RESEARCH ARTICLE

Drought Induced Biochemical Changes in *Commiphora wightii*

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ABSTRACT- The effects of water deficit induced by withholding water in soil pots were examined on the activities of various key enzymes i.e. nitrate reductase, peroxidase, acid phosphatase, α-amylase and invertase in *Commiphora wightii*. Drought stress induced decrease in the activities of nitrate reductase, peroxidase, α-amylase and invertase was observed in leaves of *C. wightii*. The decreased activity of peroxidase enzyme in *C. wightii* plants under water stress condition indicates that the plant is capable of maintaining growth vigour despite adverse conditions. On the other hand, acid phosphatase activity increased continuously in the leaves of *C. wightii* plants subjected to water stress. The results clearly indicate that regulation of enzymatic activity under drought is an essential biochemical process, which prevents the plants from drought-induced damage.

Key-words- Drought, *Commiphora wightii*, Nitrate reductase, Peroxidase, Acid phosphatase, α-amylase, Invertase

INTRODUCTION

Drought is one of the important biomass-limiting stress factors which affect practically every aspect of plant growth and metabolism [1]. Plant's responses to water deficit condition depend upon various factors such as duration and degree of stress, growth stage and time of stress exposure. During drought the water potential (ψ), relative water content (RWC) and net photosynthetic CO_2 fixation (A) significantly decrease [2]. The stomatal closure is among the first responses to the water stress, and is assumed to be the main cause of impaired photosynthesis, since the stomatal closure limits CO_2 availability to the mesophyll. On the other hand, the limitation of CO_2 fixation during drought is also influenced by the diffusion of CO_2 from the intercellular spaces to chloroplasts [3], and by other metabolic factors such as changes in the activity of ribulose-1,5-bisphosphate-carbosilase and perturbed regeneration of ribulose-1,5-bisphosphate etc [3].

C. wightii (Arnott) Bhandari (Burseraceae) is a slow growing, highly branched, and critically endangered plant that grows wild in the arid and semi-arid regions of India, Pakistan and tropical regions of Africa.

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In India, the plant is widespread in the dry and arid region of Rajasthan and Gujarat states. Plants grown in such arid and semi-arid regions often encounter various stress conditions such as high temperature, high wind regime, high light intensity, drought etc. Against drought, plants adapt themselves by different mechanisms, including change in morphological and developmental pattern as well as physiological and biochemical responses [4]. Adaptation to drought is associated with metabolic adjustments that lead to the modulation of different enzymes. It has been hypothesized that these particular changes induced under water deficit conditions enable the plant to endure drought stress. Therefore, in the present instigations, efforts were carried out to understand the biochemical basis of drought tolerance in *C. wightii* plants.

MATERIALS AND METHODS

Plant materials and Drought treatment- Two year old vegetatively propagated plants of *C. wightii* (Fig. 1 A) growing in pots at research nursery, Department of Botany, University of Rajasthan, Jaipur, India during March 2005 was used for the biochemical studies. The plants were divided into two sets (each of four plants), out of which one set was subjected to water stress by withholding of water supply till wilting symptoms appeared, while the second set was watered regularly and served as a control.

Sample preparation- Biochemical analyses of enzymes were carried out in the leaves as the leaves were devoid of resinous compounds, which interfere with biochemical

estimations. The samples were collected regularly at an interval of three days till day 15. The leaves were homogenized in pre-chilled mortar and pestle using appropriate buffers. The homogenate was centrifuged in high-speed centrifuge (KR 20000 T, KUBUTA, Japan) at 10,000 rpm for 20 minutes. The supernatant thus collected was used for enzyme assay and protein estimation. The results were averages of two replicates.

Enzyme assay- A method given by Jaworski [5] was used for assaying nitrate reductase activity. Guaiacol H_2O_2 method was used for assaying the activity of peroxidase [6]. The activity of acid phosphatase was assayed by using p-nitrophenyl phosphate as a substrate [7] and Bernfeld's method [8] was used for assaying the activity of α- amylase. A modified method of Harris and Jeffcoat [9] was used to determine the activity of acid invertase.

RESULTS AND DISCUSSION

Metabolic changes in response to water stress also include reduction in photosynthetic activity and accumulation of organic acids, such as malate, citrate and lactate accompanied by accumulation of proline, sugars and betaine [10,11]. Exposure of plants to low water potential often leads to loss of cell turgor and then plants undergo osmotic adjustments by the rapid accumulation of abscisic acid (ABA) and osmoprotectants [12]. In the present investigation, the activities of enzymes (nitrate reductase, peroxidase, acid phosphatase, α-amylase and invertase) and level of cellular metabolites (proteins and proline) were analyzed in *C. wightii* plants subjected to water deficit condition.

Nitrate reductase, which is involved in the reduction of NO^-_3 to NO^-_2, is known to be highly sensitive to all types of stresses, particularly salt and drought stresses. In the present study, a remarkable decline in the nitrate reductase activity was observed after 3rd day of water deprivation (Fig. 1 B). On day 15 of water deprivation, the activity of nitrate reductase enzyme decreased to 15.6 % (±2.02) of initial level. Similarly Inhibition of nitrate reductase under moisture stress has been reported by several workers in clusterbean [13], rice [14], corn [15], wheat [16], and Sorghum [17].

Peroxidase activity decreased continuously in the leaves of *C. wightii* plants subjected to water deficit conditions (Fig. 1 C). On day 15 the activity of this enzyme decreased to 30.7% (±1.421) of initial level. The variation in peroxidase activity in the plants growing under normal irrigated condition was insignificant. The decrease in activity is ascribed to as a change in the structural proteins [18]. In general, the peroxidase activity is inversely correlated with the plant growth rates. The decreased activity of peroxidase enzyme in plants undergoes water stress condition indicates that the plant is capable of maintaining growth vigour despite adverse conditions.

Acid phosphatase is known to maintain a certain level of inorganic phosphate in plant cells under stress [19]. In the present study, acid phosphatase activity increased continuously in the leaves of C. wightii plants subjected to water stress (Fig. 1 D). Induction of acid phosphatase activity may be due to fact that under conditions of water stress, growth is restricted and delivery of phosphate is impaired, resulting in the activation of the cellular phosphatases that release soluble phosphate from its insoluble compounds inside or outside the cells [20]. Olmos and Hellin [21] observed that acid phosphatase under salt and water stress maintained a certain level of inorganic phosphate, which could be co-transported with H+ along a gradient of proton motive force.

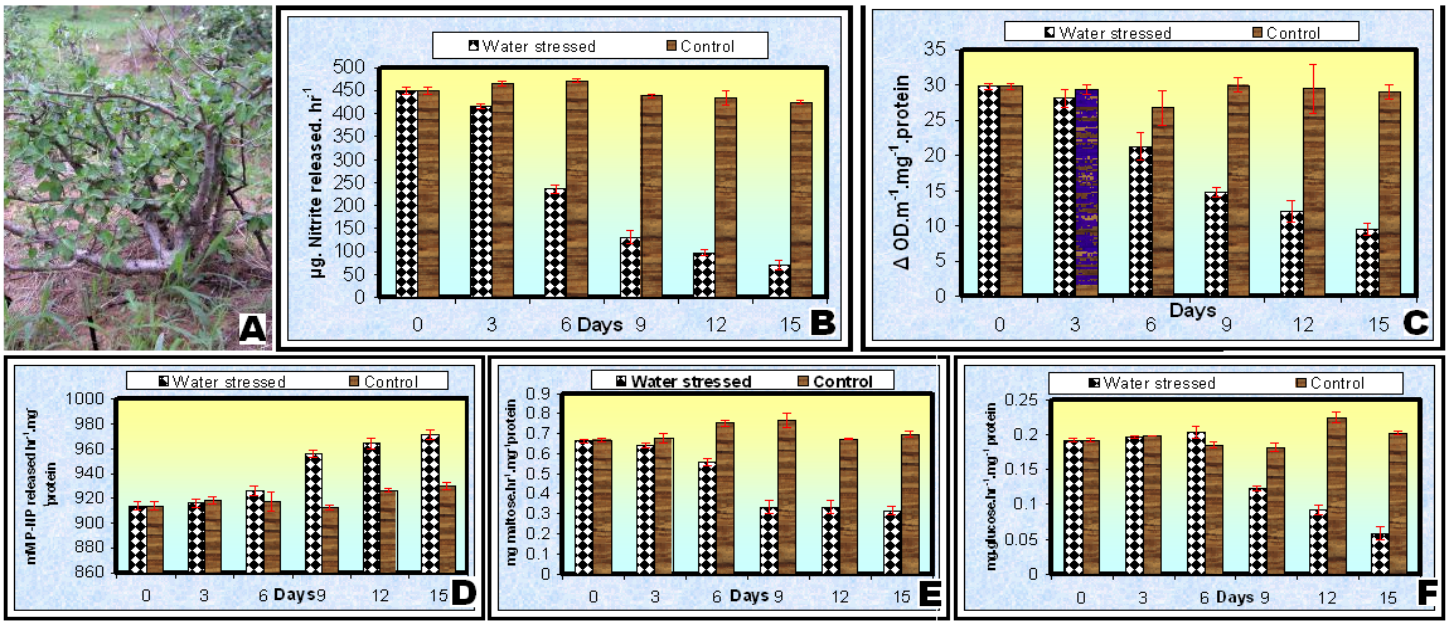

Fig. 1: (A) Mature plant of *C. wightii*; Drought induced changes in nitrate reductase, (B) Peroxidase,

(C) Acid phosphatase, (D) α-amylase, (E) Invertase, (F) *C. wightii* under drought stress

Activity of α-amylase declined marginally till day 6 and declined sharply thereafter in the guggul plants subjected to water stress. The α-amylase activity in non-stressed plants increased marginally during first nine days (Fig. 1 E). Similarly results were also observed in pearl millet [22]. Invertase activity initially increased till day 6 and then declined sharply with an increasing water starvation (Fig. 1 F). The initial increase in invertase activity in plants indicates its involvement in water stress tolerance mechanism. Thind and Malik [23] also observed an increase in acid invertase activity in wheat at low stress levels.

CONCLUSIONS

In the present investigations, biochemical basis of drought tolerance in the succulent plant *C. wightii* was studied. Biochemical analysis indicates that plants are quite resistant and well developed to endure drought stress by modulating the key enzymes such as enzymes nitrate reductase, peroxidase, acid phosphatase, α-amylase and invertase. The study will be highly beneficial in the development of transgenic plants against drought stress.

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