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Salinity Stress in Crop Plants: Effects of stress, Tolerance Mechanisms and Breeding Strategies for Improvement

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Abstract

Salinity, whether primary or secondary, is among the most destructive abiotic stresses that disturb the plants from germination to physiological maturity. This problem is more severe in arid areas that get low yearly rainfall and are prone to high evapo-transpiration. Land under salinity stress is increasing on daily basis and it is thought that about half of the fertile land would become saline by the year 2050. The effects of salinity are highly diverse and depends on large number of factors like amount, intensity and duration of salinity and crop growth stages. Increased uptake of toxic ions couples with limited uptake of essential minerals resulting in significant reduction in enzymatic activity and disturbance in cell metabolism. Moreover, increased solute potential and reactive oxygen species inside the cells act as secondary damaging factors to physiological process and plant anatomy. Plants struggle to avoid or escape the stress by closing the stomata which results in stunted growth, wilting of plants and reduced productivity. Germplasm can be evaluated using different methods to access the salinity tolerance and their further administration in different breeding programs could develop tolerant cultivars.

Key Words: Drought Tolerance Indices, PCA, Radar Diagram, Yield and Yield Components

Introduction

Abiotic stress can be referred to as the environmental conditions that decrease plant growth and yield from optimum level (Skirycz and Inze, 2010). There are a number of types of biotic and abiotic stresses which cause significant reduction in plant productivity (Aslam *et al.*, 2013 a, b, c; Aslam *et al.*, 2014 a, b; Aslam *et al.*, 2015 a, b, c; Aslam *et al.*, 2016; Maqbool *et al.*, 2015 a, b; Maqbool *et al.*, 2016; Naveed *et al.*, 2014). Among the abiotic stresses, the most common are drought, heat, salinity, water logging etc. (Nishida and Murata, 1996). Subsoil constraints can be physical (like compaction or gravel layers), chemical (like salinity, sodicity, acidity, nutrient deficiencies and toxicities) or biological (high or low microbial activity) and have the ability to limit the availability of water and nutrients, thus having a damaging effect on plant growth (Dang *et al.*, 2006). Plants respond to abiotic stresses in various complex ways (Cramer, 2010). This depends upon the intensity of stress i.e. whether it is acute or chronic as well as duration to which plant is exposed to stress (Tattersall *et al.*, 2007; Pinheiro and Chaves, 2011). Reduction in arable land is another factor contributing to yield reduction of agricultural crops caused by limited water resources, climate change and global warming (Lobell *et al.*, 2011). Due to increasing global population and excessive exploitation of natural resources, a number of problems have been emerged. These include water scarcity, salt stress, acidity of soil, high temperature, flooding and diseases by different pathogens. Due to such environmental stresses, productivity of crops has

been drastically reduced. It has been estimated that about 20% yield losses are caused by salt stress. About 17% crop reduction is by drought stress, 40% by elevated temperature, 20% by low temperature, and other factors contribute to 8% reduction in crop yield (Rehman *et al.*, 2005; Ashraf *et al.*, 2008). Overall, various abiotic stresses may cause reduction in annual yield of crops by 51-82 % at global level (Bray *et al.*, 2000).

Salinity Stress

Salt stress explains the presence of soluble salts in excessive amount in the soil which alters plant's normal physiological processes and hinders plant growth. Irrigation water or soil solution with electric conductivity of 4 dsm⁻¹ or greater (Cramer, 1993) which is equal to 40 mM NaCl is considered as saline one. Environmental contaminants specifically those containing salt radicals affect crops adversely (Kijne, 2006). Building up of soluble salts in soil column causes serious damage to agricultural production and environmental health (Rengasamy, 2006). It has been estimated that up to 2050, about 50% of fertile land may be lost due to environmental constraints (Manchanda and Garg, 2008). In order to cope with the increasing demands of growing population, 38% increase in food production is needed by the year 2025 that would further rise to 50% by the year 2050. There are many factors which are limiting the global demands mainly by physical and chemical degradation of soil due to erosion and salinity (Wild, 2003).

On the basis of source, salinity has been divided into primary and secondary salinity. The main source of primary salinity is the decay of rocks that releases variety of salts. Secondary salinity is caused by anthropogenic activities i.e. irrigation, overgrazing, deforestation and intensive cropping etc. (Ashraf, 1994).

Global scenario of salinity stress

Salinity is major issue of irrigated areas. Irrigation is practiced on approximately 17% of the cultivated land that adds to 30% of global agricultural production (Hillel, 2000). Globally, salinity is severe abiotic stress that causes huge reduction in the growth and development of plant. It has been estimated that salt stress has affected approximately 6% of land area of the world that constitutes above 800 million hectares of dry land (FAO, 2008). Out of 230 million hectares of irrigated area affected by different abiotic stresses 45 million hectares is affected by salinity worldwide (Ashraf, 2010). According to another estimate, globally, 831×10^6 hectares area has been affected by salts (Beltran and Manzur, 2005). Moreover, it is estimated that salinity has affected one-half of the irrigated land (that is about 2.5×10^8 ha; Rhoades and Loveday, 1990). About 50% of the arable land will be prone to damages by salt stress up to year 2050 (Manchanda and Garg, 2008). Saboor *et al.* (2006) stated that the salinity is degrading about 10% of land area every year in the world.

The problem of salinity is more adverse in dried areas especially the areas receiving little rainfall with high evapo-transpiration rate and temperature (Neto *et al.*, 2006). The problem of the soil salinity is elevating throughout the world due to the use of low quality water resulting in accumulation of some soluble salts in the root zone of the plants. The problem is further increasing by poor soil management practices (Misra *et al.*, 1997; Pitman and Lauchli, 2002).

Scenario of salinity stress in Pakistan

Pakistan is predominately an agricultural country and thus, its development depends upon the improvement of the agriculture sector. Pakistan comprises of 79.61 million hectares of total geographical area (Khan *et al.*, 2004) out of which 80% (62,400 km long) land is irrigated with canal water. Sustainable agriculture of Pakistan is in constant threat by a number of factors like drought, soil salinity, change in climate, low and high water stress and abiotic stresses. Among these problems, sodicity and salinity are the major ones to the soil degradation (Khan *et al.*, 2004).

Salt stress has declined soil potential for crop productivity in Pakistan. Improper cultivation practices and mismanagement of agricultural resources are serious factors for the cause of soil

erosion. Almost 14 hundred thousand hectares of agricultural area in Pakistan has been skipped out of cultivation which is about 25% irrigated land of the country (World Bank, 2006). Subsoil salinity had significantly limited crop growth and yield in Pakistan (Dang *et al.*, 2006). The soil of Pakistan is generally calcareous in nature, alkaline in chemical composition and have low organic matter (Sillanpaa, 1982 and Khattak, 1991). Approximately, 0.2 to 0.4 percent of the total cultivable area is being skipped of cultivation each year because of water logging and salinity (Qureshi, 1978) and only due to salinity problem, 33% (6.8 Mha) area of cultivation has been affected (Anonymous, 2008).

Mechanism of salt stress on plants

Osmotic effect

Plants are stressed under high salt concentrations either by increased osmotic potential or by toxic effects of high ionic concentrations (Brady and Weil, 2002). In osmotic or H₂O deficit environments, soluble salts reduce the water potential and make water not freely available to plants for uptake which is the major reason for stunted growth under salinity. It is very difficult to distinguish between either water deficiency is due to salinity or drought (Nawaz *et al.*, 2010). The water potential of soil controls new leaf formation. Rapidly growing cells have the capacity to store higher levels of salts in their expanding vacuoles, so the growth of the new leaves is not restricted due to gathering of salts in the cytoplasm (Munns, 2005a). Root and shoot growth is more disturbed because of water stress than salt specific effect during the early days of stress (Munns, 2002). At moderate osmotic stress, root growth is not much affected whereas the reduction of shoot growth is maximum (Hsiao and Xu, 2000). Damage due to osmotic effect is governed by plant species, time period of stress, types of cells and tissues and the method of stress application (Munns *et al.*, 2000).

Specific ion effect

Ion specific toxicity, generally, is because of certain ions like sodium, chloride and sulphate which are taken up in larger quantities than routine. It affects the crop right from emergence to physiological maturity. Crops fail especially when specific ions affect at lateral growth stages. Regarding tolerance against salt stress different crops have different levels of responses. Most of the higher plants especially agricultural crops are highly susceptible to this stress (Abrol *et al.*, 1988). Under saline or sodic environments, high concentrations of sodium and chloride ions coupled with low concentration of potassium ions was observed in leaves of wheat varieties (Maas *et al.*, 1986). Mostly the salts are accumulated in the older leaves of plants. With higher concentration of salt

accumulation there may be death of leaves; this happens when the salt concentration is too high, hence cannot be retained inside the vacuoles. In such cases the excessive salts go to the cytoplasm where they affect the normal mechanisms of enzyme action. On the other hand, excessive salts cause cell dehydration by being accumulated in the cell walls (Munns, 2005b). In defense against this effect, plants either try to restrict the salt entry in their bodies or reduce the amount of salts in their cytoplasm. Concentration of sodium in the cytoplasm of the root cells is from 10-30 mM (Tester and Davenport, 2003). Due to high concentrations of sodium and chloride ions inside leaf sap, root and shoot fresh weight reduces up to 50% (Parveen and Qureshi, 1992).

Nutritional imbalance

Ions discrepancy is caused by higher accretion of sodium and chloride and consequently less absorption of the other minerals such as calcium, manganese and potassium (Karimi *et al.*, 2005). Elevated Na⁺: K⁺ ratio causes enzyme inactivation and affects normal metabolic functions of the plants (Booth and Beardall, 1991). Building up of salt deposition disturbs water relations of the plants; this results in limited uptake and utilization of important nutrients. As a result, metabolic activities of the cell and functioning of the enzymes is disturbed (Lacerda *et al.*, 2003).

Nutrients and salt interaction cause deficiencies and imbalances of the major nutrients (McCue and Hanson, 1990). More uptake of Na⁺ causes reduction in the uptake of potassium and symptoms like potassium deficiency are observed (Gopal and Dube, 2003). The regulation of calcium within the plant under saline condition is a crucial parameter of plant salt tolerance (Soussi *et al.*, 2001).

Potassium is main component for protein formation, osmoregulation, photosynthesis and maintenance of cell turgor pressure (Ashraf, 2004). Decrease in potassium ion uptake in due to salinity stress was observed (Marcar *et al.*, 1991). K⁺ along with Ca⁺² are necessary for maintaining the integrity and proper working of the cell membranes (Wenxue *et al.*, 2003). Sufficient amount of K⁺ in plant cell under salinity depends on the uptake on selection basis of the potassium ions and discriminatory compartmentalization of K⁺ and Na⁺ ions in the shoots (Munns *et al.*, 2000).

Oxidative stress

A major effect of salinity is elevation in production of ROS (reactive oxygen species) e.g. H₂O₂, O²⁻, and OH⁻ (Mittler, 2002; Munns, 2002). Proteins, lipids and nucleic acids are damaged oxidatively by ROS and hence negatively affect the normal cellular metabolism (Imlay, 2003). Reduction of oxygen causes formation of these ROS that disturb plant metabolic routes (Asada, 1999). ROS production occurs at minute level

during the normal body and cell growth (Polle, 2001) but increased production occurs during stressed conditions (Laloi *et al.*, 2004). Osmotic effect inhibits the stomata opening and decreases the CO₂ supply for photosynthesis which stimulates the deposition of super oxides in chloroplast. This deposition of super oxides promotes the photoinhibition and photooxidation in plant cells (Ashraf, 2009). Plants have unique appliances to salvage these ROS such as stimulation of the enzymes of antioxidative pathway (Smirnov, 2005).

Effects of salinity on plant growth

Germination stage

Salinity severely affects both glycophytes and halophytes especially at germination stage (Sosa *et al.*, 2005). Seed germination is very important stage for the successful establishment of healthy seedlings which are very sensitive to salinity as compared to other vegetative stages. Salinity accumulates the toxic ion in plants causing a mineral imbalance. The essential ions are reduced and do not meet the demand resulting in hindrance in normal physiological activities of plant. High salt stress retards seed germination process while low salt stress causes seed dormancy (Khan and Weber, 2008). To cope with such nutritional limitation, seeds develop a mechanism of maintaining low water potential (Allen, 1994), or other specific tolerance mechanism to prevent the damage due to salt stress (Rumbaugh *et al.*, 1993).

Salinity disturbs germination in a number of ways. From reducing the osmotic potential of soil which makes decline in water imbibition by seed (Khan and Weber, 2008) to the creation of ionic toxicity which alters enzymes action involved in nucleic acid metabolism. Other impacts of salt stress on seed germination include change in metabolism of protein (Rasheed, 2009).

Seeds are more susceptible to salt stress due to close association to surface of the soil (Dodd and Donovan, 1999). With sodium chloride accumulation to a toxic level in soil, ionic stress decreases the rate of germination (Murillo-Amador *et al.*, 2002). Water absorption by the seed is reduced because of lower water potential caused by salt stress thus posing toxic effects to the developing embryo, resulting in delay in germination process (Khan and Ungar, 1984).

The average time of seed germination is dependent on salinity stress strength and genotypes. With increasing trend of salinity stress there is always decreasing rate of germination (Ditommaso, 2004). Carpici *et al.* (2009) estimated the reduction in germination index of maize cultivars because of salt stress. Salinity exerts very pronounced effect on the germination index and seed size of chickpea (Kaya *et al.*, 2008). Small sized seeds show high value of germination index as compared to large

size seeds under salinity stress. Salinity delays the time to achieve 50% germination in citrus (Zerki, 1993).

According to Farooq *et al.* (2006) toxicity of salts on rice seedlings and time to 50% germination are reduced if seedlings are treated with ethanol treatment. Salinity has negative effect on the vigor index by raising salt concentration in the growing medium (Djanaguiraman *et al.*, 2003). Bordi (2010) reported reduction in germination percentage, rate of germination and germination speed due to salinity. Salinity caused approximately 32% reduction in germination rate, 80% in root length, 78% in plumule length, 78% in shoot length and 95% in seed vigor of *Zea mays* (Khodarahmpour *et al.*, 2012).

Plant physiology

Increased amounts of salt in the soil poses a serious threat to different processes of plants which results in reduction of crop productivity. Epstein (1980) reported reduction in the uptake of essential ions in the plants due to salinity; this causes alteration in metabolic rates leading to reduction in growth rate. Excessive salt concentration in root zone of plant causes change in plant water relations. To deal with the increased amounts of salinity, the osmotic potential decreases (Rodriguez *et al.*, 1997; Gama *et al.*, 2007; Kaymakanova and Stoeva, 2008). Salinity causes reduction in turgor in plant cells due to reduction in water uptake by the plant. Low water uptake reduces cell division and regulation of stomata aperture which ultimately lead to low photosynthesis and finally death of plant tissues (Marschner, 1995; Munns *et al.*, 2002). Reduction in turgor pressure results in stomata closure which causes reduction in gaseous exchange through transpiration (Munns, 1993; Munns and Tester, 2008). Other physiological activities under the influence of salinity include changes in membrane permeability leading to destabilization of membrane proteins (Gupta *et al.*, 2002; Grattan and Grieve, 1992) and reduction in the process of photosynthesis (Sayed, 2003; Kao *et al.*, 2003; Ashraf and Shahbaz, 2003). Lowering of photosynthesis rate happens due to reduction in enzymes and pigments carrying out photosynthesis (Ashraf and Harris, 2013; Misra *et al.*, 1997; Saravanavel *et al.*, 2011). In a study on mungbean grown under saline conditions, it was found that the additional increase of leaf Na⁺ and Cl⁻ also causes the production ROS followed by reduced photosynthetic capacity leading to low plant growth (Nazar *et al.*, 2011). Many processes which are related with plant physiology and biochemistry are affected by salinity like photosynthesis (Hayat *et al.*, 2010), water conductance through stomata (Perez-Perez *et al.*, 2009), various biomolecules and plant-water relations. All these adversely

affected biological processes ultimately reduces crop yield.

Plant anatomy

Salinity has significant effects on plant anatomy. Plants adopt various strategies in response of salinity, that allow them to deal with the problem. Plants with growth in high salt concentration, have more thickness of leaves (Waisel, 1991), epidermis, cell walls and cuticles. The high salt concentration, increases mesophyll cell layers and cell size (Zekri and Parsons, 1990), due to more extension in cell wall at high turgor pressure (Munns and Termaat, 1986). Plants grown in salt stress conditions have large in number but narrow xylem vessels as compared to plant grown in salt free media (Walker *et al.*, 1985). Salinity increases the density of stomata of lower side of leaves and leaf thickness (Raafat *et al.*, 1991) with increased palisade tissues (Hussein *et al.*, 2012); however, it reduces number of cells per leaf. Salinity reduces the number of stomata on the surface of epidermis (Cavisoglu *et al.*, 2007), the total leaf area, (Awang *et al.*, 1993), leaf plastochron index (Bray and Reid, 2002). Vascular bundle length, xylem rows, number of vessels have also been reported to decline due to salinity (Hussein *et al.*, 2012). Salinity stress causes increase in suberin amounts inside the roots (Walker *et al.*, 1985).

Salinity lowered the xylem development and width of vascular bundle in mungbean (Beida and Ho, 1993; Rashid *et al.*, 2004). In rice, stem diameter was reported to be reduced (Pimpongkol *et al.*, 2002), while trichome and stomata density increased. Junghans *et al.* (2006) reported that cambial activity is also disturbed by raising the salt concentration in *Populus euphratica*. A reduction in thickness and area of mesophyll tissue around the axis of leaves of kallar grass was found by increasing salinity (Ola *et al.*, 2012). Salt stress reduced cell size, epidermal thickness of leaves, apical meristem, diameter of cortex and central cylinder (Reinhardt and Rost, 1995; Javed *et al.*, 2001). Salinity caused thickening of endo- as well as exodermis (Gomes *et al.*, 2011; Degenhardt and Gimmler, 2000) and increased development of sclerenchymatous tissues (Javed *et al.*, 2001). In other studies, lignification of intercellular spaces in exodermis in *Vracbiaria decumbens* was also observed (Degenhardt and Gimmler, 2000; Gomes *et al.*, 2011).

Plant morphology

Once the seed has germinated, next goal for plant growth is crop establishment. Salinity cause reduction in crop establishment by reducing shoot growth, blocking leaf development and expansion, reducing growth of internodes and promoting abscission of leaf (Ziska *et al.*, 1990; Zekri, 1991). While studying morphological attributes of *Suaeda salsa*, Guan *et al.* (2011)

observed that that salt stress decreased shoot length, diameter and number of branches due to increased Na⁺ and Cl⁻ contents. Salinity accelerates a number of factors in plants like osmotic stress, ion toxicity and nutrient imbalance; these are identified as most prominent causes of reduction in crop growth which finally lead to crop failure. However, different stages like germination, vegetative growth, flowering, seed establishment and grain filling of crops behave differently with salinity.

Salinity affects plant morphology in different ways. These modifications depend on cultivars used, intensity and duration of the stress (Khan *et al.*, 2003b; Munns and James, 2003). Ashraf *et al.* (2002) studied the morphogenetic parameters of germplasm of 15 guar ecotypes under varying degrees of salinity. They concluded that salinity poses a significant reduction in various vegetative parameters. In different plants the toxic ions effects were observed like burning of the leaves, chlorosis, reduction in the leaf area and the necrosis. The plants affected by salinity have dark green, thicker and succulent leaves. Salt reduced leaf area and volume (Bray and Reid, 2002). The main harmful effects of salinity are reduced germination and emergence, stand and establishment of seedlings (Wahid *et al.*, 1999), and enhanced chlorosis and senescence of leaves (Lutts *et al.*, 1996; Wahid *et al.*, 1997; Curtis and Lauchli, 1987). To cope with osmotic stress, plants reduce the leaf area and increase the rooting density (Guo *et al.*, 2002; Han and Wang, 2005).

Qureshi *et al.* (2000) and (Sanadgol, 2002) reported that in *Eucalyptus camaldulens* and *Agropyron elongatom* respectively. Salinity reduces leaf bio-mass of stem of maize (Evlagon *et al.*, 1992). Other parameters like area of leaves, root and shoot dry weights were also reduced by increasing salt levels (Ashrafuzzaman *et al.*, 2002). Salinity reduced relative growth and net assimilation rate (Khan, 2001; Ahmad, 2010). Ashraf and Bhatti (2000) stated that in rice, salinity decreased biomass and leaf area. In sugar beet, growth and leaf area expansion were reduced even at very low NaCl concentration (Terry and Waldren, 1984).

Reproductive growth stage

Salinity causes about 50% downfall in crop growth, productivity and yield throughout the globe. Isla *et al.* (1998) observed that high level of salinity caused reduction in the growth and yield of barley by 65% and increased ash content. Ahmad *et al.* (1995) indicated that with increased salinity levels, dry matter and seed cotton yield was significantly reduced. Khan *et al.* (1999) evaluated different wheat cultivars against salinity stress for their intra-varietal variability. They concluded that the grain yield decreased 69% and straw yield

decreased by 64% with increase in the root zone salinity.

Reddy and Vora (1986) proposed that salt stress decreases the yield components as a result of change in the normal plant metabolism. As already discussed, salinity affects various physiological growth parameters. The considerable effects are observed on the grain yield per panicle, seed growth. Nahar and Hasanuzzaman (2009) came with a result that that salt stress decreased different components of yield in *V. radiate* and rice (Gain *et al.*, 2004). Kafi and Goldam (2000) determined the response of plants against salinity stress. They concluded that salinity poses a serious problem in vegetative and reproductive stage in the plants.

Mechanism of salt tolerance in plants

Osmotic adjustment

Osmotic adjustment or osmoregulation is generally considered a significant adaptation to salt and water stress. The plant with a better osmotic adjustment mechanism maintains growth and yield and survives better in dry or saline soils because it improves cell turgor as well as cell volume (Munns, 1988). Osmoregulation is considered a key mechanism to ameliorate toxic effects of salinity as it causes reduction in osmotic potential because of gathering of some compatible solutes (Hasegawa *et al.*, 2000). Ions outside the root cause osmotic stress (Flowers and Colmer, 2008).

In the process of osmosis, a solvent moves from low to high solute concentration. Roots have high concentration of solutes in the form of sugars and various organic compounds as compared to the soil solution outside the roots. Water moves through selectively permeable membrane from cell walls to the root cells. However, this balance in solute concentration is disturbed when solute concentration in soil rises, thereby reduces water uptake by plants. To compensate the high concentration of solutes outside the root media, plants accumulate different osmotica in the form of sugars and organic compounds. Among the amino acids, proline is necessary for osmotic adjustment (Zhao and Harris, 1992) by lowering cell water potential. This needs energy which is provided at the cost of cut of energy supply to other metabolic activities. So due to diversion of energy and resources, overall growth of plants is reduced (Hanson *et al.*, 1999). The osmotic problem stimulates the decrease in efficiency of utility and uptake of other essential nutrients which results in an imbalance of nutrients in the plant. This is mainly because of competition of sodium with other essential elements for availability and uptake in soil solutions and in plant roots. The enzymatic activity and metabolic processes are also susceptible to the high salt concentration (Lacerda *et al.*, 2003).

Synthesis of compatible solutes

One method to avoid toxic effects of salinity is the presence of metabolites which serve the purpose of compatible solutes. Plants synthesize low molecular weight solutes to acclimate the toxic effects of salinity; these can be termed as compatible solutes (Hasegawa *et al.*, 2000). Organic osmolytes do not disturb the normal metabolic processes of plants, even at high concentration (Rhodes and Hanson, 1993); their high concentration stabilizes membrane integrity, other macromolecules assemblies and control enzyme functions (Genard *et al.*, 1991; Schwab and Gaff, 1990). These solutes play a major part in order to maintain the pH of cells, scavenging of free radicals produced during oxidation process, detoxification of harmful chemicals and serve as stored form of nitrogen (Mansour *et al.*, 2000); these molecules guard the cell from the toxic effects of salt stress and drought (Genard *et al.*, 1991; Krishnamurthy and Bhagwat, 1991).

Other metabolites that serve as compatible solute include polyols, betains (Sakamoto and Murata, 2000) and soluble sugars (Wahid, 2004) etc. Among these, osmotic adjustment is established by amino acid (Bohnert *et al.*, 1992). Now it is well established that polyamines, that maintains the solute potential between cells relative to its surroundings (Pollard and Wyn Jones, 1979).

Ionic compartmentalization

Among different ways to tolerate high salinity, one is to compartmentalize inside the cells (Carlos *et al.*, 2009). The salt stress results in an imbalance of the nutrients required for plant growth. Ionic stress causes increase of Na⁺, Cl⁻ and decrease of K⁺ ions. The imbalance of these ions is harmful for growth of plants (Abdel Kader *et al.*, 2011). Increased Na⁺ under salinity decreases Ca⁺², K⁺, Mg⁺² within the cytosol and disturbs the ionic homeostasis (Aleman *et al.*, 2011). Na⁺ is very toxic at high concentration that causes membrane destabilization, loss of functional proteins, slows down cell division and expansion, lower plant metabolic rates and alters the homeostasis of the mineral nutrients (Munns and Tester, 2008). Na⁺ also competes with potassium for cellular uptake when its external concentration is very high (Niu *et al.*, 1995). With increase in Na⁺ more than 100 mM in the cytosol, it becomes toxic (Serrano *et al.*, 1999).

Na⁺ in high concentration causes reduction in K⁺ concentration accompanied with imbalance of K⁺/Na⁺ ratio and other electrolytes vital for optimum homeostasis of cytosol (Shabala and Cuin, 2008). Plants maintain ionic homeostasis by regular control of Na⁺ entry in the cell and its compartmentalization into the vacuole (Zhu, 2003). This strategy is very effective in both halophytes and glycophytes since it helps in lowering the Na⁺ and Cl⁻ concentration within the cell and maintains

growth under high salt concentration (Hasegawa *et al.*, 2000; Blumwald *et al.*, 2000).

Synthesis of antioxidant

The disturbance of cellular homeostasis in response to different stressed conditions such as drought, salinity and cold produce reactive oxygen species (ROS) (Dubey, 2011). These are created as an additional or due to escape of electrons from electron transport processes carried out in chloroplast and mitochondria (Rishi and Sneha, 2013; Joseph and Jini, 2010). The cellular metabolism causes the production of free radical.

Breeding strategies to enhance salt tolerance in plants

Screening and selection of existing germplasm

Scientists have developed different methods to cope the salinity problem. One way is to exploit variations in genomes of the available germplasm for identification of a salt tolerant genotype that can enhance crop yield (Ashraf *et al.*, 2006). Since salinity reduces the crop growth and productivity, a proper selection criterion is needed to determine the relative salt tolerance (Francois and Mass, 1994). Different levels of tolerance against salinity exist within plants (Francois and Mass, 1994). Many scientists have developed screening criteria for different crops like wheat (Ali *et al.*, 2002), maize (Khan *et al.*, 2003a) and rice (Shannon, 1998).

Some plants show significant tolerance to salinity in a remarkable way that indicates the genetic potential in plants to tolerate the toxic effects of salinity that is required trait in agriculture (Mahmood *et al.*, 2000). Plants accumulate salts according to their capacity to salt tolerance. Plants with capability to exclude more Na⁺ ions out of the cell are considered more salt tolerant as compared to plants that accumulate Na⁺ ions (Schachtman and Munns, 1992). So the salt tolerant species of plants accumulate higher quantities of K⁺ and low quantities of Na⁺ ions as compared to salt sensitive species (Tipirdamaz and Cakirlar, 1989). However, it was proposed that there does not seem any strong relationship in removal of Na⁺ with tolerance to salinity in wheat; hence both are independent features of crops. Moreno *et al.* (2000) determined variation in genotypes in cultivars of *Phaseolus vulgaris* L. They reported that some bean cultivars had higher root growth and mineral nutrient accumulation as compared to others. Giaveno *et al.* (2007) performed a screening experiment of tropical maize for salt tolerance. The experiment was conducted at germination and seedling stages. They concluded that significant genetic variability existed at germination stage while the early seedling growth had no association with the germination under salinity stress. Akram *et al.* (2010) performed an experiment for screening at early stages of maize. The genetic variations for

salinity tolerance were determined in maize by hydroponic technique. They used ten hybrids of maize against four levels of salinity i.e. 0, 40, 80 and 120 mM. It was observed that salinity had significant effect in reducing biomass and production of maize.

Genetic variability for salt tolerance

To breed crop plants tolerant to salinity, there should be significant variation available in salinity tolerance. However, Noble and Rogers (1992) have pointed out that there was very little variation between varieties for salt tolerance. Plant-plant variations for tolerance against salinity among different varieties was reported by different researchers mostly in cross-pollinated species, e.g. in alfalfa (Al-Khatib *et al.*, 1993) and maize (Khan *et al.*, 2003a). In recent years, Ashraf (2002) has screened out plant material from F₃ population, developed by cross between LU26S (salt tolerant wheat cultivar) with cv. Kharchia from India, which grew successfully under S_{24dsm}⁻¹ and S_{36 dsm}⁻¹. The success of development of salinity tolerant germplasm depends upon the selection criterion to be followed while screening the segregating generations under saline conditions, and is also subject to heritability of the character.

The potential to select and breed for plants with increased salt tolerance may be good if the variation shown for that specific trait is genetically determined. Information on the genetic basis of tolerance would assist the breeder in two ways; firstly, it helps him to devise appropriate selection protocols for screening tolerant plant, and testing their progenies, and secondly, it provides estimates of heritability of the character. In sorghum, genetic variation demonstrated that salinity tolerance is controlled by both additive and dominant gene actions (Ashraf *et al.*, 1987). Adetimirin *et al.*, 2001 reported high interaction of epistatic effects with environment rather than other gene actions. Revilla *et al.*, 2000 revealed the role of additive and dominance × dominance gene action to govern most of the traits in control condition studies. In addition to these studies, the reports available on rice (Shannon *et al.*, 1998), maize (Rao and McNeilly, 1999), Triticeae (Xing *et al.*, 2002), cotton (Akhtar and Azhar, 2001; Azhar and Ahmad, 2000; Noor *et al.*, 2001), *Aegilops ovala* (Farooq, 2002), and lucerne (Al-Khatib *et al.*, 1994) revealed that salt tolerance was heritable in nature and we could greatly improve these species through direct selection or breeding techniques.

Summary

Salinity, as we know that it is one of the most damaging abiotic stresses, which causes serious decrease in growth, development and production of almost all biota. If soil has water the electric conductivity of which is greater than 4 dsm⁻¹, it can be declared as saline. Salinity can be

divided into primary (caused by decaying of rocks), and secondary (caused by anthropogenic activities). The problem of salinity is more severe in dry areas having low rainfall and high evapo-transpiration rates because the water in the soil is lost to atmosphere and salts cannot leach down causing increase in salinity. About 800 Mha of dry land has already fallen to salt stress. The rate of the soil becoming saline is so fast that it is assumed that by 2050, about half of the fertile soil of the globe would become saline. In Pakistan, approximately 0.2 to 0.4 percent of cultivable area is being put out of cultivation each year and almost 25% of the irrigated land has already been skipped out because of water logging and salinity by the year 2006.

Increased solute potential in soil coupled with toxic concentrations of the salt ions affect growth, development and yield of plants. Highly hypertonic conditions in the soil make water less likely to be available to plants for uptake causing stunted growth and even wilting. So it is very difficult to determine whether water deficiency in plant is due to drought or salinity. As plant cells have ability to store excessive amounts of salts in their vacuoles, so water stress seems to affect more than salt stress.

Regarding salinity, Na⁺, Cl⁻ and SO₄²⁻ most commonly affect plants right from germination till physiological maturity of the crop. The response against salinity varies from plant to plant, but on the average, when the salt concentration increases inside plant, it tends to shift it into older leaves to be stored in the vacuoles of the cells. That's why high levels of sodium and chloride ions and reduced amount of essential elements like calcium, potassium and manganese can be observed in salinity stressed plants. When the amount of salinity exceeds plants limit, the salts go into cytoplasm of the cells causing full fledge disturbance in the enzymatic activity and eventually death of cells leading to senescence. Another result of salt stress is the over production of ROS which cause photoinhibition and photooxidation.

The amount, duration and intensity of salts in the vicinity coupled with the growth stage of plant collectively determine plant's fate. Generally, every stage has its own importance regarding growth and production, but salinity at germination is quite dangerous for plant stature while that at reproductive stage is importance in lowering the production. Plants may adjust their osmotic potential to minimize salt uptake but this creates moisture stress. Another important strategy is the production of compatible solutes that play critical role in maintaining cell pH and lowering toxicity of salts. Cell compartmentalization, and production of antioxidants are also quite acceptable strategies to save as much as possible. Hence, best strategy against salinity stress so far is to develop a

screening criteria for salinity tolerance, then evaluation of diverse germplasm on the basis of that criteria, and conventional as well as molecular breeding approaches to pyramid essential traits into one ideal genotype.

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