

## **Problem soils in cool-season food legumes- Current knowledge and future prospects**

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## **Abstract**

Problem soils impose various stresses on plants and reduce crop yield significantly. As a sizeable area under food legume crops falls under the problem soils, development of tolerant varieties and appropriate agronomic practices to ameliorate problem soils are required to increase food legumes production. The current research efforts towards the development of cultivars tolerant to problem soils and soil amelioration measures are very limited. There is a lack of knowledge on physiological and genetic basis of mechanisms contributing towards tolerance to problem soils. . Recent advances in high throughput phenotyping and genotyping (next generation sequencing) platform offer new suites of technologies that facilitate the understanding of tolerance mechanisms, accelerate the genetic gains and develop improved cultivars better adapted to adverse soil conditions in a rapid manner. This review examines various edaphic problems, discuss recent efforts towards the development of appropriate technologies including tolerant varieties, soil amelioration methods and innovation in agronomic practices that are contributing to, or potentially can contribute to adaptation of food legume crops such as lentil, chickpea, faba bean, and field peas in problem soils.

Keywords: Soil acidity, salinity, phenotyping, agronomic manipulations, breeding.

## **1. INTRODUCTION**

Soil is the key environmental factor that affects plant growth and development. The properties of the soil are usually embedded in various definitions of soil fertility. Reuler and Prins (1993) defined soil fertility as the capacity of soil to provide plants with nutrients, water and oxygen. A mineral is considered as nutrient, when without it plant cannot complete its life cycle. Sixteen minerals are essential nutrients for plant growth and development. These are carbon (C), hydrogen (H), oxygen (O), nitrogen (N), phosphorus (P), potassium (K), calcium (Ca), magnesium (Mg), sulphur (S), iron (Fe), manganese (Mn), zinc (Zn), copper (Cu), boron (B), molybdenum (Mo) and chlorine (Cl). For normal growth and function, plants require all these nutrients in a balanced proportion. Except C, plant uptakes all minerals from the soil solution whereas C enters from the atmosphere, in the form of carbon dioxide (CO<sub>2</sub>) through carbon assimilation process. Soil health is important for crop production however, many of today's soils are afflicted with various problematic conditions.

There is no concrete parameters to define the concept of problem soils. Osman (2013) defined problem soils as the one that limit crop cultivation and requires special management practices to support a successful crop. In general, the problem soils are not suitable for crop production (Huq and Shoab 2013). Physical conditions such as dryness, wetness, steepness and poor

texture of soils, chemical conditions such as acidity, salinity, alkalinity and sodicity, nutrient imbalances such as boron and aluminium toxicities, and essential nutrient deficiencies fall under the problem soils (Osman 2013). These soils affect plant growth and productivity not only due to the presence of high toxic minerals but also due to the limited supply of essential nutrients (White and Greenwood 2013). Limited availability of macronutrients, nitrogen (N), Phosphorus (P) and Potassium (K), and micro-nutrients, Magnesium (Mg), Iron (Fe), Zinc (B) and Boron (B) challenges crop cultivation (Lynch 2007; Fageria et al. 2011; Mueller et al. 2012). Problem soils also exert water related stresses including restricted root growth through mechanical impedance in drying soils, limited porosity and waterlogging (Bengough et al. 2006; Hallett and Bengough 2013). It also influence the C/N ratio and hence the mineral nutritional status of the soil.

Chickpea (*Cicer arietinum* L), peas (*Pisum sativum* L), lentil (*Lens culinaris* Medik.), faba bean (*Vicia faba* L), and grass pea (*Lathyrus sativus*) are the major cool-season food legume crops (Andrews and Hodge 2010) and their cultivation improves physical, chemical and biological properties of the soil. Low carbon and water footprints, better C/N ratio and improved nitrogen economy are some of the advantages that cultivation of legumes offer to the soil (Andrews et al. 2009; Andrews and Hodge 2010). The inclusion of legumes typically benefits succeeding crops by improving soil health because of biological nitrogen fixation and other rotational effects. Legumes help to enhance net carbon sequestration and lower the carbon footprint of cereal production. One study showed that the lentil-wheat system among tested rotations had the lowest per area carbon footprint at -552 kg CO<sub>2</sub> eq/ha (Gan et al. 2014). Legume crops are well adapted to a wide range of soil conditions differing in texture and fertility. However, their yield constrains substantially when grown under difficult edaphic conditions (Mueller et al. 2012; Osman 2013). Therefore, to raise a successful legume crop in problem soils, both genetic and agronomic interventions are required as the potential yield of an improved cultivar can be realized only with the best agronomic practices. Legume breeding target to develop cultivars that can overcome mineral toxicities by improving tolerance and mineral deficiencies by improving nutrient use efficiency. However, breeding for tolerance to problem soils is a difficult task and requires specific breeding strategy. It involves rigorous phenotyping at hotspot sites over the years due to heterogeneity of the stress, dissection of tolerance into its components, pyramiding genes/QTLs associated with each components of tolerance in the desired agronomic background, multi-environment evaluation of tolerant lines for their performance and adaptability under problems soils. Some progress was made by early breeders through specialized field designs to manage field trials and use of sophisticated statistical tools to assess genotype x environments x management interactions (GEMI). However, tolerant cultivars for commercial use are limited due to physiological and genetic complexity of traits contributing to tolerance mechanism. Recent advances in imaging technologies now open up new avenues for high throughput phenotyping of plants grown under problem soil conditions. RGB image technology is useful to study soil salinity (Rajendran et al. 2009; Schilling et al. 2014) and boron deficiency (Schnurbusch et al. 2010; Hayes et al. 2013) while thermal images are useful to screen osmotic stress by diagnosing leaf water status and leaf temperature (Sirault et al. 2009; Berger et al. 2010) under water stressed conditions,

chlorophyll fluorescence images to detect N deficiency (Luand 2000; Antal et al. 2010; Donnini et al. 2013), and hyper-spectral images to screen genotypic differences for micro-nutrient deficiencies (Li et al. 2005; Zhao et al. 2005; Shi et al. 2011a; 2011b; Shi et al. 2012a; 2012b).

Similarly, recent advances in molecular tools have brightened up the scope to integrate them in mainstream breeding programs. However, in comparison to cereals, there are limited efforts in the development and deployment of molecular tools in legume crops. The whole genome sequencing projects have recently concluded for some legume crops including *Medicago*, *Lotus*, soybean (Schmutz et al. 2010), chickpea (Varshney et al., 2014), pigeonpea (Varshney et al. 2011) and lentil (Bett et al. 2016). This provides an opportunity to identify putative orthologous gene sequence resources through comparative genetics, especially those located within the Galegoid clade of the Fabaceae sub-family Papilionoideae.

The best agronomic practices would help to bring changes in the soil condition and bring some practical solution to the problem soil conditions.

In this review, we discuss production constraints of legume cultivation under various problem soils and suggest integrated approaches combining agronomy, breeding approaches and genomics, to bring problem soils under legumes cultivation.

## **2. PROBLEM SOILS**

### **2.1 Soil acidity**

The soil with pH <7 is considered as acidic soil (Slattery et al. 1999). The natural process of soil acidification involves the accumulation of hydrogen ions (H<sup>+</sup>) released from nitric acid and sulfuric acid from the acid rain into the soil over the years (Hajkowicz and Young 2005). In addition, the N compounds in the fertilizer get oxidized during the nitrification process that produces nitrous and nitric acids and favours the development of acid soils over the time (Hajkowicz and Young 2005). Soil acidity is one of the emerging agricultural problems and globally, ~3950 million ha of arable land is affected by it. It includes 38% farmland in Southeast Asia, 31% in Latin America, 20% in East Asia, 56% in Sub-Saharan Africa, and parts of North America (Wood et al. 2000; Hoekenga et al. 2006). America has about 1616 million ha of land affected, most in South America. In Australia and New Zealand, 239 million ha of agricultural land is acidic [11]. In China and India, about 12% (212 million ha) of agricultural land is classified as acidic (Bian et al. 2013). In acid soils, at pH <5.5, iron (Fe), copper (Cu), manganese (Mn), Zinc (Zn), aluminium (Al) and manganese (Mn) become more soluble; they become toxic as their concentration in the soil solution rises (Ring et al. 1993) and cause P, Mo, or Ca deficiency (Graham 1992). Aluminium (Al), the third most common element in the Earth crust is one of the most serious problems limiting crop productivity in acid soils. Low pH in topsoils may affect microbial activity, most notably decreasing the Rhizobia activity and consequent poor nodulation in legume crops. The resulting nitrogen deficiency is apparent from reddening of the stems and petioles on pasture legumes, and yellowing and

ultimate death of old leaves on grain legumes. The population of Rhizobium bacteria is reduced substantially in acid soils. Some legumes may fail to persist due to the inability of reduced Rhizobia populations to successfully nodulate roots and form a functioning symbiosis. At high pH (> 6.0), Al mostly exists in a non-soluble form. However, when soil pH decreases, Al become more soluble and the excessive Al ions ( $\text{Al}^{3+}$ ) uptake by higher plants cause  $\text{Al}^{3+}$  toxicity.  $\text{Al}^{3+}$  toxicity inhibits root growth, reduction in root penetration and branching, reduces microbial activity and causes poor nodulation in legumes (Bian et al. 2013; Scheffer-Basso and Prior 2015). The major lentil-growing countries including India, Canada, Turkey, Syria, Australia, Nepal and the United States have large acreage under acidic soils with the problem of  $\text{Al}^{3+}$  toxicity (Bian et al. 2013). Reduction in root growth is the key morphological symptom of  $\text{Al}^{3+}$  toxicity. In lentil, chickpea and faba bean reduction in root growth is evident due to  $\text{Al}^{3+}$  toxicity (Singh et al. 2012; Qi-chen et al. 2012; Choudhary and Sharma, 2014). In lentil, number of pods/plant is reduced under  $\text{Al}^{3+}$  stress conditions. The affected lentil plants show purple colour stem and leaves which resembles phosphorus deficiency under  $\text{Al}^{3+}$  toxicity environments (Singh et al. 2012). The total protein content was decreased in faba bean due to  $\text{Al}^{3+}$  toxicity (Xu et al. 2013). High concentration of  $\text{H}^+$  ions in acid soil competes with other mineral elements such as phosphorus (P), magnesium (Mg), calcium (Ca), Iron (Fe) and molybdenum (Mo), causing secondary stresses that affect plant growth and development.

## **2.2 Soil salinity**

Salinity indicates the presence of extra salts in soil. In other words, salinity measures the total amount of soluble salt of chlorides ( $\text{Cl}^-$ ) and sulphates ( $\text{SO}_4^{2-}$ ) of sodium ( $\text{Na}^+$ ), calcium ( $\text{Ca}^{2+}$ ) and magnesium ( $\text{Mg}^{2+}$ ) and to a lesser extent, salts of carbonates ( $\text{CO}_3^{2-}$ ) and bicarbonates ( $\text{HCO}_3^-$ ) on the soil surface. Weathering of parental rocks and deposition of oceanic salts (by wind and water) cause the development of saline soils in a natural way. Besides, use of saline water, shallow water tables, agronomic practices such as imbalance fertilization, an intense evapotranspiration with insufficient leaching, lack of suitable lands and lack of appropriate irrigation and drainage management are the main reasons for soil salinization (Rhoades et al. 1993; Slavich et al. 1999; Villa-Castorena et al. 2003; FAO, 2005). Saline soils are characterized by the concentration of soluble salts in the soil solution (Munns and Tester 2008). It possesses the electrical conductivity of saturation extract ( $\text{EC}_e$ ) of  $>4 \text{ dSm}^{-1}$ , (approximately 40 mM), the exchangeable sodium percentage (ESP) of  $>15\%$  and the soil pH  $<8.5$  (Abrol et al. 1988). It is a major problem in arid and semi-arid regions of the world where these factors are directly associated with levels of precipitation and temperature. Out of 1,500 million ha of dry land agriculture, 32 million ha are salt-affected (2.1%), to varying degrees by human-induced processes (Oldeman et al. 1991). As salinity levels increase, plants extract less water from soil, aggravating water stress conditions and ultimately, their growth is restricted by the stress of “pulling” water away from the salt. A high salt level interferes with the germination of new seeds. The immediate effects of salinity on plants that are not adapted to salt start from poor seed germination, later followed by leaf drop, leaf burn, stunted growth, and death. Salinity prevents plant roots from performing their osmotic activity where water and nutrients move from an area of low concentration into an area of high concentration. Excessive ions like Na, Cl, B, Li, Se, Cd, Cr and Pb may have toxic effects on legumes when their

concentration within the plants exceeds a specific level. Further, the  $\text{Na}^+$  in the soil particle often replaces  $\text{Ca}^{2+}$  and develops  $\text{Na}^+$  induced  $\text{Ca}^{2+}$  deficiencies (Ehret et al. 1990; Adcock et al. 2001). The accumulation of  $\text{CO}_3^{2-}$  and  $\text{HCO}_3^-$  in the cytoplasm competes with the uptake of other nutrients such as K, Fe, Mn, Mg, Cu, Zn and P and causes other related nutrient deficiencies (Naidu and Rengasamy, 1993). Overall, legumes are sensitive to soil salinity; they are devoid of any specially developed mechanisms to face the saline environment. The growth is retarded at lower concentrations of such ions while mortality of legumes may occur when the concentration of toxic ions are very high and beyond the tolerable levels. Experimental evidence from previous studies indicated that increased treatment of NaCl induced significant increase in  $\text{Na}^+$  and decrease in  $\text{K}^+$ ,  $\text{Mg}^{2+}$  and  $\text{Ca}^{2+}$  levels in shoot system of faba bean. Increasing salt concentrations may have a detrimental effect on soil microbial populations as well, either due to direct toxicity or through osmotic stress. Therefore, nodulation and biological nitrogen fixation (BNF) in legumes is suppressed significantly or even lost totally. Salinity inhibits nitrogen fixation by reducing nodulation, mineral nitrogen level, protein content, phosphates (acid and alkaline) and nitrogenase enzyme activities in faba bean plants (Soussi 1999; Khan 2001; Ferreira et al. 2001; Rao and Tak 2002; Yano-Melo et al. 2003; Parida et al. 2004; Zandavalli et al. 2004; Parida and Das 2005, Rabie et al. 2005; Rahman 2008).

Salinity changes photosynthetic parameters, including osmotic and leaf water potential, transpiration rate, leaf temperature, and relative leaf water content (RWC). Salt also affects photosynthetic components such as enzymes, chlorophylls, and carotenoids. Changes in these parameters depend on the severity and duration of stress (Misra et al. 1997) and on plant species (Dubey 1994). Chlorosis is a common response of salinity stress (Parida and Das 2005; Jamil et al. 2007) that can also limit the photosynthetic efficiency. Salinity can affect chlorophyll content through inhibition of chlorophyll synthesis or an acceleration of its degradation (Reddy and Vora 1986). Reduction in chlorophyll concentrations is probably due to the inhibitory effect of the accumulated ions of various salts on the biosynthesis of the different chlorophyll fractions (Ali et al. 2004).

### **2.3 Soil sodicity/alkalinity**

Sodic soil commonly refers to soils, which have salts of the  $\text{Cl}^-$ ,  $\text{SO}_4^{2-}$ ,  $\text{HCO}_3^-$  and  $\text{CO}_3^{2-}$  of mainly  $\text{Na}^+$  (Abrol et al. 1988). It is also known as alkaline soils. Sodic soil usually possesses poor soil structure with low infiltration capacity. The low level of adsorbed  $\text{Na}^+$  (6%) in the exchange sites can cause severe soil structural degradation (Northcote and Srene 1972). Sodic soils often look dry, hard and black in colour. Usually, they possess a hard calcareous pan at 0.5 to 1 m below the soil surface. Sodic soils are characterized by  $\text{pH} > 8.5$  with the ESP  $> 15\%$  and  $\text{EC}_e$  of  $< 4 \text{ dS m}^{-1}$  (Abrol et al. 1988). Since the dominance of NaCl in saline soils favours adsorption of  $\text{Na}^+$  by soil particles, it induces to be more sodic during soil leaching processes; most of times saline and sodic soils occur together (Rengasamy et al. 2003). The high soil strength of sodic soil creates huge mechanical impedance for roots to grow and penetrate inside the soil column (Masle and Passioura 1987). The poor soil structure of sodic soil sometimes does not allow seeds to germinate at dry condition (Abrol et al. 1988). Sodic soils reduce the

porosity and permeability of soil and results slow water penetration and distribution in the soil column (Oster and Jayawardane 1998). The poor infiltration capacity of the sodic soil allows water stagnation and limits the choice of crop for cultivation. Further, the exchangeable  $\text{Na}^+$  displaces exchangeable  $\text{Ca}^{2+}$  in the soil particle, sodic soils develops  $\text{Na}^+$  induced  $\text{Ca}^{2+}$  deficiencies (Adcock et al. 2001; Ehret et al. 1990). They have high risk of getting  $\text{CO}_3^{2-}$  and  $\text{HCO}_3^-$  toxicities and deficiencies of other nutrients such as K, Fe, Mn, Mg, Cu, Zn and P in plants (1993). Overall, plants growing in sodic soils do not get sufficient quantity of water, oxygen and nutrients, which is essential to obtain high yield and productivity.

## **2.4 Nutrient imbalances**

Although most of the problem soils are deficient in certain nutrients, they are grouped into two major categories, according to whether abundance or deficiency of a mineral predominates. The problem soils including acidic, saline and sodic impose mineral toxicities and induced mineral deficiencies in legumes. On the other hand, deficiency of mineral nutrients such as nitrogen (N), phosphorus (P), sulphur (S), iron (Fe), zinc (Zn) and boron (B) reduce the productivity in the major cool-season food legume producing areas. They are described in the following sections.

### **2.4.1 Nutrient deficiency**

Nutrient depletion limits crop production in all types of soils. Continuous cultivation of crops can cause mineral deficiency, which can be corrected by application of respective fertilizers in the soil and adoption of best agronomic practices (Arnon 1954). This is a serious problem for small landholder farmers in developing countries, where much of the grain legume production occurs, and many farmers cannot afford to buy fertilizers. Sanchez (2002) suggested average annual nutrient depletion rates of N, P and K as  $22 \text{ kg N ha}^{-1}$ ,  $2.5 \text{ kg P ha}^{-1}$ , and  $15 \text{ kg K ha}^{-1}$  across 37 African countries. Out of 135 pulse growing districts in India, soils were low in available P in 68 districts and medium in 62 districts (Ghosh and Hasan 1979). Drought has confounding effect on total soil soluble organic P with reduced uptake of both P and K by plants in the Mediterranean soils (Sardans and Penuelas 2007). There is a negative interaction between yield and N use efficiency when drought and soil acidity exist together. For instance, the wheat belt of Australia, which grows lentils, peas and chickpeas in rotation, can experience greater negative effects on yield and nitrogen use efficiency under climate change scenario (Hernandez et al. 2009).

Under difficult edaphic conditions, the rhizobial strain in the root nodules of legumes did not express in its full capacity to fix atmospheric nitrogen ( $\text{N}_2$ ) (Brockwell et al. 1995; Zahran 1999). Some 40–60 million metric tons (Mt) of  $\text{N}_2$  are fixed by agriculturally important legumes annually, and another 3–5 million Mt fixed by legumes in natural ecosystems (Smil 1999). This helps to balance C/N ratios and prevents the benefits of the N fertilization effects of the atmospheric  $\text{CO}_2$  being realized.

The K and Zn use efficiency is one of the important strategies of lentils and faba bean to be cultivated in the saline soils respectively (Ashraf and Zafar 1997; Saleh and Maftoun et al. 2009).

### **2.4.2 Nutrient toxicity**

A mineral becomes a toxin when plant uptakes higher than the optimum level of requirement. For example, Fe is an essential micronutrient but when present at high concentrations in soil solution and gets accumulated into the plant system, it is one of the major problems of rice cultivation under wetland conditions. A higher concentration of  $\text{Fe}^{2+}$  in the soil solution causes leaf bronzing, necrotic rust leaf spots on the leaf tips, stained leaf edges, stunted shoot growth and poor root development in susceptible rice genotypes (Sahrawat 2004; Elec et al. 2013). Similarly, B toxicity is the one of the major problems of plants under cultivation on the arid and semi-arid areas all over the world. It can occur through natural process from marine evaporates and marine argillaceous sediments (Nable et al. 1997). In addition, irrigation water contaminated with wastes from surface mining, fly ash and industrial chemicals increases the soil B availability and become toxic to plants. An excess of B in the contaminated irrigation water or in the soil solution can cause toxicity to a variety of crops. The symptoms of B toxicity develops by yellowing of leaflets of the lower leaf followed by the necrosis progressed towards the base of leaflets and leaf margins. Under severe stress, it causes dropping of leaflets; it begins from older leaf at the bottom to the younger leaf at the top (Yau and Erskine 2000). As described earlier, Al and Mn become toxic under acid soil conditions (Ring et al. 1993) and sometimes, the presence of toxic minerals in the irrigation water causes this problem. The use of Arsenic (As) contaminated ground water for irrigation leads to arsenization of soils. The Gangetic alluvium of Bangladesh is a good example of arsenization of soils (Huq and Shoaib 2013). The toxicity caused by Al is one of the most significant limiting factors in the growth and development of legume plants in acid soils. Plant roots become thin and dark colored, resulting in lower absorption of water and nutrients and this effect is more pronounced in seedlings than in adult plants (Merino-Gergichevich et al. 2010).

### **2.5 Waterlogging**

Waterlogging is a major problem in certain pocket of pulse growing areas and causes considerable losses to food legumes (Jackson 2010). Waterlogging is the condition in which plants get very limited supply of oxygen with other gases such as carbon dioxide, methane, nitrogen and affect crop production (Huq and Shoaib 2013b). Waterlogging conditions arise due to the poor internal capillary drainage or high soil clay composition in some regions. On the other hand, cropping systems such as rice followed by legumes often develop waterlogging condition in some areas (Cornelious et al. 2005). It reduces germination, seedling emergence, root and shoot growth and plant density up to 80%, besides causing seedling diseases. Yield losses due to waterlogging may reach almost 100% (Siddique 2000). In Australia, transient waterlogging occurs primarily in sandy duplex soils where rainfall rapidly penetrates the sandy topsoil and accumulates above the compacted clay subsoil (Tennant et al. 1992). Poorly drained duplex soils occupy ~3.8 million ha of the agricultural land in Victoria, and in Western Australia, 60% of the cropping land is prone to waterlogging (McFarlane and Cox 1992; Setter and Waters 2003), causing significant losses in crop yields (Siddique et al. 1993). Even though extensive field trials have been conducted on adaptation, growth and yields of cool-season grain legumes in south-western Australia (Siddique and Sykes 1997; Thomson and Siddique

1997; Thomson et al. 1997; Siddique et al. 1998, 1999), research on the effects of waterlogging on growth and yield of grain legumes has been limited. Field trials conducted in Western Australia have demonstrated the potential of faba bean as a crop on duplex soils in medium- and high-rainfall regions (Loss and Siddique 1997; Loss et al. 1997a, 1997b, 1998; Siddique and Loss 1999; Siddique et al. 2001). Field observations showed some degree of waterlogging tolerance in faba bean and its high yield potential particularly in wet years suggests that faba bean is potentially the most productive cool-season grain legume crop for the waterlogging-prone soils of the Mediterranean-type environments of southern Australia (Siddique et al. 1993, 1999, 2001; Siddique 2000). Tolerance to waterlogging as indicated by root and shoot growth (as % of drained controls) was ranked as follows: faba bean > yellow lupin > grass pea > narrow-leaved lupin > chickpea > lentil > field pea (Solaiman et al. 2007). Faba bean produced adventitious roots and parenchyma leading to increased root porosity (9% gas volume per unit root volume).

### **3. MANAGEMENT OF PROBLEM SOILS**

Management of problem soils is not complete unless proper remedial measures are taken to restore the soil fertility and structure of the soil. Integrated management including tolerant varieties, reclamation measures and suitable agronomic practices is the only option for improving the productivity of legumes under the problem soils. Followings are the important management options to overcome these problems.

#### **3.1 Breeding for tolerance to problem soils: conventional approaches**

##### **3.1.1 Genetic variability**

Genetic variation has been reported for tolerance to acidity (Camargo et al. 1995; Foy 1996), salinity (Munns et al. 2000; Munns and James 2003), sodicity (Rao et al. 2008; Singh et al. 2002), toxic soils (Hasnain et al. 2011; Yau and Erskine 2000), and waterlogging (Cornelious et al. 2005; Saqib et al. 2013) in different crop plants. This variation is based on traits such as symptoms of toxicities and deficiencies, physiological parameters such as leaf elongation rate, root and shoot growth, relative growth rate, ion concentrations in various tissues, *etc.*, (Table 1). Among legumes, pea is more tolerant to B toxicity than faba bean, chickpea and lentil. Chickpea germplasm showed significant genotypic variation for salinity tolerance (Flowers et al. 2010). As Cl is more toxic than Na in chickpea yields, more efforts were made to study tolerance to Cl toxicity under saline conditions. Similarly, faba bean is also more sensitive to Cl than Na. Genotypes tolerant to Cl and Na have been identified based on pot and field screening. Screening of pea germplasm against salinity and alkaline/acidity showed that salinity tolerant accessions have mostly originated from Greece and Sha'anxi province in China (Leonforte, Forster, Redden, Nicolas, and Salisbury, 2012; B. Redden, Leonforte, Ford, Croser, and Slattery, 2005). Sha'anxi was one of the first Chinese provinces to develop irrigation systems over 2000 years ago, possibly leading to more areas of soil salinity and subsequent selection pressure for salinity tolerant germplasm, but the same does not hold true for salinity tolerant germplasm originating from Greece.

**Table 1.** The list of tolerant genotypes of food legumes for various problem soil conditions.

Problem soils	Crops	Identified source of tolerance	References
Al tolerance	Lentil	L7903, L4602, ILL6002	Singh <i>et al.</i> (2012)
Salinity tolerance	Lentil	NEL2704	Mamo <i>et al.</i> (1996)
		Masoor 93, Mansehra 89	Yasin <i>et al.</i> (2002)
		ILL5845, ILL6451, ILL6788, ILL6793, ILL6796	Ashraf and Waheed (1990)
		ILL 6796	Ashraf and Zafar (1997)
	Chickpea	DLG-103, LC-50, LC-53 and Sehore 74-3	Rai and Singh (1999)
		DZ-10-16-2	Mamo <i>et al.</i> (1996)
		SG-11, DHG-84-11	Singh and Singh (2001)
		ILC1919	Soussi <i>et al.</i> (2003)
	ICC10755, ICC13124, ICC13357, ICC15406, ICC15697, ICCV92318, ICCV92337, ICCV95332, ICCV95334, Jumbo 2	Serraj <i>et al.</i> (2004)	
	MCA 103, MCA 131 and MCA 250	Sadiki and Rabih (2001)	
Faba bean	Giza Blanca and Giza 674	Gaballah and Gomaa (2004)	
Alkalinity / Sodicity tolerance	Lentil	DLG-103, LC-50, LC-53 and Sehore 74-3	Rai and Singh (1999)
Boron tolerance	Peas	NGB1430 and NGB 2126	Bagheri <i>et al.</i> (1994)
	Lentil	ILL2024, ILL213A	Hobson <i>et al.</i> (2003)
		ILL8009, ILL5888, ILL8010, ILL2580	Yau and Erskine (2000)
Waterlogging tolerance	Lentil	ILL6439, ILL6778 and ILL6793	Ashraf and Chishti (1993)
	Chickpea	Line 946-512	Cowie <i>et al.</i> (1995)

Screening of primary, secondary and tertiary gene pools of field peas, lentil, chickpea and faba bean showed more frequent occurrence of tolerance to problem soils among landraces and wild species. Screening of 100 accessions of *Lens culinaris* subsp. *orientalis* under the greenhouse conditions (Fig. 1) showed 17 accessions as salt tolerant with average STR 1.5 (Table 2). Under hydroponic screening, only eight accessions (ILWL487, ILWL368, ILWL485, ILWL417, ILWL297, ILWL475, ILWL455 and ILWL136670) confirmed having high salt tolerant at 120 mM NaCl.

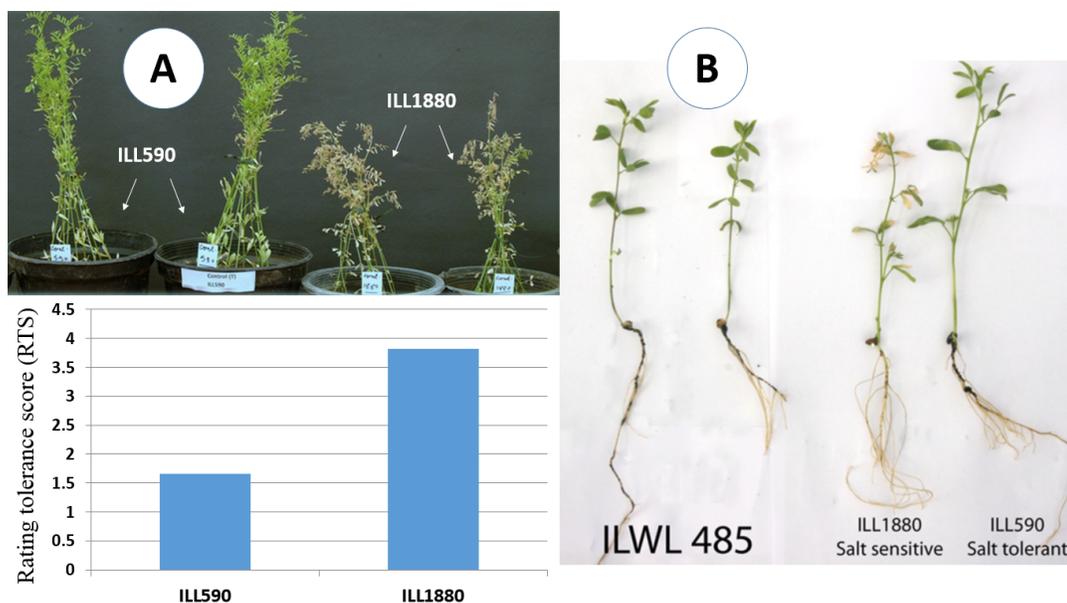
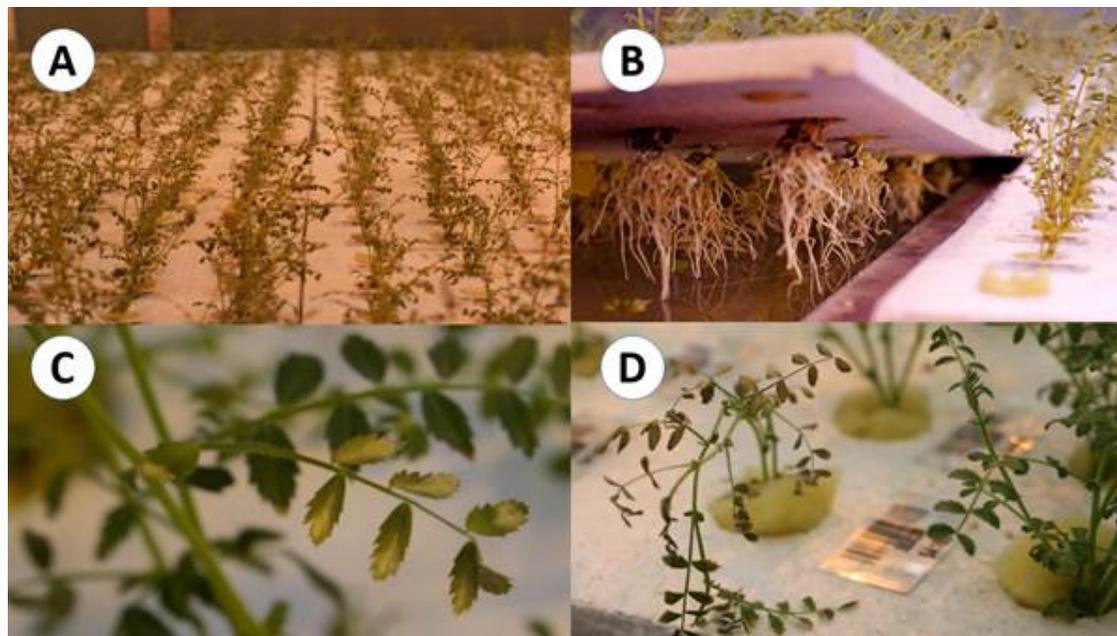


Figure 1. Screening of wild genotypes for salt tolerance in lentil. A) Comparison of ratings tolerance score (RTS) between accession ILL 590 (salt tolerant) and ILL 1880 (salt sensitive), B) ILWL485 was selected as salt tolerant by using bot and hydroponic culture at concentration 120 mM NaCl.

**Table 2:** Salt tolerant accessions of wild lentils and their salt tolerance rate (STR).

	<b>IG/ILWL</b>	<b>Replication 1</b>	<b>Replication 2</b>	<b>Average</b>
1	297	1	1	1
2	301	1	2	1.5
3	317	2	1	1.5
4	326	1	2	1.5
5	348	1	2	1.5
6	359	2	1	1.5
7	368	1	1	1
8	371	1	1	1
9	417	1	1	1
10	426	1	1	1
11	455	1	1	1
12	475	1	1	1

13	479	1	2	1.5
14	480	1	1	1
15	485	1	1	1
16	487	1	1	1
17	136670	1	1	1



**Figure:** Screening for salt tolerance in chickpea. A) high throughput hydroponic system, B) the roots immersed in Hoagland solution supplemented by the 100 mM NaCl, C) the primary symptoms appeared on the susceptible genotypes after salt treatment, D) completely plant death.

The crop response to salt stress depends on the crop growth stage. Genotypes that show salt tolerant at germination stage may show salt sensitivity at flowering and pod filling stages. Faba bean is more sensitive to salt stress during vegetative phase and less sensitive during flowering and pod filling stage (Al- Tahir and Al-Abdulssalam 1997).

Genetic variation for nutrient use efficiency has been reported in legume crops by comparing differential response of genotypes in terms of plant biomass and grain yield under low fertile/ input soil conditions with the normal soil conditions (Zhang et al. 2009; Parentoni et al. 2010).

Screening of two F<sub>2</sub> populations (LM2 and DR4) in half strength Hoagland and Arnon hydroponic culture (Asagawa 1985) supplemented with up to 100mM salt NaCl (Figure 1) showed that a single dominant gene (following ratio of 3:1) governs salt tolerance in chickpea. Sodium accumulation in the leave tissue showed higher concentration in susceptible parents as compared to resistant parents (Table 3).

**Table 3.** Primary results of the chickpea parents of the two F<sub>2</sub> population developed at ICARDA chickpea breeding program. These populations derived from salt tolerant and salt sensitive parent.

Pop. Code	Parents	STR	μmol Na/1g
LM2	ILC5309	4.8	1713.7
	ILC3397	1.6	718.6
DR4	S03087	4.4	1557.0
	FLIP87-8C	2.9	966.2

### 3.1.2 Breeding approach

In the case of complex polygenic traits like tolerance to salinity and boron toxicity, bulk and recurrent selection is adopted in order to increase the frequency of minor genes with additive effect. Single plant selections and progenies are tested in multiple rows/plots with many replications under stress conditions for several years at different locations. GEM interaction is studied to improve regional and general adaptability. Lentil cultivars with improved tolerance to NaCl have been released in Australia (Materne and Siddique 2009). Development of dwarf semi-leafless varieties of field peas has made a major significant in breeding for tolerance to problem soils.

Classical phenotyping methods usually involve measurement of genotypic differences for shoot and root parameters, observe relative changes in the physiological parameters including leaf elongation rate, leaf area expansion rate, relative growth rate and relative water content, *etc.*, and monitor health status by visual scoring in Zadoks scale (Hasnain et al. 2011). Development of stress tolerance indices and ranking of genotypes based on their score is a common practice (Genc et al. 2007; El-Hendawy et al. 2007; El-Hendawy et al. 2009; Ashkan and Moemeni 2013). It is helpful to screen and identify genotypic differences for a wide range of problem soils conditions across various food legume crops. However, there are several factors affecting the efficiency of classical phenotyping methods. Most of the classical methods focus on single physiological observation rather than whole plant response under stressed environment. As no single physiological parameter can contribute the whole plant tolerance to various abiotic stresses, it is recommended to measure the changes in the whole plant response rather than individual physiological observations (Hasegawa et al. 2000). Visual score of symptoms of nutrient deficiencies and toxicities is often objective, produces imprecise and unrepeatable results (Greenway and Munns 1980; Shannon 1985). Sometimes, symptoms of some nutrient deficiencies are often get confused with other nutrient deficiencies, plant damages caused by pests, diseases and other related stresses. For some soil nutrient

deficiencies such as Al tolerance, B tolerance, nitrogen deficiency and phosphate deficiency, roots need to be phenotyped. It is difficult to study phenotypic variation for root characteristics through classical methods, as they require huge amount of labour and time (Sirault et al. 2009). Further, most of the screening assays require destructive sampling, for example, harvesting of leave and shoot samples for nutrient analysis is expensive for screening Mg deficiency in faba bean in a large number of genotypes (Hariadi and Shabala 2004) and the same plant can not be further examined once it is disturbed or harvested completely.

Recent advances in the non-destructive imaging technology have allowed researchers to monitor the changes in growth, health and physiological status of whole plant response over time non-destructively. It is rapid, consumes less labour, time and produces reliable results. It is objective and helps to obtain quantitative data of symptoms of plant damages in a mapping population for further QTL analysis. Images taken using visible light (RGB) infrared rays and chlorophyll fluorescence are quite useful to detect changes in morphological and physiological status of the plants grown under stressed environment and detect nutritional deficiency tolerance at various stages of plant ontogeny. The pictures of the projected shoot area captured over time is useful to monitor changes in the growth rate of plants grown under stressed environments (Rajendran et al. 2009; Schilling et al. 2014). It was also quite useful to quantify macro-nutrients (N, P and K) deficiencies in faba bean and peas (Wiwart et al. 2009).

Plants grown under problem soil conditions demonstrate physiological responses similar to drought stress. This includes closure of stomata and reduction in the photosynthetic area. Therefore, it is possible to utilize the benefits of infra-red thermography to phenotype plants grown under various problems soil conditions (Sirault et al. 2009). It distinguishes genotypes based on the occurrence of stomatal conductance and hence the canopy temperature. Tolerant genotypes usually have high rate of stomatal conductance and possess cooler leaves than the sensitive ones (Berger et al. 2010). As the GE interactions influence the results under field conditions, it is used to detect genotypic differences in the stomatal conductance and transpiration rate of genotypes grown under controlled environments rather in the field environment (Berger et al. 2010; Furbank and Tester 2011). However, the physics of the heat flux is highly variable and complicates the measurement in many times (Fiorani and Schurr 2013). Frequently, a combination of RGB images with infrared thermography is useful to study leaf orientation, canopy structure and canopy temperature of plants grown under stressed environments (Leinonen and Jones 2004). Likewise, chlorophyll fluorescence has a wide application in high throughput plant phenotyping from lab to field studies. It is useful to measure photosynthetic efficiency, electron transport rate and the extent of non-photochemical quenching. The parameters of photochemical and non-photochemical quenching coefficients are useful to study Mg deficiency in broad beans (Hariadi and Shabala 2004b). Most of the times, portable instruments used to measure photosynthetic status of plants create difficulties in terms of robustness, reproducibility and data analysis (Fiorani and Schurr 2013). Along with RGB imaging, chlorophyll fluorescence can provide information about the leaf area, growth rate and leaf senescence. Hence, the interpretation of results from both RGB images and

fluorescence images could be more efficient to do early detection of nutrient deficiencies and toxicities and to quantify plant damages.

Hyper-spectral imaging utilizes the wavelength from 400 to 2500 nm, which falls between the spectral range of visible and near-infrared (NIR) regions. It has a major application in remote sensing (Kokaly et al. 2009). Hyper-spectral reflectance measurements are useful to identify wave band signatures that help to indicate plant stress levels. It is useful to determine reflectance vegetation indices and helps to estimate biomass, healthiness, pigment composition, photosynthetic status, leaf thickness, growth habit and relative water content (Berger et al. 2010; Fiorani and Schurr 2013). In the current field of research it is used to detect deficiencies caused by N (Shi et al. 2011a; Shi et al. 2012a; Shi et al. 2011b), P (Shi et al. 2012b) and Ca (Li et al. 2005) in various crops. NIR spectral imaging needs extensive calibration before usage. Once calibrated, it facilitates non-destructive quantification of water content, protein content and other related compounds for large-scale phenotyping. All these high throughput plant-imaging technologies often necessitate highly automatic and rapid imaging of plants grown under controlled environments. Greenhouse facilities setup with the conveyor belts for delivery of plants to the imaging system, watering and weighing of plants in an automated way would help to do more precision fast phenotyping. Such greenhouse facilities have already been used to phenotype plant traits including plant height, width, area and biomass, chlorophyll, anthocyanin and foliar water content, in corn, soybean, and cotton (Hyundae et al. 2014). It is also used to detect deficiencies of Ca (Story et al. 2010) and Mg (Chaerle et al. 2007) in various crops. The AGH facilities are now available at Crop Design in Belgium (<http://www.cropdesign.com/general.php>), Leibniz Institute of Plant Genetics and Crop Plant Research-IPK in Germany (<http://www.ipk-gatersleben.de/>), Australian Plant Phenomics Facility in Australia (<http://www.plantphenomics.org.au/>). They are often useful to do screening in germplasm and breeding populations to identify QTL and candidate gene(s) under controlled conditions; later these results can be validated in the well-planned field experimentation.

## **3.2 Genomics approaches**

### ***3.2.1 Marker Assisted Selection***

In peas, a total of 36,188 single nucleotide polymorphism (SNP) markers associated with expressed sequence tags (ESTs) were developed and used for mapping and QTL analysis (Leonforte et al. 2013). Sequences associated with the mapped molecular markers were used for comparative genomic analysis with other legume species. They found higher levels of conserved synteny were observed with the genomes of *Medicago truncatula* Gaertn. and chickpea (*Cicer arietinum* L.) than with soybean (*Glycine max* [L.] Merr.), *Lotus japonicus* L. and pigeon pea (*Cajanus cajan* [L.] Millsp.). QTLs for salinity tolerance were identified on linkage groups Ps III and VII, with flanking SNP markers suitable for selection of resistant cultivars. Comparison of sequences underpinning these SNP markers to the *M. truncatula* genome defined genomic regions containing candidate genes associated with saline stress

tolerance (Table 4). This constitutes an important set of tools for marker-assisted selection (MAS) programs aimed at performance enhancement of field pea cultivars.

**Table 4.** Identification of QTLs for salt tolerance on the Kaspia x Parafield genetic map in peas (Leonforte et al. 2013).

Marker	Linkage group	Position (cM)	Phenotypic variance (%)
SNP_100000313 - SNP_100000353	PS III	179- 184	12
SNP_100000318 - SNP_100000130	PS VII	218 - 222	19
SNP_100000313 - SNP_100000353	PS III	179 - 184	12
SNP_100000318 - SNP_100000130	PS VII	218 -222	17

In lentil, Kaur et al. (2014) used a subset of 546 SSRs and 768 SNPs for genetic mapping of an intraspecific mapping population (Cassab × ILL2024) that exhibits segregation for B tolerance in lentil. A single genomic region (single gene segregation) was associated with variation for B tolerance based on evaluation performed over 2 years (2008-2009), on LG4.2 in the 7.5 cM interval between the markers SNP\_20002998 and SNP\_20000246. The comparison of flanking markers to genome sequences of model species (*M. truncatula*, soybean and *Arabidopsis thaliana*) identified candidate genes that are functionally associated with B tolerance, and could potentially be used for diagnostic marker development in lentil.

In chickpea, Varshney et al. (2009) generated salinity responsive ESTs from root tissues of NaCl treated plants of salinity-tolerant and salinity-sensitive genotypes. They developed 8,258 ESTs (2,595 unigenes) from salinity challenged libraries. The salinity-sensitive genotype (ICCV 2) exhibited a relatively more stunted growth pattern than salinity-tolerant genotype (JG 11) when these genotypes were exposed to salinity stress (80 mM). Their results indicated a high copy number of transcripts corresponding to the ABR gene (UniProt ID: Q06931) were moderately expressed in ICCV 2\_Salinity library (55 transcripts).

### 3.2.2 Transcriptomics

To investigate the molecular mechanisms of Al toxicity in leaves of broad bean, a suppression subtractive hybridization (SSH) library was constructed to identify up-regulated genes: cDNA from leaves subjected to 12, 24, 48 and 72 h of 50 and 100 µM AlCl<sub>3</sub> stress were used as testers and cDNA from leaves subjected to 0 µM AlCl<sub>3</sub> treatment for the same lengths of time as above were used as a driver. The SSH analysis identified 156 non-redundant putative Al stress-responsive expressed sequence tags (ESTs) out of 960 clones. The ESTs were categorized into ten functional groups, which were involved in metabolism (0·21), protein synthesis and protein fate (0·10), photosynthesis and chloroplast structure (0·09),

transporter (0.08), cell wall related (0.06), signal transduction (0.05), defence, stress and cell death (0.05), energy (0.03), transcription factor (0.03) and unknown proteins (0.30). The effect of Al treatment on expression of 15 selected genes was investigated by reverse transcription polymerase chain reaction (RT-PCR), confirming induction by Al stress. The results indicated that genes involved in organic acid metabolism, transport, photosynthesis and chloroplast structure, defence, stress and cell death might play important roles under Al stress.

### 3.2.3 Transgenics

When a plant is subjected to abiotic stress, a number of genes are turned on, resulting in increased levels of several metabolites and proteins, some of which may be responsible for conferring a certain degree of protection against these stresses. A key to progress towards better crops under stress has been to understand the changes in cellular, biochemical and molecular machinery that occurs in response to stress. Modern molecular techniques involve the identification and use of molecular markers that can accelerate genetic gains. However, the introgression of genetic proteins (QTLs) involved in stress tolerance often brings along undesirable agronomic characteristics from the donor parents. This is because of the lack of proper understanding of the key genes underlying the QTLs. Therefore, the development of genetically engineered plants by the introduction and/or over expression of selected genes seems to be a viable option to hasten the breeding of “improved” plants. Following these logical steps, various transgenic techniques have been used to improve stress tolerance in plants (Allen 1995).

A large number of studies have evaluated different transgenic constructs in different plant species, and to different stresses such as drought, salinity and cold. The expression of the genes inserted as well as altered levels of metabolites have been reported in great details. However, less detail is given with regard to methods used to evaluate the stress response (Bhatnagar-Mathur et al., 2008). This lack of details applies mostly to drought stress (Holmstrom et al., 2000). Stress conditions used to evaluate the transgenic material in most of the cases are usually too severe (Shinwari et al., 1998; Garg et al., 2002) as plants are very unlikely to undergo such stresses under field conditions. While the use of PEG (polyethylene glycol) in hydroponics can be useful to test certain response of plants under given osmotic potential as reported by Pilon-Smits et al. (1996; 1999), it offers relatively different conditions than in the soil.

In a salt stress study in *Lotus japonicus* metabolite profiling revealed gradual increase of many soluble small molecules that were compared to similar stress in forage legume *Lotus* species. The results showed that only a few salt responsive metabolites were common in all species examined (Sanchez et al. 2012). Many workers stated that the best discrimination between sensitive and tolerant lines in hydroponic or sand culture was achieved at ‘moderate’ stress levels, which depended on the species concerned, being 25 mM NaCl for chickpea (Sadiki and Rabih 2001), 75 mM for faba bean (Cordovilla et al. 1995) and either 70 or 75 mM for pea (Hernández et al. 2000; El-Hamdaoui et al. 2003; Gómez et al. 2004). Lentil responses varied with both growth stage and salinity level (Ameen 1999), ranging from ILL 5582 (susceptible at flowering and sensitive based on seed yield) to ILL 5845 (tolerant at

flowering and tolerant based on seed yield). In ICRISAT and ICARDA several chickpea RIL populations have been developed to understand the nature of tolerance (Table 4). These populations will help to identify how many genes controlling salt tolerance in chickpea and to identify markers linked to these gene(s).

Table 4. List of population developed between parents showing different responses to salinity resistance in chickpea.

Population code	Cross	Generation	Chickpea seed type
ICCRIL01	ICCV 2 × JG 62	F10	Desi
ICCRIL08	ICC 6263 × ICC 1431	F8	Desi
ICCRIL09	ICCV 2 × JG 11	F8	Desi
Salt4	ILC10722 X CPI060546	F5	Kabuli
LM2	ILC5309 X ILC3397	F5	Kabuli
DR4	S03087 X FLIP87-8C	F5	Kabuli

### 3.3 Agronomic manipulations

Problem soils have serious physical and chemical limitations to crop cultivation. Special management techniques are required for satisfactory production from such soils. Agronomic practices such as irrigation, leaching and drainage practices, soil conservation measures, cover crops mulching, application of amendments, manuring, tillage, whichever is appropriate to a particular problem soil condition, hold promise to bring back these problem soils into crop cultivation. Saline soils can be managed through adequate irrigation, leaching, draining, and salt-tolerant crops. Sodic soil management needs chemical amendments such as application of sulfur and gypsum along with leaching and draining. Management of acid soils involves liming, leaching, and safe disposal of acid-wash water along with cultivating acid-tolerant crops (Khan, 2013). Management options for acid/acid sulphate soils, saline soils, and alkaline/sodic soils are discussed hereunder:

#### 3.3.1 Application of soil amendments

Soil amendments are materials such as lime, gypsum or calcium chloride, sulphur or sulphuric acids, organic matter and plant roots, which are commonly used to reclaim the problems soils. The kind and quantity of a chemical amendment to be used for replacement of exchangeable sodium in the soils depend on the soil characteristics including the extent of soil deterioration, desired level of soil improvement including crops intended to be grown and economic considerations. Saline soils are often dominated by neutral salts and therefore, an amendments may not be needed (Siyal et al. 2002). In case of alkaline/sodic soils with high amount of sodium salts, an amendment needs to be added for losing sodium from the soil before it can be leached.

Gypsum is the most commonly used amendment for removing sodium from the soil particles, as it is cheaper and readily available (Raza et al. 2001; Ardakani and Zahirnia, 2006). Gypsum is a slightly soluble salt of calcium and sulfate. Therefore, it reacts in the soil very slowly for a long period. Organic matter (i.e. straw, farm and green manures), decomposition

and plant root action also help dissolve the calcium compounds found in most soils, thus promoting reclamation but this is relatively a slow process. Addition of organic matter in conjunction with gypsum reduce the adverse soil properties associated with sodic soils (El-Banna et al. 2004; Abou El-Defan et al. 2005; Moustafa 2005; Wong et al. 2009; Abd Elrahman et al. 2012). Application of organic manure in composition with gypsum decreases the pH (Negm et al. 2003), EC and ESP (Abou El-Defan et al. 2005) and pH, ECe, soluble ions, SAR and DTPA-extractable heavy metals (Abd Elrahman et al. 2012). These amendments increase the soluble Ca concentrations leading to enhance flocculation of soil colloids (Abd Elrahman et al. 2012). Citrate increased the dissolution rate by two fold in comparison to without citrate and complexed Al in solution and also directly from the mineral phase (Jones and Kochian 1996). Calcium chloride is also used to reclaim alkali soils.  $\text{CaCl}_2$  converts  $\text{Na}_2\text{CO}_3$  into  $\text{NaCl}$  precipitating  $\text{CaCO}_3$ . Sodium chloride ( $\text{NaCl}$ ) is drained off by leaching water. Spent acids ( $\text{HCl}$ ,  $\text{H}_2\text{SO}_4$ , etc.) can also be used to reduce the excess  $\text{Na}_2\text{CO}_3$  in the soil. Urea can also be used to reduce soil alkalinity/salinity primarily in those regions where it is available cheaply (Abrol et al. 1988). The  $\text{NH}_4$  (ammonium) present in urea which is a weak cation releases the strong cation Na from the soil structure into water. Thus, alkali soils absorb/consume more urea compared to other soils.

Liming of drainage water has been applied to reclaim acid sulfate soils in Australia. The second strategy is to try to limit pyrite oxidation by maintaining a high groundwater table. A precondition is the availability of sufficient water. This method also requires substantial investments in water management, while the potential danger of acidification remains present. This strategy is widely followed, both in temperate regions and in the tropics, often with ingenious adaptations to suit local conditions and practices. Incorporation of lime into the upper cultivable soil layer is an effective and dominant practice to raise soil pH and reduce acidity-related constraints to improve crop yields (Fageria and Baligar 2008). The quantity of lime required depends on the soil type, quality of liming material, costs and crop species or cultivars (Fageria and Baligar 2008). Banding or pelleting lime onto the seed at sowing is also a common practice used to aid with the establishment of temperate pasture legumes. Several papers have summarized positive effects of lime use for reclamation of acid soils (Lathwell 1979; Lopes 1983; Miranda et al. 1990; The et al. 2006; Moreira and Fageria 2010; Kisinyo, 2011; Verde et al. 2013; Kisinyo et al. 2014). Moreover, application of lime tends to raise the soil pH by displacement of  $\text{H}^+$ ,  $\text{Fe}^{2+}$ ,  $\text{Al}^{3+}$ ,  $\text{Mn}^{4+}$  and  $\text{Cu}^{2+}$  ions from soil adsorption site (Onwonga et al. 2010; Kisinyo et al. 2012). More than increasing soil pH, it also supplies significant amounts of Ca and Mg, depending on the type. Indirect effects of lime include increased availability of P, Mo and B, and more favourable conditions for microbial mediated reactions such as nitrogen fixation and nitrification and in some cases, it also improves soil structure (Crawford et al. 2008). In addition to correcting soil acidity, lime also diminishes the P-fixation capacity by precipitating exchange Al as  $\text{Al}(\text{OH})_3$  (Haynes 1982) and stimulates root development at depth depending on rate of incorporation and time after application (Lopes 1983; Goedert 1987). Lime should be incorporated as deeply as possible and targeted for residual effect by using a coarser lime (Lopes and Guilherme 1994). After this preparatory

period, one third or one half of the lime requirement can be applied in the clayey and sandy soils without ploughing (Lopes 1996).

### 3.3.2 Leaching of salts and drainage

Leaching excess salts and maintaining a favourable salt balance remain the best strategy to prevent detrimental salt accumulation in the soil profile. In order to improve salt affected soils, it is necessary not only to leach salts but also to have adequate drainage (Siyal et al. 2002). The drainage system must provide an outlet for the removal of the leachates as well as keep the water table deep enough to prevent salt laden groundwater moving up to the root zone. This is particularly a problem for soils with a shallow saline water table (Schilgaarde 1974). Where shallow water tables limit the use of leaching, artificial drainage may be needed. Cut drainage ditches in fields below the water table level to channel away drainage water allow the salts to leach out. Drainage tile or plastic drainpipe can also be buried in fields for this purpose. Proper design and construction of a drainage system is critical.

Clay soils in high annual rainfall (more than 1000 mm) zones do not generally suffer from high alkalinity as the rainwater runoff reduces the soil salts to comfortable levels if proper rainwater harvesting methods are followed. In some agricultural areas, the use of subsurface "tile lines" are used to facilitate drainage and leach salts. Continuous drip irrigation would lead to alkali soils formation in the absence of leaching/drainage water from the field.

Amount of water required to leach down the salts depends upon the amount and type of salts in the soil, soil texture, level of salts desired in the root zone (depends on the crop to be grown) and depth of reclamation (Table). The leaching requirement of the soil varies with texture of soil as found by CSSRI (Anon. 2004). However, leaching of salts from the root zone may increase the salt content of the groundwater. Choice of leaching salts from root zone can create another problem and therefore may not be a generally accepted practice.

Table. Leaching requirement and water requirement for different types of soil

Soil type	Leaching requirement(cm cm <sup>-1</sup> soil depth)	Water required to leach 60 cm of soil profile(cm)
Coarse textured	0.5-0.6	30-36
Medium textured	0.6-0.8	36-48
Heavy textured	0.8-1.0	48-60

### 3.3.3 Improved irrigation practices

**Pre-plant Irrigation:** Legumes are more susceptible to salt injury during germination or in the early seedling stages. An early-season application of good quality water, designed to fill the root zone and leach salts from the upper 6 to 12 inches of soil, may provide good enough conditions for the crop to grow through its most injury-prone stages.

**Irrigation frequency management:** Salts are most efficiently leached from the soil profile under higher frequency irrigation (shorter irrigation intervals). Keeping soil moisture levels higher between irrigation events effectively dilutes salt concentrations in the root zone, thereby reducing the salinity hazard.

Most surface irrigation systems (flood or furrow systems) cannot be controlled to apply less than 3 or 4 inches of water per application and are not generally suited to this method of salinity control.

***Irrigation methods:*** Double-row bed systems require uniform wetting toward the middle of the bed. This leaves the sides and shoulders of the bed relatively free from injurious levels of salinity. Without uniform applications of water (one furrow receiving more or less than another), salts accumulate closer to one side of the bed. Periodic leaching of salts down from the soil surface and below the root zone may still be required to ensure the beds are not eventually salted out.

Alternate furrow irrigation may be desired for single-row bed systems. This is accomplished by irrigating every other furrow and leaving alternating furrows dry. Salts are pushed across the bed from the irrigated side of the furrow to the dry side. Care is needed to ensure enough water is applied to wet all the way across the bed to prevent build up in the planted area. This method of salinity management can still result in plant injury if large amounts of natural rainfall fill the normally dry furrows and push salts back across the bed toward the plants. This phenomenon also occurs if the normally dry furrows are accidentally irrigated.

Sprinkler-irrigated fields with poor water quality present a challenge because it is difficult to apply enough water to leach out the salts and utilize row or bed configurations to manage accumulation. Growers should monitor the soil EC and irrigation water salinity. Sprinkler systems, particularly center-pivot and linear-move systems configured with low energy precision application (LEPA) nozzle packages or properly spaced drop nozzles, and drip irrigation systems provide the best control to allow this type of salinity management.

#### ***3.3.4 Nutrient management***

Application of organic and green manures also helps reclaim acid soils. Application of organic manures to the acid soils increases soil pH (Adeniyani et al. 2011), Mg availability (Adeleye et al. 2010) and available P and reduces Al toxicity (Kisinyo et al. 2014). Use of P fertilizer also increases the available P in P deficit tropical acid soils (The et al. 2006; Kisinyo et al. 2014). Similar increases of available P in acid soils of western Kenya reported by Opala et al. (2007; 2010a). Integrated nutrient management involving lime application, P fertilization, organic manures, seed treatment and seed inoculation with rhizobium reduced the acidity and increased the productivity of acid soils (Bal 2001; Verde et al. 2013; Kasinya et al. 2014). Manure is a source of nutrients, which are released through mineralization, thus supplying the necessary elements for plant growth (Chiezey and Odunze 2009), and when combined with P fertilizers it increased nutrient supply which enhanced vegetative growth, affecting plant height and yields (Umoetok et al. 2007).

Organic matter has been implicated in the reduction of aggregate breakdown in sodic and alkali soils (Gupta and Abrol 1990; Barzegar et al. 1997; Badia 2000; Harris and Rengasamy 2004). Application of organic matter to these soils enhances microbial activity that transforms the newly added organic material into polysaccharides and long chain aliphatic compounds capable of binding and stabilizing soil aggregates (Parr et al. 1989; Lado et al. 2004). Organic materials including crop straw (Barzegar et al. 1997), cottage cheese whey and

fly ash (Graber et al. 2006), synthetic organic polymers and farmyard manure (Negm et al. 2003), slaughterhouse wastes and poultry excreta (Qadir et al. 2001) and green manuring (Gupta and Abrol 1990; Ghai et al. 1988; Ilyas et al. 1997) are used to ameliorate sodic and alkali soils but at a slow rate. Combined use of organic and inorganic amendments accelerates the rate of soil amelioration (Negm et al. 2003; Abou El-Defan et al. 2005; Wong et al. 2009; Abd Elrahman et al. 2012). An example is the application of farmyard manure or green-manuring of common vetch (*Vicia sativa* L.) with gypsum.

### **3.3.5 Tillage management**

Crop productivity in some sodic and alkali soils is often associated with low macroporosity resulting in restricted infiltration rate because of fine texture, excess of silt, hard pan, or stratification. A number of tillage options have been used to create a rough and thoroughly disturbed root zone with improved hydraulic properties. These options include: deep ploughing, subsoiling, sanding, minimum tillage, hauling and profile inversion. Deep-ploughing consists of ploughing to a depth from about 0.5 m to more than 1.0 m. This method may be used on stratified soils having impermeable layer(s) lying between permeable layers (Kovda et al. 1973) and gypsiferous soils if the Ca<sup>2+</sup>-rich layer occurs in the subsoil (Rasmussen et al. 1972). Therefore, deep tillage to the subsoil is beneficial for the improvement of the saline soil (Xiong et al. 2012). The depth of deep-ploughing should not be so shallow that only the A and a part of B horizons are mixed, otherwise the physical condition of the surface is likely to be worsened or, at best, only short-lived change may result (McAndrew and Malhi 1990). The cost of deep-ploughing and requirements of heavy machinery, however, make it unacceptable to subsistence farmers (Grevers and de Jong 1993).

On the other hand, agricultural mechanization and intensive and continuous tillage operations over the decades lead to soil degradation, declining fertility and increasing soil salinity (Niu and Wang, 2002). Conservation tillage is an effective method to improve soil fertility and structure and leads to a reduction of soil evaporation and salinity (Li et al. 2010). Maintaining plant residues atop soil surface in conservation agriculture practice can limit soil evaporation and salinity at that layer (Ma et al. 2010). The conservational tillage results in increased organic matter and soil permeability, salt leaching from the surface to deeper layers and ultimately amendment of salty and sodic soils (Hulugalle et al. 1997; Chatterjee and Lal (2009); Qingjie et al. 2014).

### **3.3.6 Crop residue management**

Crop residue or mulch to the soil reduces evaporative water losses, thus decreases the upward movement and accumulation of salts into the root zone (Provin and Pitt, 2001). . Evaporation and thus, salt accumulation, tends to be greater in bare soils. Fields need to have 30 to 50 per cent residue cover to significantly reduce evaporation. Under crop residue, soils remain wetter, allowing fall or winter precipitation to be more effective in leaching salts, particularly from the surface soil layers where damage to crop seedlings is most likely to occur. Plastic mulches used with drip irrigation effectively reduce salt concentration from evaporation. Sub-surface drip irrigation pushes salts to the edge of the soil wetting front, reducing harmful effects on seedlings and plant roots.

Various agricultural residues have been tested to ameliorate soil acidity, but their application is often constrained by their limited efficiency, especially in strongly acidic soils. Firstly, the alkalinity released during the decomposition of residues may not be adequate to meet the need of highly acidic soil (Haynes and Mokolobate 2001). Secondly, plant residues with high alkalinity and low CN ratios can decompose and release alkalinity rapidly, but nitrification of organic N may reduce the liming potential of these materials at later stages of decomposition (Mao et al. 2010). Residues with low CN ratios tend to exhibit net N mineralization, while the residues with high CN ratios exhibit net N immobilization (Van Kessel et al. 2000; Qian and Schoenau 2002). The contribution of the N cycle on soil pH change is therefore important. Crop residues with high CN ratio combined with a liming agent such as alkaline slag ameliorate soil acidity efficiently (Wang et al. 2012). The liming effect of alkaline slag is likely to be negated if added in combination with residues with high N contents.

### **3.3.7 Crop and cropping systems**

Selection of crop and cropping systems can be a good management tool for moderately salt-affected soils. Just as crops differ in tolerance to high salt concentrations, they also differ in their ability to withstand high sodium concentrations.

The recommended cropping sequences for saline soils are pearl millet- barley/ wheat/ mustard, sorghum-wheat/ barley/ mustard, cluster bean- wheat/ barley, and cotton, wheat/ barley. Pearl millet or sorghum based cropping sequence is more remunerative than cotton based, as the winter crops that follow cotton are less productive. It can be possible to include cool-season legumes in these cropping systems for better productivity under salt affected soil condition. Release of H<sup>+</sup> by legume crops might increase the Na removal from sodic calcareous soils (Qadir et al. 2003).

### **3.4 Crop-mediated amelioration (Phytoremediation)**

Crop-mediated amelioration – phytoremediation – refers to the improvement of acidic, sodic and alkali soils by growing certain plant species that can withstand ambient levels of soil acidity, salinity and sodicity without the application of an amendment, i.e., growing a range of crops together with the application and infiltration of excess water during the cropping season. Synonymous terms for phytoremediation include vegetative bioremediation, bioremediation or biological reclamation. This crop-mediated strategy works through plant root action to dissolve native or precipitated calcite in sodic soils to provide sufficient Ca<sup>2+</sup> to cause an effective Na<sup>+</sup>–Ca<sup>2+</sup> exchange at the cation exchange sites resulting in a marked decrease in soil sodicity levels (Robbins 1986; Ahmad et al. 1990; Qadir et al. 1996; Qadir and Oster 2004). Many sodic and alkali soils of the world are calcareous with the occurrence of calcite at varying depths.

Typical plant-based strategies for contaminated soils, such as those having elevated levels of heavy metals, work through the cultivation of specific plant species capable of hyper-accumulating target ionic species in their shoots, thereby removing them from the soil (McGrath et al. 2002). In contrast, phytoremediation of sodic and alkali soils is achieved by the ability of plant roots to increase the dissolution rate of calcite. The salinity–sodicity

combination present in the soil solution during phytoremediation maintains adequate soil structure and aggregate stability that enhance the amelioration process (Oster et al. 1999).

The process of Na<sup>+</sup> removal from calcareous sodic soils during phytoremediation is driven by (1) increased PCO<sub>2</sub> within the root zone (Shainberg and Letey, 1984; Qadir et al., 1996); (2) enhanced proton (H<sup>+</sup>) release in the rhizosphere in case of certain N<sub>2</sub>-fixing crops (Qadir et al., 2003); and (3) enhanced Na<sup>+</sup> uptake in the shoots, which is removed through harvesting of above-ground biomass and its subsequent export from the field (Gritsenko and Gritsenko, 1999). In addition, roots of the phytoremediation crops play an important role in improving soil aggregation and hydraulic properties within the root zone (Ilyas et al., 1993; Akhter et al., 2004). The collective effects of these factors ultimately lead to soil amelioration, provided drainage is present and adequate leaching occurs (Qadir and Sharma 2006). Among these factors, PCO<sub>2</sub> has been identified as the single largest driving force for soil amelioration, suggesting the need to identify different crops and crop management practices that enhance CO<sub>2</sub> production within the root zone to ameliorate sodic and alkali soils more efficiently, especially in areas where chemical amendments are not available or are too expensive.

The phytoremediation by halophyte is more suitable as it can be executed very easily without any problems. Several halophyte species including grasses, shrubs, and trees can remove the salt from different kinds of salt-affected problematic soils through salt excluding, excreting, or accumulating by their morphological, anatomical, physiological adaptation in their organelle level and cellular level. Exploiting halophytes for reducing salinity can be good sources for meeting the basic needs of people in salt-affected areas as well (Hasanuzzaman et al. 2014). A recent study revealed an increased efficiency of *Atriplexhalimus* with increasing salinity which suggest it a good candidate for soil desalination in arid and semi-arid regions (Abdul-Kareem and Nazzal 2013).

#### **4. Future perspectives**

The non-destructive imaging platforms provide an opportunity for better characterization of the stress and precision phenotyping of germplasm. From images, it is possible to characterize the shape of a plant's shoot using various mathematical descriptors such as compactness and centre of mass. In addition to alterations in plant growth, these plant measurements would be useful to screen germplasm for osmotic stress tolerance. These identified sources of variability will help to develop tolerant cultivars. Genetic variation in *Aegilops tauschii* for salinity tolerance was useful to develop synthetic hexaploids of bread wheat with improved salinity tolerance (Schachtman et al. 1992) through conventional breeding approach. On the other hand, marker assisted selection has facilitated to improve salinity tolerance of wheat cultivars and those cultivars can yield more than 25% in the saline soils in Australia (James et al. 2011). Past research has been useful to understand the variability of the traits related to problem soil conditions. In most studies, the genetic control of tolerance mechanisms are in question which seriously limits the plant breeding efforts for developing tolerant varieties. Recent studies suggest polygenic inheritance for tolerance to problem soils in which several minor genes with small individual effects are in play. The expression of these traits is highly subjected to the environmental conditions. In wheat, however, the monogenic inheritance was observed for Al

tolerance while in barley and maize, it has been confirmed as a polygenic trait with additive gene actions (de Almeida et al. 2002; Ferreira et al. 2006; Minella and Sorrells 1997). Incidence of genotype-environment (GxE) interaction causes difficulty on visual selection of traits with polygenic inheritance (Cobb et al. 2013). Besides, GxE interactions, other physiological and genetic interactions between physiological pathways and genes controlling various component traits also mask the phenotypic effect. Development of reliable phenotyping technique that can accurately examine these interactions and distinguish genotypes for these differences could help to develop a successful tolerant cultivar.

#### *Focusing on plants root traits under problem soil conditions*

Similar to shoots, roots also show rapid reduction in growth rate under stress conditions. Root length, root density, number of nodules, reduction in growth rate, recovery rate, etc., can be monitored. Very little work has been done so far, however, to study the effect of salt on plant root growth because it is hard to characterize *in vivo* plant root responses under stressed environments. A rhizotron system oriented with an imaging device is widely preferred by various researchers to study the response of wheat under stressed conditions (Andr n *et al.*, 1996; Pan *et al.*, 1998). Similarly, a rhizotron attached with digital and near infra-red spectroscopy could be useful to characterize the changes in the biological processes of root growth, development, activity and longevity under salt stressed environments (Vamerli *et al.*, 2012; French *et al.*, 2012).

#### *Use QTL pyramiding approach to develop tolerant cultivar for the near future*

One of the usefulness of marker assisted selection would be pyramiding genes linked to all tolerance components and developing a cultivar which is suitable to grow under different environmental conditions. A gene pyramiding approach has been suggested by various researchers to develop salinity tolerant cultivars in various crop species (Lin et al., 2004). It involves selection of physiological traits which can correlate well with the salinity tolerance and combine the alleles with similar effects from different loci (Xu, 2010). A QTL pyramiding approach would help to develop cultivars with more salinity tolerance and it can better survive than existing commercial cultivars under saline environments.

The lines tolerant to B toxicity were developed for Southern grain belt. In contrast, B deficiency limits lentil cultivation in Nepal. Cultivars with efficient Fe uptake is required for alkaline soils of Syria and Australia.

## **5. Conclusion**

The HT phenotyping technologies are useful to study the phenotypic changes in the growth, health and morphological changes of plants grown under different problem soils. The advantages of HT phenotyping are well recognised by breeders, physiologists and soil-scientists involved in the current field of interest. Along with the advances plant genomics research area, recent developments in plant phenomics would help to do more precise

measurement of plant traits, identify new plant traits and gene loci towards tolerance to various problem soils and helpful to either modify or develop new cultivars for future need.

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