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YIELD ATTRIBUTES AND BIOCHEMICAL CONSTITUENTS OF MEDICINAL PLANT SENNA (*CASSIA ANGUSTIFOLIA* VAHL.) AS AFFECTED BY SALINITY

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ABSTRACT

An pot experiment was conducted under natural conditions of screen house to estimate various yield parameters and biochemical constituents under the influence of chloride and sulphate dominated salinity in Senna (*Cassia angustifolia* Vahl.). In this experiment, plants were raised by sowing seeds in dune sand filled polythene bags, at varying EC levels viz. 0, 4, 8 and 12 dSm^{-1} of Cl^- dominated and SO_4^{2-} dominated salinity along with nutrients. Results revealed a significant delay in the flower initiation and pod maturity was observed by increase of salinity and relatively more delay in both under sulphate dominated salinity. Build up of salinity irrespective of salinity type, in the growing medium was found deleterious to various reproductive yield attributes such as number of pods, pod dry weight, number of seeds, seed yield per plant and 100-seed weight. Relatively higher reductions were noticed under SO_4^{2-} dominated salinity. Significant accumulation of both total soluble carbohydrates and proline was recorded with the increase of salinity but relatively higher accumulation of proline was observed under SO_4^{2-} dominated salinity. The results demonstrated that in spite of better osmotic adjustment under sulphate dominated salinity treatments, the sulphate ions were more deleterious to the plants as compared to chloride ions.

INTRODUCTION

Abiotic stresses; drought, high and low temperatures, heavy metals and salinity are exercising drastic effects on crop biochemical processes, plant growth and development which result in low agricultural productivity and food insecurity (Kokani *et al.,* 2014). On a world scale, no toxic substance restricts plant growth more than does salt. Salt stress presents an increasing threat to plant agriculture (Rani and Sharma, 2015). Osmotic stress induced by conditions like drought, salinity and heat etc. impairs plant growth and development by affecting plant physiological processes (Nagar *et al.*, 2015). Despite a number of advances in biotechnology and crop improvement techniques for pests and diseases, salt stress remained elusive due to the fact that it is a complicated biochemical and physiological phenomenon (Tavakkoli *et al.,* 2011). Total global area of salt affected soil including saline and sodic soil is 831 million hectare (Martinez-Beltran and Manzur, 2005). In India about 12 million hectare of land is affected by salinity and/or alkalinity. In the state of Haryana, 0.63 million hectare of land having this problem. Conductivity is a very important parameter for determining the water quality for drinking as well as agricultural purposes. Conductivity based on total concentration of various ions (Deshmukh and Urkude, 2014). Salt stress creates both ionic as well as osmotic stress on plants. Also, ionic toxicity generated from salt contaminated soil has negative effects on plant growth and development (Munns *et al.*, 2006). However, there are many defense mechanisms in plants which are tolerant to water deficit and salt stresses, such as osmoregulation, ion homeostasis, antioxidant and hormonal systems (Mahajan and Tuteja, 2005), helping plants to survive and grow under severe environmental conditions prior to their reproductive stages. Osmotic adjustment, i.e. reduction of osmotic potential due to net solute accumulation, is an important mechanism of salt tolerance in plants (Sairam and Tyagi, 2004). The reduction in osmotic potential in salt-stressed plants may stem from accumulation of inorganic ions (Na^+, Cl^-, Ca^{2+} and K^{+}) and compatible solutes (soluble carbohydrates, amino acids, proline) (Kavi Kishore *et al.*, 2005). Biochemical and physiological parameters in higher plants cultivated in salt or water-deficit conditions have been developed as effective indices for tolerant screening in plant breeding programs (Ashraf and Fooland, 2007). Salinity adversely affects the process of germination, seedling establishment, plant growth and yield in almost all the cultivated crops by lowering the osmotic potential of water in the growing medium or by causing specific ionic toxicity or both. Na^+, Cl^-, SO_4^{2-}, CO_3^{2-} and HCO_3^{2-} are the potential ions in saline habitats that are inimical to plants. Carbohydrate metabolism in plants is found to be affected by salinity as well as by the type of salt ions contributing to the salinity (Polonenko *et al.,* 1983). Accumulation of proline and other free amino acids under stress have long been regarded as an adaptive phenomenon to salt and water stress (Hellebust, 1976). Sugars have been correlated with the salt tolerance in many plants (Rathert, 1983). *Cassia angustifolia* Vahl. commonly known as Indian senna in English and Senai in Hindi is an important medicinal plant species belonging to the family caesalpiniaceae. The leaves and pods of this species produce crude drug senna. The leaves and pods of *Cassia angustifolia* are cathartic, contains sennosides A, B, C, D, rhein and aloe-emodin in free and compound form of these sennosides. Sennosides A and B are present in large amount than others. The leaves and leaflets also contain koempferol, anthraquinone, essential oil, calcium salt and isorhamnetin. It is used as expectorant, wound dresser, carminative and laxative. It is also useful in loss of apetite, hepatomegaly, spleenomegaly, indigestion, malaria, skin diseases, jaundice and anaemia. There is growing global demand for medicinal plants. With increasing demand of food for an everincreasing population in India, it is not possible to bring lands under cultivation for aromatic and medicinal plants. Salt affected lands due to high soluble salt content, excessive exchangeable Na^+ and poor physical properties are not suitable for raising conventional food crops. So, marginal and salt lands could be exploited for the cultivation of medicinal plants such as Senna. The present experiment was planned to study yield attributes and biochemical changes in Senna in response to salt stress.

MATERIALS AND METHODS

The experimental site was in the screen house, Department of Botany and plant physiology, Chaudhary Charan Singh Haryana Agricultural University, Hisar, Haryana. Seeds of Senna var. sona for these experiments were obtained from the Institute of Herbal Heritage (A unit of Asian Medicinal plants and Health care trust) Sonamukhi Nagar, Sangaria Fanta, Salawas Road, Jodhpur-342005 (Rajasthan), INDIA.

Culture conditions

The plants were raised in polythene bags (18" × 15"), each containing 12 kg of dune sand. The sand filled polythene bags were saturated with the solution of respective salinity treatment along with the nutrient (Hogland and Arnon, 1950) before sowing. Two types of salinity i.e. chloride (Cl^-:SO_4^{2-} (7:3); Na^+:$Ca^{2+}+Mg^{2+}$ (1:1); Ca^{2+}:Mg^{2+} (1:3)) and sulphate (SO_4^{2-}:Cl^- (7:3); Na^+:$Ca^{2+}+Mg^{2+}$ (1:1); Ca^{2+}:Mg^{2+} (1:3) dominated salinity with three replication was given at 4 different salinity level such as 0 (control), 4, 8 and 12 dSm^{-1}. 15 seeds of Senna were sown in February 2010 on variously treated sand beds in polythene bags. The moisture in the bags was maintained at field capacity by adding water as and when required. After establishment of seedlings thinning was done to maintain 3 plants of uniform size in each bag. Number of days taken by the plants to initiate flowering in each treatment were recorded through constant observation. Days to pod maturity recorded, when the pods became almost 50% brown. Number of pods formed per plant till pod maturity were counted and recorded. Number of seeds per pod were counted and recorded. Available number of seeds was air-dried for 72 h and their combine weight was determined and expressed on 100-seed weight basis. Total soluble carbohydrate, Proline content and Chlorophyll contents was observed in fourth leaf at vegetative stage. The total soluble carbohydrate (mg g^{-1}) was estimated by the methods of Yemms and Willis (1954) using anthrone reagent. Proline content (mg g^{-1}) was estimated spectrophotometrically according to Bates *et al.* (1973) using Acid ninhydrin and sulphosalicylic acid. Factorial CRD design was used for statistical analysis. Significance was tested at 5% level of critical differences (CD).

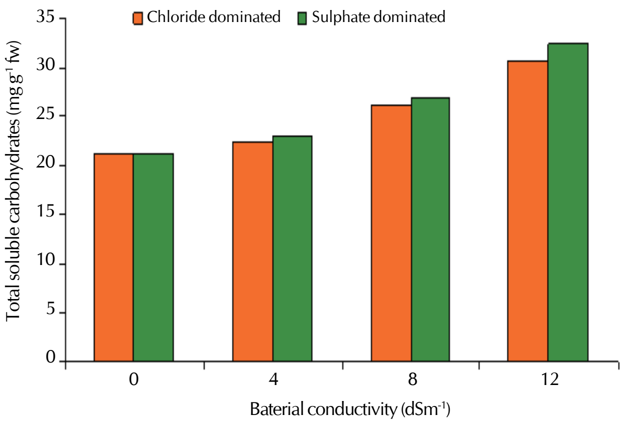
RESULTS AND DISCUSSION

A significant delay in days to flower initiation and pod maturity was recorded with the progressive increase of salinity in the growing medium. No significant effect was observed at 8 dSm^{-1} EC level in number of days to pod maturity. Sulphate dominated salinity caused more significant delay in days to flower initiation than chloride dominated salinity. The two salinity types, however, were indifferent in influencing the days to pod maturity. Reports on deferred flower initiation in the medicinal plant german chamomile (Deepika, 2015) and in an ornamental *Calceoria hybrida* (Fornes *et al*., 2007) are in consonance with the present findings. Preponement or delay in flowering and fruiting has been observed in some crop plants due to salinity by Flower *et al*. (1988).

Salinity has been known to affect the economic yield of crop plants through its adverse effect on vegetative as well as reproductive growth i.e. yield attributes. Seed yield is affected by the number of pods and number of seeds per pod. The mass of individual seed might also be reduced where plant growth is reduced. Pod number is a function of the number of flowers, successful fertilization and photosynthate available for seed filling. Reproductive yield attributes of Senna were recorded at pod maturity. The number of pods per plant, pod dry weight per plant, number of seeds per pod, 100-seed weight and seed yield per plant were adversely affected with the increasing EC levels in the growing medium. The two salinity types did not differ in their response on these yield attributes indicating that the reduction in various yield parameters were due to osmotic effects and not because of ionic toxicity.

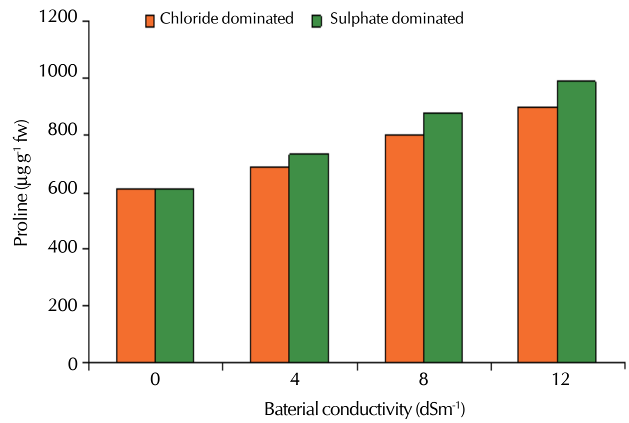
Table 1: Yield attributes (reproductive yield) of Senna under varying chloride and sulphate dominated salinity

Parameters	Salinity Types (ST) Chloride dominated salinity EC levels (dSm^{-1})					Sulphate dominated salinity					CD at 5%	
	0	4	8	12	Mean	0	4	8	12	Mean		
Days to flower initiation	69.7	75.3	84.0	92.7	80.4	69.7	79.3	89.3	95.7	83.5	ST = 2.2EC = 3.1	ST*EC= N.S.
Days to pods maturity	141.7	146.3	146.7	148.7	145.8	141.7	143.7	145.3	149.0	144.9	ST = N.S. EC = 2.3	ST×EC= N.S.
Number of pods /plant	9.78	8.11	8.00	7.45	8.33	9.78	7.89	7.56	7.11	8.08	ST = N.S. EC = 0.618	ST×EC = N.S.
Pod dry wt/plant (g)	1.588	1.405	1.214	1.089	1.324	1.588	1.285	1.172	0.991	1.259	ST = N.S.EC = 0.114	ST×EC = N.S.
Number of seeds/pod	4.80	4.13	4.08	3.40	4.10	4.80	3.87	3.76	3.33	3.94	ST = N.S EC = 0.47	ST×EC = N.S.
100-seed weight (g)	2.435	2.346	2.266	2.193	2.310	2.435	2.290	2.218	2.155	2.275	ST = N.S.EC = 0.062	ST×EC = N.S.
Seed yield/plant	0.726	0.648	0.515	0.422	0.578	0.726	0.602	0.498	0.414	0.516	ST = N.S.EC = 0.053	ST×EC = N.S



CD at 5%, ST = N.S.; EC = 1.14; ST×EC = N.S.

Figure 1: Total soluble carbohydrates (mg g^-1 fresh weight) content of leaves of Senna at vegetative stage under salinity



CD at 5%, ST=23.6, EC=33.4, ST×EC=47.1

Figure 2: Proline (µg g^-1 fresh weight) content of leaves of Senna at vegetative stage under salinity

Reduction in seed yield with the increase of EC levels was due to decrease in the number of pods per plant, number of seeds per pod and reduced 100-seed weight. Similar results on various yield attributes were reported chickpea by Singh (2014), in *calendula officinalis* by Kusum, (2013) and in isabgol by Surajkala (2014). Salinity has been reported to reduce final yield in other leguminous crops also *viz*. Chickpea (Asha and Dhingra, 2007), pigeon pea (Joshi and Nimbalkar, 1983). Infact, reduction in seed number in pods occur at the very beginning of fruit development due to failure of fertilization or abortion of fertilized ovule due to competition for assimilate supply. Thus, reproduction seems to be a sensitive development stage for Senna exposed to salinity.

In the present experimental study significant accumulation of TSC content was observed in leaves at the vegetative stage with increasing EC level of the growing medium irrespective of salt type. The accumulation of TSC helped to bring down the osmotic potential of cell sap below that of growing medium, enabling the uptake of water by cells under salt stress. The two salinity type remained indifferent in influencing the TSC content. Relatively higher accumulation of TSC in leaves of German chamomile (Deepika *et al.*, 2015), however, was found under sulphate dominated salinity as compared to chloride dominated salinity. Contrasting results were obtained by Nehru (2003) in isabgol and Singh (2004) in chandrashura where more accumulation of TSC was occurred under chloride salt stress. The accumulation of TSC has been shown to be associated with the hydrolysis of starch content (Karadge and chavan, 1983). The accumulation of total soluble carbohydrates under salt stress has been reported in several plant species such as barley (Kukreja *et al.*, 2010), isabgol (Surajkala, 2014), and german chamomile (Deepika *et al*. 2015). The results also revealed accumulation of proline content of fourth leaves (from the top of the plant) at the vegetative stage under the influence of salinity. Relatively more accumulation of proline was found under sulphate dominated salinity than chloride dominated salinity. The increase in proline content was observed right from 4 dSm^{-1} EC level under both salinity types and the differential response of the two salinity types was more pronounced at higher EC levels. These results corroborate the finding of Deepika *et al*. (2015) in German chamomile. Contrastingly, Nehru (2003) reported more accumulation of proline under chloride salt stress as compared to sulphate salt stress in isabgol genotypes. Similar results were obtained by Singh (2004) also in chandrashura. Increase of protein degradation may contribute to proline accumulation during salt stress. Cytoplasmic accumulation of this amino acid is thought to be involved in osmotic adjustment of stressed tissue (Roy *et al.*, 2003). Enhancement of proline content under salt stress was also observed in senna (Arshi *et al.*, 2004), pepper (Chookhampaeng, 2011), lemon grass (Sapna, 2011), durum wheat (Kahrizi *et al.*, 2012), *Calendula officinalis* (Kusum, 2013) and chickpea (Singh and Dhingra, 2014). The accumulation of proline under stress has long been considered as an adaptive mechanism to both salt and water stress (Hellebust, 1976).

Proline has been observed to lower the generation of free radical and lipid peroxidation linked membrane deterioration under salt stress. Its accumulation also protects the thylakoid membrane against photodamage in *Brassica juncea* (Alia *et al.*, 1995). In addition to acting as osmoprotectant, proline also serve as a sink for energy it regulate redox potentials, a hydroxyl radical scavenger, as a solute that protects macromolecules against denaturation, as means of reducing the acidity in the cell and acts as storage compound and nitrogen source for rapid growth after stress.

Senna is a highly salt tolerant plant species with varied medicinal values. Plants survived and complete its life cycle even at highest salinity level (12 dSm^{-1}). This plant can therefore be grown in crop field or wastelands affected by salts.

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