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Growth response and antioxidant enzyme activity of *Cassia occidentalis* exposed to soil moisture stress

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ABSTRACT

Biotic and abiotic stresses exert a considerable influence on the growth and physiology of plants. Water stress is one of the most important abiotic stress factors. The study was carried out to elucidate the effect of drought stress on growth, physiology and antioxidant enzyme activity of *Cassia occidentalis* experiencing four water regimes with soil relative water content ranging from 65% to 12%. Plants were grown in pots and treated with three irrigation levels (100ml, 200ml and 300ml).The experiment was laid out in complete randomized design (CRD) with three replicates. Data on growth, physiological parameters and antioxidant enzyme activity were recorded and analyzed statistically. Physiological parameters like leaf area index, biomass accumulation, relative water content and membrane stability index decreased significantly with decrease in water level, but mild level of water treatment enhanced the growth. The plant exhibited an increase in antioxidant enzyme activity under stressed condition which proves its tolerance against drought. Thus it can be concluded that *Cassia occidentalis* plant is able to tolerate a limited water levels by enhancing its antioxidant defense but excessive drought markedly affected the enzyme activity and so depleted the plant growth.

Key words : Relative water content, biomass, membrane stability index, leaf area index, antioxidant enzymes, stress.

INTRODUCTION

Coffee senna (*Cassia occidentalis*, family Fabaceae) is a potent medicinal herb. It is used as natural medicine in the tropical and subtropical regions throughout the world. Its roots are considered as diuretic, seeds are brewed into a coffee like beverage for asthma and flower infusion is used for bronchitis. Its crushed leaves are anti-inflammatory. The *Cassia* plants are well known for a group of chemicals called anthraquinons. Due to this group of chemicals, the plant is widely used as purgative and laxative (Hennebelle *et. al.*, 2009).

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Drought or water deficit stress is the major environmental factor that has negative impact on agricultural yield (Parida and Dagaonkar, 2007). Drought is known to adversely affect plant height and weight (Lahlou and Ledent 2005) reduces leaf size (Jefferies and MacKerron 1987), photosynthesis and dry matter accumulation (Deblonde *et al.*1999). Drought impacts include growth, yield, membrane integrity, pigment content, osmotic adjustment water relations, and photosynthetic activity (Praba *et al.*, 2009).

The susceptibility of plants to drought stress varies in dependence of stress degree, different accompanying stress factors, plant species, and their developmental stages. Water stress induces generation of active species, such as superoxide radical, hydrogen peroxides and hydroxyl radical causing lipid peroxidation and consequently membrane injury, protein degradation, enzyme inactivation, pigment bleaching and disruption of DNA strands (Arora *et al* ; 2002).

Survival under stressful environment is, however, made possible because of the inherent capability of plants to upregulate the antioxidant system, providing a mechanism for efficient detoxification of the ROS (Asada, 2006).

Plants of Fabaceae family are generally tolerant to drought stress. Although, vast and comprehensive research were done about drought stress on crop plants, behavior of medicinal plant in field conditions were not studied. The present study evaluates the effect of drought stress on various physiological parameters viz. relative water content, biomass accumulation, membrane stability index and leaf area index of Indian senna.

MATERIALS AND METHODS

Surface sterilized mature seeds [with 0.1% $HgCl_2$ for one minute (Ramakrishna *et. al.*, 1991)] of *Cassia occidentalis* were treated with Conc. H_2SO_4 for 40 minutes in order to break the dormancy due to hard lignified seed coat (Srivastava *et al*., 2012).

Seeds were sown in earthenware pots containing garden soil. After 15 days of sowing, plants were irrigated with 100ml, 200ml and 300ml water per pot daily. One set was treated as control and was irrigated regularly with 600-650 ml water. The soil relative water content (SRWC, the ratio of current soil water content to water content at field capacity) for four levels of water treatments was: 12% (Severe stress), 24% (Moderate stress), 36% (Mild stress) and 60-65% (Control) respectively.

Leaf samples were collected at all growth stages and enzymatic parameters were studied at every 10 days starting from 25 days after sowing (DAS). Membrane stability index, relative water content and leaf area index were calculated by the method of Deshmukh *et. al.*(1991), Schonfeld *et. al*. (1988) and Singh (1970) respectively. Biomass was determined by measuring the dry weight of plant. Antioxidant enzymes viz., catalase, peroxidase and SOD were studied by the methods of Chance and Maehly (1955), Shannon *et. al.* (1966) and Giannopolites and Ries (1977) respectively.

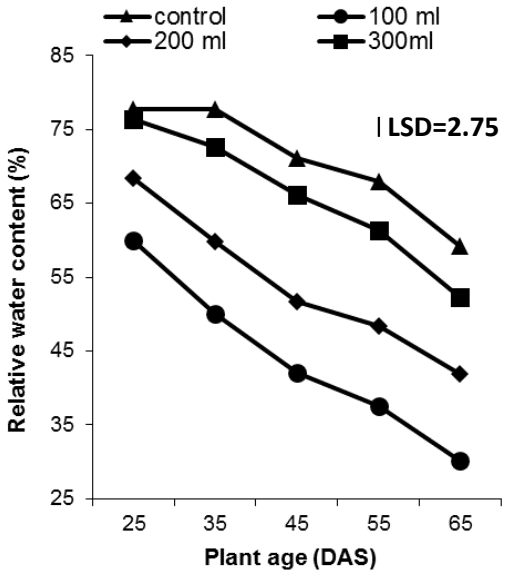
RESULTS AND DISCUSSION

Cassia occidentalis plants subjected to three water regimes (sever, moderate and mild) showed a significant variations in all growth and enzymatic parameters studied.

Effect of water stress on Relative water content (RWC):

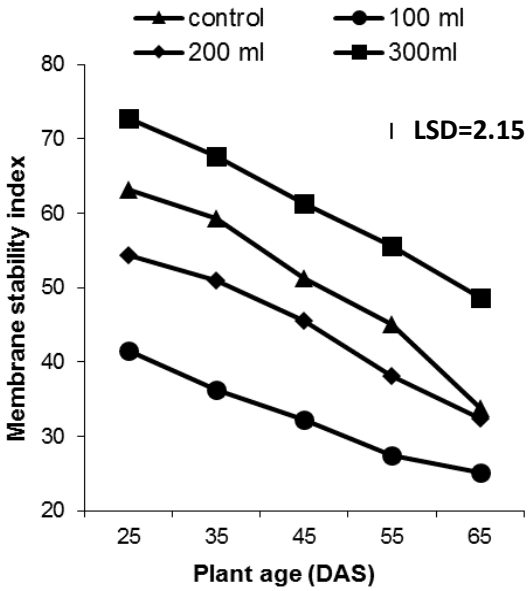
Water stress reduced RWC of plants treated with all three levels of water (Fig-1). Sever stress significantly lowered the RWC of the plant at every stage of plant life. However mild and moderate stress had a minor effect on RWC ($P<0.05$). The result is in accordance with the result of Saraswathi and Paliwal (2011) who studied the effect of drought on *Cassia siamia*.

Figure-1. *Cassia occidentalis:* Relative water content at different age of plant growth under different regimes of water



It is suggested that higher relative water content under mild and moderate stress might help the plant to perform physio-biochemical processes more efficiently under stress condition.

Figure-2. *Cassia occidentalis*: Membrane stability index at different age of plant growth under different regimes of water



Effect of water stress on Membrane stability index (MSI):

Membranes are the main loci affected under water stress condition. Plant showed a steep reduction in membrane stability index (MSI) when treated with sever (100ml) water stress. Moderate stress also showed a minor reduction in MSI but mild stress slightly enhanced the MSI of the plant (P<0.05) (Fig-2).

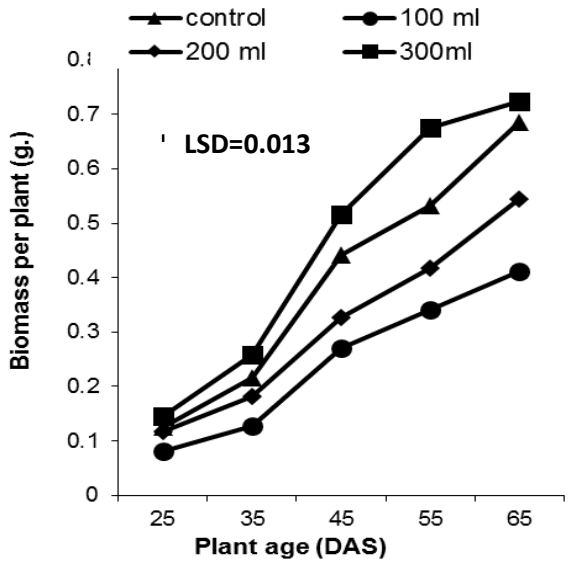
The lower membrane stability index under severe stress condition reflects the extent of lipid peroxidation, which in turn is a consequence of higher oxidative stress due to water stress conditions. Moussa and Moustafa (2008) also reported similar findings in maize.

Effect of water stress on Biomass:

Drought stress impaired physiological functions and biomass production. Sever and moderate stress markedly reduced the biomass accumulation under different moisture regimes. However mild water treatment increased it slightly (P<0.05) (Fig. 3). Xu and Zhou (2005) reported slight increase in MSI and biomass in *Leymus chinensis* plants treated with different water stress condition.

Low availability of moisture around the roots, restricted proliferation of root biomass or limited absorption of nutrients (Staniszewska *et al*., 2003) are the factors that can be responsible for the reduction of biomass. The reduction in dry weight can be due to reduced protein synthesis (Kaya *et al.*, 2002). Mild stressed plants showed a slight increase in biomass accumulation which may be due to increased water use efficiency as reported by Saraswathi and Paliwal (2008) in woody plants.

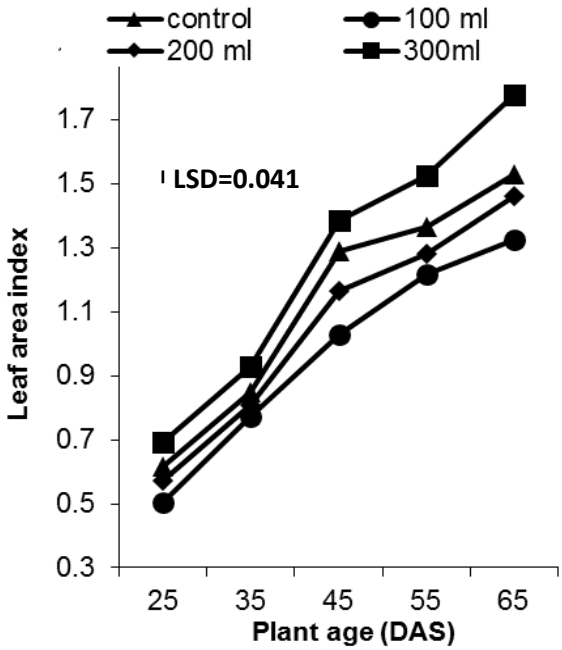
Figure-3. *Cassia occidentalis:* Biomass at different age of plant growth under different regimes of water



Effect of water stress on Leaf Area Index (LAI):

Water stress significantly reduced leaf area of sever and moderately stressed plants. However mild stress slightly enhanced leaf area index of *Cassia* plants (P<0.05) (Fig. 4). The decrease in leaf area index might be due to lesser number of leaves and or decrease area of leaf. The reduction in biomass and LAI in severe stressed plants can also be related with oxidative damage of proteins, DNA and lipids due to massive formation of reactive oxygen species (ROS) like oxygen ions, free radicals and peroxides (Apel and Hart, 2004).

Figure-4. *Cassia occidentalis:* Leaf area index at different age of plant growth under different regimes of water



Effect of water stress on Antioxidant enzymes:

SOD constitutes the first line of defense via detoxification of superoxide radicals (Sairam *et al*., 2000). The activity of SOD in drought treated plants was significantly higher than that of the control (P<0.05). Moderately stressed plants showed maximum SOD activity followed by mild and severely stressed ones (Fig.5).

Peroxidase, an important H_2O_2 scavenging enzyme, followed the similar trend. The present study indicated that the activity of POD was significantly higher in mild, moderate and severe stressed plants than that of control plants (P<0.05) (Fig.6). Enhancement in POX activity under various stress conditions has been linked with protection from oxidative damage, lignifications and cross-linking of cell wall to prevent from such adverse conditions (Dalal and Khanna-Chopra, 2001).

Figure-5. *Cassia occidentalis:* Superoxide dismutase activity at different age of plant growth under different regimes of water

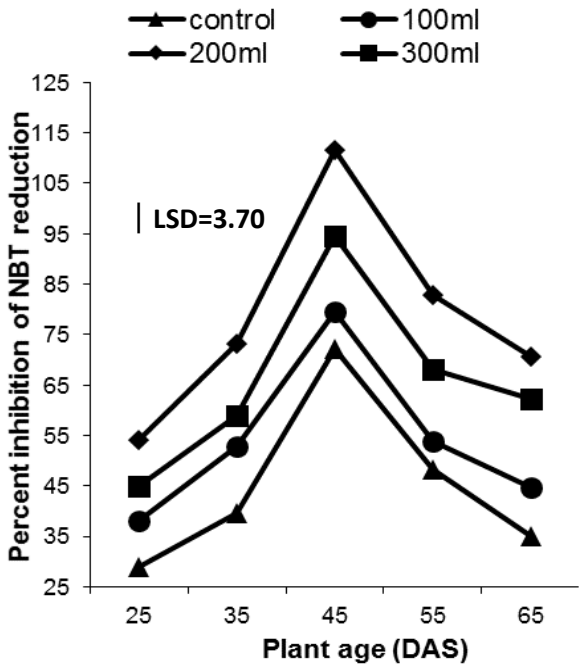
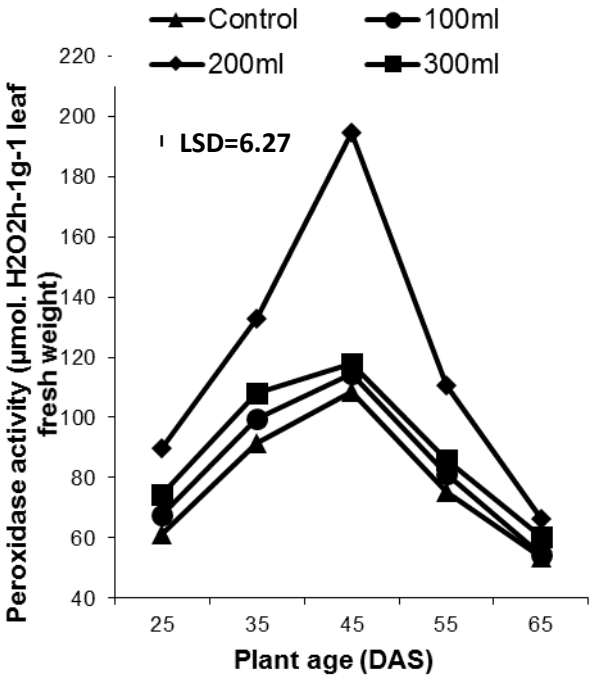
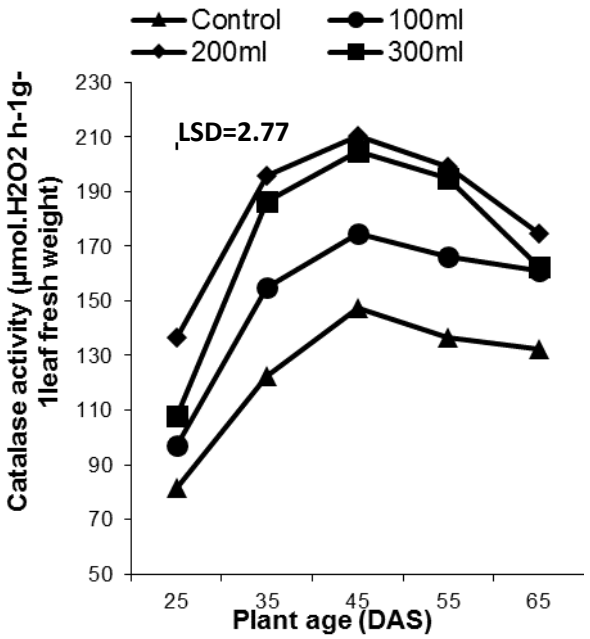


Figure-6. *Cassia occidentalis:* Peroxidase activity at different age of plant growth under different regimes of water



Catalase activity increased significantly under water stress conditions at all the three regimes of water ($P<0.05$) (Fig.7). The moderate drought stressed plants had the highest CAT activity all through the growth period.

Figure-7. *Cassia occidentalis:* Catalase activity at different age of plant growth under different regimes of water



The activity pattern of above three enzymes under different drought stress levels was significantly higher than that of the control, which indicated that *Cassia* plants were able to increase the metabolism of $O_2^{\cdot -}$ by enhancing the activity of antioxidant enzymes and reducing the rate of membrane lipid peroxidation, to resist the effects imposed by the external environment. Cunhua *et. al.*, (2010) and Paritha bhanu and M. Deepak (2015) in pigweed and Khammari *et. al*., (2012) in Indian Senna also found the same pattern of antioxidant activity under drought stress.

CONCLUSION

From the above study we can conclude that *Cassia occidentalis* has a high tolerance against water stress. Water stress enhanced the growth and antioxidant defense in *Cassia occidentalis* under moderate and mild level of water regimes. However excessive drought stress markedly affect physiological pattern of *Cassia occidentalis*. So this is suggested that *Cassia occidentalis* have the ability to cope with stress and can be cultivated in areas prone to drought. *Cassia* is a potent medicinal plant and with the help of suitable water level, we can enhance its growth and yield.

Conflict of Interests

The authors declare that there is no conflict of interests regarding the publication of this paper.

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