



Applying hyperspectral imaging to explore natural plant diversity towards improving salt stress tolerance



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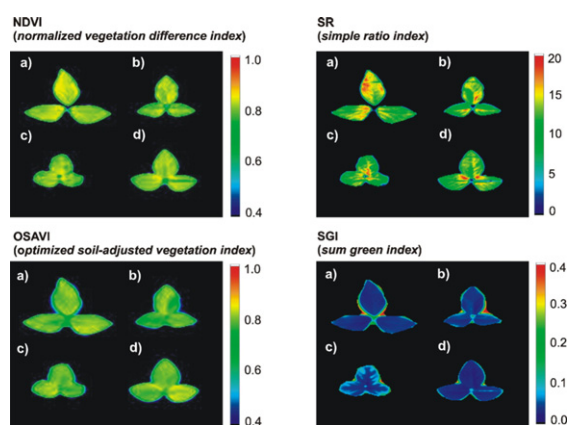
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HIGHLIGHTS

- Salinity represents an abiotic stress constraint affecting growth and productivity of plants
- Better solutions is to improve the level of salt resistance using natural genetic variability within crop species
- Phenomic methodology employing different non-invasive imaging systems for detecting quantitative and qualitative changes caused by salt stress at the whole plant and canopy level. Hyperspectral imaging techniques provide unique opportunities for fast and reliable evaluation of numerous characteristics associated both with various structural, biochemical and physiological traits
- Salt-soil-plant interaction and sustainable coastal agriculture need powerful phenotyping tools

GRAPHICAL ABSTRACT



Examples of the structural reflectance parameters derived from hyperspectral imaging of soybean leaves treated with salt stress. a) and b) are leaves from non-salt conditions and c) and d) are leaves from plants treated with 250 mM NaCl concentration. Pictures of a) and c) show the soybean leaf of genotype SA-108 and b) and d) shows the soybean leaf of genotype SA-136. Leaves (c) shown a typical symptomatic of damage induced by salt stress conditions

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ABSTRACT

Salinity represents an abiotic stress constraint affecting growth and productivity of plants in many regions of the world. One of the possible solutions is to improve the level of salt resistance using natural genetic variability within crop species. In the context of recent knowledge on salt stress effects and mechanisms of salt tolerance, this review present useful phenomic approach employing different non-invasive imaging systems for detection of quantitative and qualitative changes caused by salt stress at the plant and canopy level. The focus is put on hyperspectral imaging technique, which provides unique opportunities for fast and reliable estimate of numerous characteristics associated both with various structural, biochemical

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and physiological traits. The method also provides possibilities to combine plant and canopy analyses with a direct determination of salinity in soil. The future perspectives in salt stress applications as well as some limits of the method are also identified.

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The salinity represents the harsh environmental stress limiting crop production on ~20% of irrigated land worldwide (Mudgal et al., 2010). In arid and semi-arid areas, the irrigation is often used to compensate the lack of rainfall in drought prone regions; however, the intensive long-term irrigation of arable lands tends to increase salinity of soil to the extent which is unacceptable for crop plants (Flowers, 2004; Tang et al., 2015). Regarding the classification of plants based on their capability to growth on high salt solutions, the species can be divided into glycophytes or halophytes. The most of the crop plants are considered as glycophytes, not able tolerate a high concentration of salts (above 1–3 g L⁻¹). Typical examples of glycophytes are beans and rice. On the other hand, the representatives of halophytes can tolerate higher quantity of salts in the root environment. There are a few of the crop species, which can be denoted as a typical halophyte. As an example, *Salicornia bigelovii* (dwarf glasswort) grows well at the root medium containing 70 g L⁻¹ of dissolved salts (Glenn et al., 1998). The marginal halophytes, such as date palm (*Phoenix dactylifera*) and barley (*Hordeum vulgare*) are able to tolerate about 5 g L⁻¹ (Glenn and Brown, 1999). Numerous species also belong among the potentially useful halophytes, such as *Atriplex* (saltbush, orache, orach), *Attalea speciosa* (babassu), *Anemopsis californica* (yerba mansa, lizard tail), *Panicum virgatum* (switchgrass), *Spartina alterniflora* (smooth cordgrass), *Salicornia bigelovii* (dwarf glasswort, pickleweed), *Tetragonia tetragonoides* (warrigal greens, kōkihi, sea spinach).

Thus, the majority of crop species represent the glycophytes notwithstanding the conditions of saline soils (Gupta and Huang, 2014). The future prognosis is rather pessimistic: in 2050 the rising soil salinization will influence more than 50% of all arable land (Wang et al., 2003) which together with a growing world population may create a need to develop crops that are tolerant to salt stress. Continual management of sustainable water and soil resources and more adequate utilization of genetic diversity are the basic steps that are necessary to increase productivity (McCouch et al., 2012; Shao et al., 2005, 2009).

Building of the knowledge on the biology, ecology and evolutionary patterns of the halophytes and glycophytes may support the efficient development of salt resistant crops. The selective pressure of human in breeding with parameters of resistance and reproductive security was used for domestication evolution of crop plants. Unexpected consequences of this selection process are the loss of tolerance to different stresses and erosion of crop genetic diversity. Therefore, the exploiting the genetic variability of crop plants and their wild relatives, discovering the genetic structures of currently evolved halophytes, endemic halophytes and 'minor' crops are extremely important. The combination of the classical empirical approaches with the modern phenomics tools used for assessment of plant genetic resources can contribute to stabilization of plant biodiversity and creation salt-tolerant crop plants (Cheeseman, 2015).

Understanding the mechanism of stress tolerance and stress signaling network in adaptive reaction is crucial for the advancement of this field. Salinity stress response is multigenic because a number of processes implicated in the tolerance mechanism. The growth decline is listed as a major morphological change under salinity effect. Damaging effects of salts on growth can be connected with the toxicity of specific ions, increase in alkalinity or elevation of osmotic pressure, which may inhibit the water availability or affect metabolic and other cellular pathways. During the primary phases of salinity stress, capacity of root systems to absorb water decreases. The leaf water loss is increased due to osmotic stress of high salt accumulation in soil and plants. Thus, salinity stress

is also called as hyperosmotic stress (Munns, 2005) and also considered as a hyper-ionic stress. One of the most negative effects of salinity stress is the accumulation of Na⁺ and Cl⁻ ions in plant tissues under effects of high NaCl concentrations. Excess uptake of both Na⁺ and Cl⁻ ions into the cells cause firstly serious ion imbalance and then significant physiological disbalance. High concentration of Na⁺ can suppress the uptake of essential elements, such as K⁺, resulting in lower productivity and may lead to death (James et al., 2011). The necrosis, chlorosis and tip burn can appear in the leaves when salinity affects imbalances in concentrations of other mineral elements, like Ca, Na, Cl, B. This explains the fact that the phenotypic effects of salt stress are very variable.

For practical reasons, in addition to mechanistic, molecular and genetic studies of different plant genotypes, the scientific focus was actually switched also on phenotypic traits. In the previous years, the fast development of phenomics approach was documented by an increasing number of works employing the modern technical tools in plant assessment and the remote sensing aimed at wetland and salinity stress species. Recently, there are numerous well-established or emerging techniques useful for detection of salinity stress effects at the plant or canopy level. Discussing the techniques and methodologies, it is important to identify the aspects, priorities and challenges that still need to be explored (Adam et al., 2010). In the last few years, a high progress has been achieved in the field of highly automated, non-destructive plant phenotyping systems. The automated phenomics facilities enable an extensive evaluation of complex plant features such as architecture, growth, development, physiology, resistance, tolerance, ecology, and yield, as well as the analysis of particular quantitative parameters that provide the background for more complex features.

An important step towards the more complex phenotyping has been done by involvement hyperspectral imaging (HSI) and subsequent analyses (Kuska et al., 2015). This technique has been successfully used in remote sensing applications to estimate the level of salinity in soils, using numerous indices to assess the concentration of salt according to different wavelengths of reflectance (Poss et al., 2006; Hamzeh et al., 2013). It has been used also in the remote sensing assessment of salt stress effects in many different crops and plants: sugarcane (Hamzeh et al., 2013), reed, cogon grass, cotton, saltcedar, corn, suaeda or aeluropus (Zhang et al., 2011a, 2011b). However, the more precise imaging and analyses of reflectance spectra at the plant level are required to provide desired information useful in phenomics systems. The focus of this review is put on the possible role of novel non-destructive emerging techniques in exploring the salt stress effects on crop performance, physiology as well as the biodiversity of cropping systems. Because of the novelty and usefulness in detections, the emphasis will be placed on hyperspectral imaging techniques. The specific features and applications of this technique in connection with other analytical tools and methods will be discussed.

1. Integration of imaging techniques in high throughput phenotyping systems

The technical progress in data caption, storage and analysis as well as commercial availability of the complex imaging systems led to a fast development stage of high-throughput plant phenotyping. Recently, the plant phenotyping is based preferentially on automated imaging techniques that can create many different images of plants per day. Such a volume of information is sufficient to perform the QTL analyses,

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