in transgenic groundnut (*Arachis hypogaea* L.) improves salt and drought tolerance. Mol Biotech. DOI: 10.1007/s12033-011-9399-1

Aslam M, Qureshi RH, Ahmed N (1993) A rapid screening technique for salt tolerance in rice (*Oryza sativa* L.). Plant Soil 150:99–107

Baisakh N, Subudhi PK, Bhardwaj P (2008) Primary responses to salt stress in a halophyte, smooth cordgrass (*Spartina alterniflora* Loisel). Funct Integr Genomics 8:287–300

Baisakh N, Rajasekharan K, Deleon T, Biradar H, Parco A, Singh P, Subudhi PK (2009) Overexpression of myo-inositol phosphate synthase gene from a halophyte *Spartina alterniflora* confers salt tolerance in transgenic tobacco and rice. Plant and Animal Genome

XVII, San Diego, CA, Jan 10–14 2009, Poster No. 616, Final abstract guide pp 117

Bhatnagar-Mathur P, Vadez V, Sharma KK (2008) Transgenic approaches for abiotic stress tolerance in plants, retrospect and prospects. Plant Cell Rep 27: 411–424

Bonilla P, Dvorak J, Mackill DJ, Deal K, Gregorio GB (2002) RFLP and SSLP mapping of salinity tolerance genes in chromosome 1 of rice (*Oryza sativa* L.) using recombinant inbred lines. Philipp Agric Sci 85:68–76

Brini F, Hanin M, Mezghani I, Berkowitz GA, Masmoudi K (2007) Overexpression of wheat Na^+/H^+ antiporter *TNHX1* and H^+-pyrophosphatase *TVP1* improve salt- and drought-stress tolerance in *Arabidopsis thaliana* plants. J Exp Bot 58:301–308

Byrt CS, Platten JD, Spielmeyer W, James RA, Lagudah ES, Dennis ES, Tester T, Munns R (2007) *HKT1,5*-like cation transporters linked to Na^+ exclusion loci in wheat, *Nax2* and *Kna1*. Plant Physiol 143:1918–1928

Chen GP, Ma WS, Huang ZJ, Xu T, Xue YB, Shen YZ (2003) Isolation and characterization of *TaGSK1* involved in wheat salt tolerance. Plant Sci 165: 1369–1375

Chitteti BR, Peng Z (2007) Proteome and phosphoproteome differential expression under salinity stress in rice (*Oryza sativa*) roots. J Proteome Res 6:1718–1727

Collard BCY, Mackill DJ (2008) Marker assisted selection: an approach for precision plant breeding in the twenty-first century. Phil Trans Roy Soc Lond B Biol Sci 363:557–572

Collard BCY, Jahufer MZZ, Brouwer JB, Pang ECK (2005) An introduction to markers, quantitative trait loci (QTL) mapping and marker-assisted selection for crop improvement: the basic concepts. Euphytica 142:169–196

Collard BCY, Veracruz CM, McNally KL, Virk PS, Mackill DJ (2008) Rice molecular breeding laboratories in the genomics era: current status and future considerations. Int J Plant Genomics 2008:524847. doi:10: 1155/2008/524847

Collins NC, Tardieu F, Tuberosa R (2008) Quantitative trait loci and crop performance under abiotic stress: where do we stand? Plant Phys 147:469–486

Colmer TD, Munns R, Flowers TJ (2005) Improving salt tolerance of wheat and barley: future prospects. Aust J Exp Agric 45:1425–1443

Colmer TD, Flowers TJ, Munns R (2006) Use of wild relatives to improve salt tolerance in wheat. J Exp Bot 57:1059–1078

Cotsaftis O, Plett D, Johnson AAT, Walia H, Wilson C, Ismail AM, Close T, Tester M, Baumann U (2011) Root-specific transcript profiling of contrasting rice genotypes in response to salinity stress. Mol Plant 4:25–41

Cuin TA, Tian Y, Betts SA, Chalmandrier R, Shabala S (2009) Ionic relations and osmotic adjustment in durum and bread wheat under saline conditions. Funct Plant Biol 36:110–119

Cuin TA, Parsons D, Shabala S (2010) Wheat cultivars can be screened for salinity tolerance by measuring leaf chlorophyll content and shoot sap potassium. Funct Plant Biol 37:255–263

Das-Chatterjee A, Goswami L, Maitra S, Ghosh-Dastidar K, Ray S, Majumder AL (2006) Introgression of a novel salt-tolerant L-*myo* inositol 1-phosphate synthase from *Porteresia coarctata* (Roxb.) Tateoka (*PcINO1*) confers salt tolerance to evolutionary diverse organisms. FEBS Lett 580:3980–3988

Davenport R, James RA, Zakrisson-Plogander A, Tester M, Munns R (2005) Control of sodium transport in durum wheat. Plant Physiol 137:807–818

Dubcovsky J, Santa-Maria G, Epstein E, Luo MC, Dvo ák J (1996) Mapping of the K/Na discrimination locus *Kna1* in wheat. Theor Appl Genet 92:448–454

Dubouzet JG, Sakuma Y, Ito Y, Kasuga M, Dubouzet EG, Miura S, Seki M, Shinozaki K, Yamaguchi-Shinozaki K (2003) OsDREB genes in rice, *Oryza sativa* L., encode transcription activators that function in drought, high salt and cold-responsive gene expression. Plant J 33:751–763

Dasgupta M, Sahoo MR, Kole PE, Mukherjce A (2008) Evaluation of orange-fleshed sweet potato (I pomea betatas L) genotypes for salt tolerance through shoot apex culture under in vitro Nace mediated salinity stress condition. Plant Cell Tiss. Organ Culture 94:161–170

Dvorak J, Noaman MM, Goyal S, Gorham J (1994) Enhancement of the salt tolerance of *Triticum turgidum* L by the *Kna1* locus transferred from *Triticum aestivum* L. chromosome 4D by homoeologous recombination. Theor Appl Genet 87:872–877

Dwivedi S, Upadhyaya H, Subudhi PK, Gehring C, Bajic V, Ortiz R (2010) Enhancing abiotic stress tolerance in cereals through breeding and transgenic interventions. Plant Breed Rev 33:31–114

Dziadczyk P, Bolibok H, Tyrka M, Hortynski JA (2003) In vitro selection of strawberry (*Fragaria* × *ananassa* Duch.) clones tolerant to salt stress. Euphytica 132:49–55

El-Hendawy SE, Hu Y, Schmidhalter U (2007) Assessing the suitability of various physiological traits to screen wheat genotypes for salt tolerance. J Integr Plant Biol 49:1352–1360

Ellis RP, Forster BPP, Gordon DC, Handley LL, Keith RP, Lawrence P, Meyer R, Powell W, Robinson D, Scrimgeour CM, Young G, Thomas WTB (2002) Phenotype/genotype associations of yield and salt tolerance in a barley mapping population segregating for two dwarfing genes. J Exp Bot 53:1163–1176

FAO (2010) FAO Land and plant nutrition management service. http://www.fao.org

Feuillet C, Langridge P, Waugh R (2008) Cereal breeding takes a walk on the wild side. Trends Genet 24:24–32

Flowers TJ (2004) Improving crop salt tolerance. J Exp Bot 55:307–319

Flowers TJ, Colmer TD (2008) Salinity tolerance in halophytes. New Phytol 179:945–963

Flowers TJ, Flowers SA (2005) Why does salinity pose such a difficult problem for plant breeders? Agric Water Management 78:15–24

Forster BP, Thomas WTB (2005) Doubled haploids in genetics and plant breeding. Plant Breed Rev 25: 57–88

Fukuda A, Nakamura A, Tagiri A, Tanaka H, Miyao A, Hirochika H, Tanaka Y (2004) Function, intracellular localization and the importance in salt tolerance of a vacuolar Na^+/H^+ antiporter from rice. Plant Cell Physiol 45:146–159

Gandonou C, Errabii T, Abrini J, Idaomar M, Senhaji N (2006) Selection of callus cultures of sugarcane (*Saccharum* sp.) tolerant to NaCl and their response to salt stress. Plant Cell Tiss Org Cult 87:9–16

Gao JP, Chao DY, Lin HX (2007) Understanding abiotic stress tolerance mechanisms: recent studies on stress response in rice. J Integr Plant Biol 49:742–750

Garg AK, Kim JK, Owens TG, Ranwala AP, Choi YD, Kochian LV, Wu R (2002) Trehalose accumulation in rice plants confers high tolerance levels to different abiotic stresses. Proc Natl Acad Sci USA 99: 15898–15903

Garthwaite AJ, von Bothmer R, Colmer TD (2005) Salt tolerance in wild Hordeum species is associated with restricted entry of Na^+ and Cl^- into the shoots. J Exp Bot 56:2365–2378

Genc Y, Oldach K, Verbyla AP, Lott G, Hassan M, Tester M, Wallwork H, McDonald GK (2010) Sodium exclusion QTL associated with improved seedling growth in bread wheat under salinity stress. Theor Appl Genet 121:877–894

Germanà MA (2010) Anther culture for haploid and doubled haploid production. Plant Cell Tiss Organ Cult 104:283–300

Ghassemi F, Jakeman A, Nix HA (1995) Salinisation of land and water resources: human causes, extent, management and case studies. UNSW Press, Sydney, Australia

Gorham J (1994) Salt tolerance in the Triticeae: K/Na discrimination in some perennial wheat grasses and their amphiploids with wheat. J Exp Bot 45:441–447

Gorham J, Hardy C, WynJones RG, Joppa LR, Law CN (1987) Chromosomal location of a K/Na discrimination character in the D genome of wheat. Theor Appl Genet 74:584–588

Gorham J, Bridges J, Dubcovsky J, Dvo ák J, Hollington PA, Luo MC, Khan JA (1997) Genetic analysis and physiology of a trait for enhanced K^+/Na^+ discrimination in wheat. New Phytol 137:109–116

Greenway H, Munns R (1980) Mechanisms of salt tolerance in non-halophytes. Ann Rev Plant Physiol 31:149–190

Gregorio GB, Senadhira D, Mendoza RD (1997) Screening rice for salinity tolerance. IRRI Discussion Paper Series 22. International Rice Research Institute, P.O. Box 933, Manila 1099, Philippines

Gregorio GB, Senadhira D, Mendoza RD, Manigbas NL, Roxas JP, Guerta CQ (2002) Progress in breeding for associated salinity tolerance and associated traits in rice. Field Crops Res 76:91–101

Gu L, Liu Y, Zong X, Liu L, Li DP, Li DQ (2010) Overexpression of maize mitogen-activated protein kinase gene, *ZmSIMK1* in Arabidopsis increases tolerance to salt stress. Mol Biol Rep 37:4067–4073

Haq TU, Gorham J, Akhtar J, Akhtar N, Steele KA (2010) Dynamic quantitative trait loci for salt stress components on chromosome 1 of rice. Funct Plant Biol 37:634–645

Hasthanasombut S, Supaibulwatana K, Mii M, Nakamura I (2011) Genetic manipulation of *Japonica* rice using the *OsBADH1* gene from *Indica* rice to improve salinity tolerance. Plant Cell Tiss Organ Cult 104:79–89

Hauser F, Horie T (2010) A conserved primary salt tolerance mechanism mediated by HKT transporters: a mechanism for sodium exclusion and maintenance of high K^+/Na^+ ratio in leaves during salinity stress. Plant Cell Environ 33:552–565

He X, Hou X, Shen Y, Huang Z (2011) *TaSRG*, a wheat transcription factor, significantly affects salt tolerance in transgenic rice and Arabidopsis. FEBS Lett 585:1231–1237

Horie T, Schroeder JI (2004) Sodium transporters in plants. Diverse genes and physiological functions. Plant Physiol 136:2457–62

Houshmand S, Arzani A, Maibody SAM, Feizi M (2005) Evaluation of salt-tolerant genotypes of durum wheat derived from in vitro and field experiments. Field Crops Res 91:345–354

Hu H, Dai M, Yao J, Xiao B, Li X, Zhang Q, Xiong L (2006) Overexpressing a NAM, ATAF, and CUC (NAC) transcription factor enhances drought resistance and salt tolerance in rice. Proc Natl Acad Sci USA 35:12987–12992

Huang S, Spielmeyer W, Lagudah ES, James RA, Platten RD, Dennis ES, Munns R (2006) A sodium transporter (*HKT7*) is a candidate for *Nax1*, a gene for salt tolerance in durum wheat. Plant Physiol 142:1718–1727

Huang W, Ma X, Wang Q, Gao Y, Xue Y, Niu X, Yu G, Liu Y (2008) Significant improvement of stress tolerance in tobacco plants by overexpressing a stress-responsive aldehyde dehydrogenase gene from maize (*Zea mays*). Plant Mol Biol 68:451–463

Husain S, von Caemmerer S, Munns R (2004) Control of salt transport from roots to shoots of wheat in saline soil. Funct Plant Biol 31:1115–1126

Inan G, Zhang Q, Li P, Wang Z, Cao Z, Zhang H, Zhang C, Quist TM, Goodwin SM, Zhu J, Shi H, Damsz B, Charbaji T, Gong Q, Ma S, Fredricksen M, Galbraith DW, Jenks MA, Rhodes D, Hasegawa PM, Bohnert HJ, Joly RJ, Bressan RA, Zhu JK (2004) Salt Cress. A halophyte and cryophyte Arabidopsis relative model system and its applicability to molecular genetic analyses of growth and development of extremophiles. Plant Physiol 135:1–20

IPCC (Intergovernmental Panel on Climate Change) (2007) Climate change 2007: the physical science basis (Summary for policy makers). IPPC Secretariat, WMO, Geneva, Switzerland, p 21

Ismail AM, Heuer S, Thomson MJ, Wissuwa M (2007) Genetic and genomic approaches to develop rice germplasm for problem soils. Plant Mol Biol 65:547–570

Ito Y, Katsura K, Maruyama K, Taji T, Kobayashi M, Seki S, Shinozaki K, Yamaguchi-Shinozaki K (2006) Functional analysis of rice DREB1/CBF type transcription factors involved in cold responsive gene expression in transgenic rice. Plant Cell Physiol 47:141–153

Jacobs A, Ford K, Kretschmer J, Tester M (2011) Rice plants expressing the moss sodium pumping ATPase *PpENA1* maintain greater biomass production under salt stress. Plant Biotech J. doi:10.1111/j.1467-7652.2011.00594.x

James RA, Davenport RJ, Munns R (2006) Physiological characterization of two genes for Na+exclusion in durum wheat, Nax1 and Nax2. Plant Physiol 142: 1537–1547

James RA, Blake C, Byrt CS, Munns R (2011) Major genes for Na^+ exclusion, *Nax1* and *Nax2* (wheat *HKT1,4* and *HKT1,5*), decrease Na^+ accumulation in bread wheat leaves under saline and waterlogged conditions. J Exp Bot. doi:10.1093/jxb/err003

Jha B, Sharma A, Mishra A (2010) Expression of *SbGSTU* (*tau* class glutathione S-transferase) gene isolated from *Salicornia brachiata* in tobacco for salt tolerance. Mol Biol Rep. 38:4823–4832

Jithesh MN, Prashanth SR, Sivaprakash KR, Parida AK (2006) Monitoring expression profiles of antioxidant genes to salinity, iron, oxidative, light and hyperosmotic stresses in highly salt tolerant grey mangrove, *Avicennia marina* (Forsk.) Vierh by mRNA analysis. Plant Cell Rep 25:865–876

Karaba A, Dixit S, Greco R, Aharoni A, Trijatmiko KR, Marsch-Martinez N, Krishnan A, Nataraja KB, Udayakumar M, Pereira A (2007) Improvement of water use efficiency in rice by expression of *HARDY*, an *Arabidopsis* drought and salt tolerance gene. Proc Natl Acad Sci USA 104:15270–15275

Karan R, Singla-Pareek SL, Pareek A (2009) Histidine kinase and response regulator genes as they relate to salinity tolerance in rice. Funct Integr Genomics 9:411–417

Khan MA, Shirazi MU, Ali M, Mumtaz S, Sherin A, Ashraf MY (2006) Comparative performance of some wheat genotypes growing under saline water. Pak J Bot 38:1633–1639

Kijne JW (2006) Abiotic stress and water scarcity: identifying and resolving conflicts from plant level to global level. Field Crops Res 97:3–18

Kong X, Pan J, Zhang M, Xing X, Zhou Y, Liu Y, Li D, Li D (2011) *ZmMKK4*, a novel group C MAPK kinase in maize (*Zea mays*), confers salt and cold tolerance in transgenic Arabidopsis. Plant Cell Environ. doi:10.1111/j.1365-3040.2011.02329.x

Koyama ML, Levesley A, Koebner RMD, Flowers TJ, Yeo AR (2001) Quantitative trait loci for component physiological traits determining salt tolerance in rice. Plant Physiol 125:406–422

Kumari S, Sabharwal VP, Kushwaha HR, Sopory SK, Singla-Pareek SL, Pareek A (2009) Transcriptome map for seedling stage specific salinity stress response indicates a specific set of genes as candidate for saline tolerance in *Oryza sativa* L. Funct Integr Genomics 9:109–123

Lang NT, Yanagihara S, Buu BC (2001) A microsatellite marker for a gene conferring salt tolerance on rice at the vegetative and reproductive stages. SABRAO J Breed Genet 33:1–10

Lee KS, Choi WR, Ko JC, Kim TS, Gregoria GP (2003a) Salimity tolerance of *japonica* and *indica* (*Bryza sativa*) at the seedling stage. Planta 216:1043–1046

Lee SY, Lee JH, Kwon TO (2003b) Selection of salt-tolerant doubled haploids in rice anther culture. Plant Cell Tiss Organ Cult 74:143–149

Lee SY, Ahn JH, Cha YS, Yun DW, Lee MC, Ko JC, Lee KS, Eun MY (2007) Mapping QTLs related to salinity tolerance of rice at the young seedling stage. Plant Breed 126:43–46

Li JY, He XW, Xu L, Zhou J, Wu P, Shou HX, Zhang FC (2008) Molecular and functional comparisons of the vacuolar Na^+/H^+ exchangers originated from glycophytic and halophytic species. J Zhejiang Univ Sci B 9:132–140

Li WH, Zhang Q, Kong XQ, Wu CX, Ma XL, Zhang H, Zhao YX (2009) Salt tolerance is conferred in Arabidopsis by over-expression of the vacuolar Na^+/H^+ antiporter gene *SsNHX2*, an alternative splicing variant of *SsNHX1*, from *Suaeda salsa*. J Plant Biol 52:147–153

Li F, Guo S, Zhao Y, Chen D, Chong K, Xu Y (2010a) Overexpression of a homopeptide repeat-containing bHLH protein gene (*OrbHLH001*) from Dongxiang wild rice confers freezing and salt tolerance in transgenic Arabidopsis. Plant Cell Rep 29:977–986

Li TX, Zhang Y, Liu H, Wu YT, Li WB, Zhang HX (2010b) Stable expression of Arabidopsis vacuolar Na^+/H^+ antiporter gene *AtNHX1*, and salt tolerance in transgenic soyabean for over six generations. Chinese Sci Bull 55:1127–1134

Lin HX, Zhu MZ, Yano M, Gao JP, Liang ZW, Su WA, Hu XH, Ren ZH, Chao DY (2004) QTLs for Na^+ and K^+ uptake of the shoots and roots controlling rice salt tolerance. Theor Appl Genet 108:253–260

Lindsay MP, Lagudah ES, Hare RA, Munns R (2004) A locus for sodium exclusion (*Nax1*), a trait for salt tolerance, mapped in durum wheat. Funct Plant Biol 31:1105–1114

Lisa LA, Seraj ZI, Elahi CMF, Das KC, Biswas K, Islam MR, Salam MA, Gomosta AR (2004) Genetic variation in microsatellite DNA, physiology and morphology of coastal saline rice (*Oryza sativa* L.) landraces of Bangladesh. Plant Soil 263:213–228

Liu X, Baird WV (2003) Differential expression of genes regulated in response to drought or salinity stress in sunflower. Crop Sci 43:678–687

Lu SY, Peng XX, Guo ZF, Zhang GY, Wang ZC, Wang CY, Pang CS, Fan Z, Wang JH (2007) In vitro selection of salinity tolerant variants from triploid bermudagrass (*Cynodon transvaalensis* × *C-dactylon*) and their physiological responses to salt and drought stress. Plant Cell Rep 26:1413–1420

Lv S, Zhang KW, Gao Q, Lian LJ, Song YJ, Zhang JR (2008) Overexpression of an H^+-PPase gene from *Thellungiella halophila* in cotton enhances salt tolerance and improves growth and photosynthetic performance. Plant Cell Physiol 49:1150–1164

Ma L, Zhou E, Huo N, Zhou R, Wang G, Jia J (2007) Genetic analysis of salt tolerance in a recombinant inbred population of wheat (*Triticum aestivum* L.). Euphytica 153:109–117

Maas EV, Hoffman GJ (1977) Crop salt tolerance-current assessment. J Irrig Drain Div ASCE 103:115–134

Mahalakshmi S, Christopher GSB, Reddy TP, Rao KV, Reddy VD (2006) Isolation of a cDNA clone (PcSrp) encoding serine-rich-protein from *Porteresia coarctata* T. and its expression in yeast and finger millet (*Eleusine coracana* L.) affording salt tolerance. Planta 224: 347–359

Majee M, Maitra S, Ghosh Dastidar K, Pattnail S, Chatterjee A, Hait NC, Das KP, Majumdar AL (2004) A novel salt-tolerant l-myo-inositol-1-phosphate synthase from *Porteresia coarctata* (Roxb.) Tateoka, a halophytic wild rice: molecular cloning, bacterial overexpression, characterization, and functional introgression into tobacco-conferring salt tolerance phenotype. J Biol Chem 279:28539–28552

Manneh B, Stam P, Struik PC, Bruce-Oliver S, van Eeuwijk FA (2007) QTL-based analysis of genotype-by-environment interaction for grain yield of rice in stress and non-stress environments. Euphytica 156:213–226

Mano Y, Takeda K (1997) Mapping quantitative trait loci for salt tolerance at germination and the seedling stage in barley (*Hordeum vulgare* L.). Euphytica 94: 263–272

Mao X, Zhang H, Tian S, Chang X, Jing R (2010) TaSnRK2.4, an SNF1-type serine/threonine protein kinase of wheat (*Triticum aestivum* L.), confers enhanced multistress tolerance in Arabidopsis. J Exp Bot 61:683–696

Mao X, Jia D, Li A, Zhang H, Tian S, Zhang X, Jia J, Jing R (2011) Transgenic expression of *TaMYB2A* confers enhanced tolerance to multiple abiotic stresses in Arabidopsis. Funct Integr Genomics. doi:10.1007/s10142-011-0218-3

Melloul A, Collin M (2006) Hydrogeological changes in coastal aquifers due to sea level rise. Ocean Coastal Management 49:281–297

Munns R (2002) Comparative physiology of salt and water stress. Plant Cell Environ 25:239–250

Munns R, James RA (2003) Screening methods for salinity tolerance: a case study with tetraploid wheat. Plant Soil 253:201–218

Munns R, Tester M (2008) Mechanisms of salinity tolerance. Annu Rev Plant Biol 59:651–681

Munns R, Hare RA, James RA, Rebetzke GJ (2000) Genetic variation for improving the salt tolerance of durum wheat. Aust J Agric Res 51:69–74

Munns R, Rebetzke GJ, Husain S, James RA, Hare RA (2003) Genetic control of sodium exclusion in durum wheat. Aust J Agr Res 54:627–635

Nakashima K, Tran LS, Van Nguyen D, Fujita M, Maruyama K, Todaka D, Ito Y, Hayashi N, Shinozaki K, Yamaguchi-Shinozaki K (2007) Functional analysis of a NAC-type transcription factor *OsNAC6* involved in abiotic and biotic stress-responsive gene expression in rice. Plant J 51:617–630

Obata T, Kitamoto HK, Nakamura A, Fukuda A, Tanaka Y (2007) Rice shaker potassium channel *OsKAT1* confers tolerance to salinity stress on yeast and rice cells. Plant Physiol 144:1978–1985

Oh SJ, Song SI, Kim YS, Jang HJ, Kim M, Kim YK (2005) *Arabidopsis* CBF3/DREB1A and ABF3 in transgenic rice increased tolerance to abiotic stress without stunting growth. Plant Physiol 138: 341–351

Oh SJ, Chang-Woo Kwon CW, Dong-Woog Choi DW, Sang Ik Song SI, Kim JK (2007) Expression of barley HvCBF4 enhances tolerance to abiotic stress in transgenic rice. Plant Biotech J 5:646–656

Ohta M, Hayashi Y, Nakashima A, Hamada A, Tanaka A, Nakamura T, Hayakawa T (2002) Introduction of a Na^+/H^+ antiporter gene from *Atriplex gmelini* confers salt tolerance to rice. FEBS Lett 532:279–82

Ortel B, Atzorn R, Hause B, Feussner I, Miersch O, Wasternack C (1999) Jasmonate-induced gene expression of barley (*Hordeum vulgare*) leaves the link between jasmonate and abscisic acid. Plant Growth Regul 29:113–122

Ozturk ZN, Talame V, Deyhoyos M, Michalowski CB, Galbraith DW, Gozukirmizi N, Tuberosa R, Bohnert HJ (2002) Monitoring large-scale changes in transcript abundance in drought-and salt-stressed barley. Plant Mol Biol 48:551–573

Pandit A, Rai V, Bal S, Sinha S, Kumar V, Chauhan M, Gautam RK, Singh R, Sharma PC, Singh AK, Gaikwad K, Sharma TR, Mohapatra T, Singh NK (2010) Combining QTL mapping and transcriptome profiling of bulked RILs for identification of functional polymorphism for salt tolerance genes in rice (*Oryza sativa* L.). Mol Genet Genom 284:121–136

Prashanth SR, Sadhasivam V, Parida A (2008) Over expression of cytosolic copper/zinc superoxide dismutase from a mangrove plant *Avicennia marina* in *indica* Rice var Pusa Basmati-1 confers abiotic stress tolerance. Transgenic Res 17:281–291

Qiu Y, Li X, Zhi H, Shen D, Lu P (2009) Differential expression of salt tolerance related genes in *Brassica campestris* L. ssp. *chinensis* (L.) Makino var. *communis* Tsen et Lee. J Zhejiang Univ Sci B 10:847–851

Quarrie SA, Steed A, Calestani C, Semikhodskii A, Lebreton C, Chinoy C, Steele N, Pljevljakusi D, Waterman E, Weyen J, Schondelmaier J, Habash DZ, Farmer P, Saker L, Clarkson DT, Abugalieva A, Yessimbekova M, Turuspekov Y, Abugalieva S, Tuberosa R, Sanguineti M-C, Hollington PA, Aragués R, Royo A, Dodig D (2005) A high-density genetic map of hexaploid wheat (*Triticum aestivum* L.) from the cross Chinese Spring x SQ1 and its use to compare QTLs for grain yield across a range of environments. Theor Appl Genet 110:865–880

Queirs F, Fidalgo F, Santos I, Salema R (2007) In vitro selection of salt tolerant cell lines in *Solanum tuberosum* L. Biol Plant 51:728–734

Rahman MS, Das KC, Das DK, Biswas K, Chowdhury MBH, Karim NH, Salam MA, Seraj ZI (2010) Breeding and anther derived lines of rice (*Oryza sativa* L.) for saline coastal areas of Bangladesh. Bangladesh J Bot 39:71–78

Rahnama A, Poustini K, Munns R, James RA (2010) Stomatal conductance as a screen for osmotic stress tolerance in durum wheat growing in saline soil. Funct Plant Biol 37:255–263

Ren Z, Gao J, Li L, Cai X, Huang W, Chao D, Zhu M, Wang Z, Luan S, Lin H (2005) A rice quantitative trait locus for salt tolerance encodes a sodium transporter. Nat Genet 37:1141–1146

Rengasamy P (2010) Soil processes affecting crop production in salt affected soils. Funct Plant Biol 37: 613–620

Ruan SL, Ma HS, Wang SH, Fu YP, Xin Y, Liu WZ, Wang F, Tong JX, Wang SZ, Chen HZ (2011) Proteomic identification of *OsCYP2*, a rice cyclophilin that confers salt tolerance in rice (*Oryza sativa* L.) seedlings when overexpressed. BMC Plant Biol 11:34. doi:10.1186/1471-2229-11-34

Sabouri H, Sabouri A (2008) New evidence of QTLs attributed to salinity rice. Afr J Biotech 7:4376–4383

Sahi C, Agarwal M, Reddy MK, Sopory SK, Grover A (2003) Isolation and expression analysis of salt stress-associated ESTs from contrasting rice cultivars using a PCR-based subtraction method. Theor Appl Genet 106:620–628

Schachtman DP, Munns R, Whitecross MI (1991) Variation of sodium exclusion and salt tolerance in *Triticum tauschii*. Crop Sci 31:992–997

Senadhira D, Zapata-Arias FJ, Gregorio GB, Alejar MS, de la Cruz HC, Padolina TF, Galvez AM (2002) Development of the first salt-tolerant rice cultivar through *indica/indica* anther culture. Field Crops Res 76:103–110

Sengupta S, Majumder AL (2009) Insight into the salt tolerance factors of a wild halophytic rice, *Porteresia coarctata*: a physiological and proteomic approach. Planta 229:911–929

Shah SH, Gorham J, Forster BP, Wyn Jones RG (1987) Salt tolerance in the Triticeae: the contribution of the D-genome to cation selectivity in hexaploid wheat. J Exp Bot 38:254–269

Shavrukov Y, Gupta NK, Miyazaki J, Baho MN, Chalmers KJ, Tester M, Langridge P, Collins NC (2010) *HvNax3*-a locus controlling shoot sodium exclusion derived from wild barley (*Hordeum vulgare ssp. spontaneum*). Funct Integr Genomics 10:277–291

Singh RK, Mishra B (1995) Screening F_1 anther culture derivatives of rice for salt tolerance. In: Sharma B et al. (eds) Genetic research and education: current trends and the next fifty years. Indian Society of Genetics and Plant Breeding New Delhi. Ind J Genet Plant Breed (Special Issue): 509–513

Singh RK, Mishra B, Senadhira D (1992) Promising salt tolerant F_1 anther culture derivatives (ACDs). Int Rice Res Newslett 17:17

Singla-Pareek SL, Yadav SK, Pareek A, Reddy MK, Sopory SK (2008) Enhancing salt tolerance in a crop plant by overexpression of glyoxalase II. Transgenic Res 17:171–180

Subudhi PK, Baisakh N (2011) *Spartina alterniflora* Loisel., a halophyte grass model to dissect salt stress tolerance. In Vitro Cell Dev Biol-Plant 47:441–457

Sun ZB, Qi XY, Li PH, Wu CX, Zhao YX, Zhang H, Wang ZL (2008) Overexpression of a *Thellungiella halophila* CBL9 homolog, *ThCBL9*, confers salt and osmotic tolerances in transgenic *Arabidopsis thaliana*. J Plant Biol 51:25–34

Taiz L (1992) The plant vacuole. J Exp Biol 172: 113–122

Tajbakhsh M, Zhou M, Chen Z, Mendham NJ (2006) Physiological and cytological response of salt-tolerant and non-tolerant barley to salinity during germination and early growth. Aust J Exp Agr 46:555–562

Taji T, Seki M, Satou M, Sakurai T, Kobayashi M, Ishiyama K, Narusaka Y, Narusaka M, Zhu JK, Shinozaki K (2004) Comparative genomics in salt tolerance between Arabidopsis and Arabidopsis-related halophyte salt cress using Arabidopsis microarray. Plant Physiol 135:1697–1709

Takasaki H, Maruyama K, Kidokoro S, Ito Y, Fujita Y, Shinozaki K, Yamaguchi-Shinozaki K, Nakashima K (2010) The abiotic stress-responsive NAC-type transcription factor OsNAC5 regulates stress-inducible genes and stress tolerance in rice. Mol Genet Genomics 284:173–183

Takeda S, Matsuoka M (2008) Genetic approaches to crop improvement: responding to environmental and population changes. Nat Rev Genet 9:444–457

Takehisa H, Shimodate T, Fukuta Y, Ueda T, Yano M, Yamaya T, Kameya T, Sato T (2004) Identification of quantitative trait loci for plant growth of rice in paddy field flooded with salt water. Field Crops Res 89:85–95

Tanksley SD, McCouch SR (1997) Seed banks and molecular maps: unlocking genetic potential from the wild. Science 277:1063–1066

Thalji T, Shalaldeh G (2007) Screening wheat and barley genotypes for salinity tolerance. J Agron 6:75–80

Thomson MJ, de Ocampo M, Egdane J, Rahman MA, Sajise AG, Adorada DL, Tumimbang-Raiz E, Blumwald E, Seraj ZI, Singh RK, Gregorio GB, Ismail AM (2010) Characterizing the *Saltol* quantitative trait locus for salinity tolerance in rice. Rice 3: 148–160

USDA-ARS (2005) Agricultural weather facility, Global crop production review. 1–5

USDA-ARS (2008) Research Databases. (http://www.ars.usda.gov/Services/docs.htm?docid=8908)

Verma D, Singla-Pareek SL, Rajagopal D, Reddy MK, Sopory SK (2007) Functional validation of a novel isoform of Na^+/H^+ antiporter from *Pennisetum glaucum* for enhancing salinity tolerance in rice. J Biosci 32:621–628

Verslues PE, Agarwal M, Katiyar-Agarwal S, Zhu J, Zhu JK (2006) Methods and concepts in quantifying resistance to drought, salt and freezing, abiotic stresses that affect plant water status. Plant J 45:523–539

Walia H, Wilson C, Condamine P, Liu X, Ismail AM, Zeng L, Wanamaker SI, Mandal J, Xu J, Cui X, Close TJ (2005) Comparative transcriptional profiling of two contrasting rice genotypes under salinity stress during the vegetative growth stage. Plant Physiol 139: 822–835

Walia H, Wilson C, Wahid A, Condamine P, Cui X, Close TJ (2006) Expression analysis of barley (*Hordeum vulgare* L.) during salinity stress. Funct Integr Genomics 6:143–156

Walia H, Wilson C, Zeng L, Ismail AM, Condamine P, Close TJ (2007) Genome-wide transcriptional analysis of salinity stressed *japonica* and *indica* rice genotypes during panicle initiation stage. Plant Mol Biol 63:609–623

Wang QY, Guan YC, Wu YR, Chen HL, Chen F, Chu CC (2008) Overexpression of a rice *OsDREB1F* gene increases salt, drought and low temperature tolerance in both *Arabidopsis* and rice. Plant Mol Biol 67:589–602

Wang ZF, Wang JF, Bao YM, Wu YY, Zhang HS (2011) Quantitative trait loci controlling rice seed germination under salt stress. Euphytica 178:297–307

Wassmann F, Hien NX, Hoanh CT, Tuong TP (2004) Sea level rise affecting the Vietnamese Mekong Delta: water elevation in the flood season and implications for rice production. Climate Change 66:89–107

Wasternack C, Miersch O, Kramell R, Hause B, Ward J, Beale M, Boland W, Parthier B, Feussner I (1998) Jasmonic acid:biosynthesis, signal transduction, gene expression. Fett-Lipid 100:139–146

Wedzony M, Forster BP, Zur I, Golemiec E, Szechynska-Hebda M, Dubas E, Gotebiowska G, Wedzony M (2009) Progress in doubled haploid technology in higher plants. In: Touraev A, Forster BP, Jain SM (eds) Advances in haploid production in higher plants. Springer, Berlin, pp 1–34

Wei W, Bilsborrow P, Hooley P, Fincham D, Forster B (2001) Variation between two near isogenic barley *(Hordeum uulgare)* cultivars in expression of the B subunit of the vacuolar ATPase in response to salinity. Hereditas 135:227–231

Wheatley AO, Ahmad MH, Asemota HN (2003) Development of salt adaptation in in vitro greater yam (*Dioscorea alata*) plantlets. In Vitro Cell Dev Biol-Plant 39:346–353

Wild A (2003) Soils, land and food: managing the land during the twenty-first century. Cambridge University Press, Cambridge, UK

Witcombe JR, Hollington PA, Howarth CJ, Reader S, Steele KA (2008) Breeding for abiotic stresses for sustainable agriculture. Philos Trans R Soc B Biol Sci 363:703–716

Witzel K, Weidner A, Surabhi GK, Borner A, Mock HP (2009) Salt stress-induced alterations in the root proteome of barley genotypes with contrasting response towards salinity. J Exp Bot 60:3545–3557

Wu W, Su Q, Xia XY, Wang Y, Luan YS, An LJ (2008) The *Suaeda liaotungensis* kitag betaine aldehyde dehydrogenase gene improves salt tolerance of transgenic maize mediated with minimum linear length of DNA fragment. Euphytica 159:17–25

Wu C, Gao X, Kong X, Zhao Y, Zhang H (2009) Molecular cloning and functional analysis of a Na^+/H^+ antiporter gene *ThNHX1* from a halophytic plant *Thellungiella halophila*. Plant Mol Biol Rep 27:1–12

Xu GY, Rocha PSCF, Wang ML, Xu ML, Cui YC, Li LY, Zhu YX, Xia X (2011) A novel rice calmodulin-like gene, OsMSR2, enhances drought and salt tolerance and increases ABA sensitivity in Arabidopsis. Planta. doi:10.1007/s00425-011-1386-z

Xue ZY, Zhi DY, Xue GP, Zhang H, Zhao YX, Xia GM (2004) Enhanced salt tolerance of transgenic wheat (*Triticum aestivum* L.) expressing a vacuolar Na^+/H^+ antiporter gene with improved yields in saline soils in the field and a reduced level of leaf Na^+. Plant Sci 167:849–859

Xue D, Huang Y, Zhang X, Wei K, Westcott S, Li C, Chen M, Zhang G, Lance R (2009) Identification of QTLs associated with salinity tolerance at late growth stage in barley. Euphytica 169:187–196

Yan S, Tang Z, Su W, Sun W (2005) Proteomic analysis of salt stress-responsive proteins in rice root. Proteomics 5:235–244

Yoshida S, Forno DA, Cock JH, Gomez KA (1976) Laboratory manual for physiological studies of rice 3rd edition. International Rice Research Institutes, Manila, Philippines, p 61

Zair I, Chlyah A, Sabounji K, Tittahsen M, Chlyah H (2003) Salt tolerance improvement in some wheat cultivars after application of in vitro selection pressure. Plant Cell Tiss Org Cult 73:237–244

Zang J, Sun Y, Wang Y, Yang J, Li F, Zhou YL, Zhu LH, Reys J, Mohammadhosein F, Xu JL, Li Z (2008) Dissection of genetic overlap of salt tolerance QTLs at the seedling and tillering stages using backcross introgression lines in rice. Sci China C Life Sci 51:583–591

Zeng L, Lesch SM, Grieve CM (2003a) Rice growth and yield respond to changes in water depth and salinity stress. Agric Water Manage 59:67–75

Zeng L, Poss JA, Wilson C, Draz ASE, Gregorio GB, Grieve CM (2003b) Evaluation of salt tolerance in rice genotypes by physiological characters. Euphytica 129:281–292

Zhang L, Zhang L, Luo J, Chen W, Hao M, Liu B, Yan Z, Zhang B, Zhang H, Zheng Y, Liu D, Yen Y (2011a) Synthesizing double haploid hexaploid wheat populations based on a spontaneous alloploidization process. J Genet Genomics 38:89–94

Zhang X, Wang L, Meng H, Wen H, Fan Y, Zhao J (2011b) Maize *ABP9* enhances tolerance to multiple stresses in transgenic Arabidopsis by modulating ABA signaling and cellular levels of reactive oxygen species. Plant Mol Biol 75:365–378

Zhao FY, Zhang H (2006) Salt and paraquat stress tolerance results from co-expression of the *Suaeda salsa* glutathione S-transferase and catalase in transgenic rice. Plant Cell Tiss Org Cult 86:349–358

Zhu GY, Kinet JM, Bertin P, Bouharmont J, Lutts S (2000) Crosses between cultivars and tissue culture-selected plants for salt resistance improvement in rice, *Oryza sativa*. Plant Breed 119:497–504

Zhu JQ, Zhang JT, Tang RJ, Lv QD, Wang QQ, Yang L, Zhang HX (2009) Molecular characterization of *ThIPK2*, an inositol poly- phosphate kinase gene homolog from *Thellungiella halophila*, and its heterologous expression to improve abiotic stress tolerance in *Brassica napus*. Physiol Plantarum 136:407–425