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Effect of Abscisic Acid on Drought Tolerance of Selected Plant Species

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ABSTRACT

Abscisic acid is a phytohormone produced by plants and has various roles in plants growth and development. ABA function in plants is still not well understood, despite of its involvement in abscission. However, ABA has a critical role in reducing a variety of stress reactions. Drought stress is one of the abiotic stress and is defined as a prolonged period of reduced water supply. ABA controls plants response to various conditions such as salinity, low temperature, and drought. It plays a vital role in regulating a variety of processes involved in plant growth, such as floral induction, seed dormancy, embryo maturation, seed germination and root growth. The present research work is carried out to analyse the drought tolerance and leaf chlorosis by the application of ABA on selected three medicinal plant species *Talinum paniculatum, Bryophyllum pinnatum* and *Sauropus androgynus.*

Key words : *Phytohormone, Abscisic acid, Wilting, Drought*, *Chlorosis*

Introduction

The primary phytohormone that increases crop plants ability to withstand abiotic stress is Abscisic acid (Shinozaki and Yamaguchi-Shinozaki, 2000; Schroeder *et al*., 2001). Drought, excessive heat, and high salinity are examples of stress conditions that cause increased content of Abscisic acid (ABA) in plants significantly, creating stress-tolerance effects that assist plants to adapt and survive in demanding environmental situations (Ng *et al*., 2014). ABA has a critical role in reducing a variety of stress reactions, hence it is called as stress hormone. Responses to ABA can be both long-lasting and very quick. Evidence suggests that some responses of ABA are received on the plasma membrane's outer surface, whereas other responses appear to perceive inside the cell (Zeevaart, 2003).

Biosynthesis of ABA occurs under drought-stress situations, and is generated during protein biosynthesis. ABA biosynthesis occurs from β-carotene via Zeaxanthin, Neoxanthin, Xanthoxin, and ABA-aldehyde (C40 pathway). Under drought stress, formation of Xanthoin from Neoxanthin is rate-limiting in C40 pathway. Biochemical studies and genetic evidences support the C40 pathway and the Neoxanthin cleavage is an important step in ABA production (Shinozaki, 2003).

Adverse environmental factors, such as pathogen infections and insect herbivores, have a significant impact on plant growth, productivity, and food security (Daryanto *et al.* 2016; Farooq *et al.* 2017). Examples of these factors are drought, alkalinity, temperatures, nutrient shortages, and heavy metals.

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Drought stress is one of the abiotic stress and is defined as a prolonged period of reduced water supply. Stress signals are received by plant cells, which is the first step taken by the plant to overcome the stress. These signals are linked to genes and cellular signalling pathways, which in turn regulate molecular and physio-morphological responses (Zhu, 2016). The first step in the drought-stress signalling pathways is the detection of stress by transmembrane histidine kinase, a plasma membrane receptor protein. This histidine kinase domain-containing protein (AtHKT1) stimulates the signals to induce gene expression in *Arabidopsis thaliana* (Kuromori *et al.,* 2014). When intracellular calcium levels are disturbed, a phosphorylation cascade is triggered by the calcium sensors. Calcium (Ca^{2+}) plays an important factor in the signalling pathways which allows many plant species to adapt to changes in their surroundings (Shahid *et al.,* 2018). The stimuli produced by a rise in intracellular Ca^{2+} concentration are perceived by plants cellular organelles such as the nucleus, cytosol, mitochondria, chloroplast etc., and the intracellular Ca^{2+} concentration rises either immediately or after a delay. Calmodulins (CaMs), Calcineurin B-like proteins (CBLs), Calmodulin-like proteins (CMLs), and Calcium-Dependent Protein Kinases (CDPKs/CPKs) are calcium detecting sensors can detect rising of cellular transient Ca^{2+}. This also targets subsequent processes like protein phosphorylation and gene expression (Reddy and Reddy, 2004; Day *et al.,* 2002).

Lack of precipitation during drought results in water shortages in the soil, which is referred to as drought stress and lack of water in the soil causes the level of groundwater to drop, which may be detrimental to plant growth and survival. Disturbance of numerous metabolic processes, including drought stress,carbon absorption, and exchange of gases, enhanced oxidative damage and turgor pressure may have detrimental consequences on plants.Ion balance, enzymatic activity, stem extension, leaf size, and root proliferation are some other metabolic functions that may be affected. The result of all the aforementioned losses is brought on by stress is a reduced yield (Chowdhury *et al.,* 2016; Hussain *et al.* 2018). Plant species have signalling pathways that allow for optimal growth and development but they don't have defined immune system. As central integrators, plant hormones for instance, are responsible for developmental pathways and adaptive stress processes. It plays a vital role in regulating a variety of processes involved in plant growth, such as floral induction,seed dormancy, embryo maturation, seed germination and root growth. Additionally, ABA minimises the consequences of drought stress on plants. Plants can maintain physiological functions, which also helps them adapt to a constantly changing environment. Stomatal closer and opening is controlled by altering ABA-responsive genes expression thereby controlling other physiological processes of plants (Zhu, 2002; Dong *et al*., 2015). The present work main objective is to study the effect of ABA on the physiological functions like drought and leaf chlorosis in selected three medicinal plants species *Talinum paniculatum, Bryophyllum pinnatum* and *Sauropus androgynus*.

Materials and Methods

Three plant species were selected from the Herbal Medicinal garden of Department of Biosciences and Sericulture, Sri Padmavati Mahila Visvavidyalayam (Women's University), Tirupati, *Talinum paniculatum*, *Bryophyllum pinnatum* and *Sauropus androgynus*.

Talinum paniculatum (Jacq.) Gaertn.

T. paniculatum (Jacq.) Gaertn. is a succulent subshrub belongs to the family Talinaceae. Commonly known as fameflower. Its leaves are edible and used to treat diseases such as diuretic, healing, emollient and anti-infective. It is also eaten in salads (Fig. 1).

Bryophyllum pinnatum (Lam.) Oken

It is a succulent, perennial plant that reach a height of about 1 m, has thick cylindrical stems, with a reddish tinge (Fig. 2). It is commonly referred to as cathedral bells, air plant, life plant, miracle leaf, and Goethe plant and is a member of the Crassulaceae family. Leaf juice is additionally used to treat kidney stones in conventional medicine. Additionally, it treats cancer, inflammation, and headache (Hermann, 1983).

Sauropus androgynus (L.) Merr.

S. androgynus, also known as katuk, star gooseberry, or sweet leaf, is a shrub grown as a leaf vegetable in some tropical regions that belongs to Euphorbiaceae family. Stems can grow upto 2.5 metres in height with 5-6 cm long dark green oval leaves. It is one of the most popular leafy vegetables in South and Southeast Asia, and it is known for its high yields and flavour (Padmavathi and Prabhakara, 1990). The leaves are used as a medicine for cough, to soothe the lungs, as a tonic, for weight loss, used as vegetable and as a febrifugal to relieve internal fever (Fig. 3). The plant has high levels of vitamin K, vitamins B and C, protein, minerals, and provitamin A carotenoids, but can cause lung damage due to high concentrations of the alkaloid papaverine (Kao *et al.*, 1999).

The selected three species differ in succulence nature of the leaf. *B. pinnatum* is more succulent than *T. paniculatum* and *S. androgynus.* To study the drought tolerance in the selected plants, ABA is sprayed at a concentration of 25 mg/l (30 ml) in three replications (Waterland *et al.* 2010). The potted Plants of one year are selected and are maintained in greenhouse environment and they were irrigated 12 hrs before the ABA is sprayed. Control plants were watered daily and experimental plants were withheld after ABA is sprayed until the plants showed symptoms of wilt. When plants were observed with wilt symptoms the wilted plants were rewatered daily to observe if the stressed plants were recovered from their wilting. And daily visual observations were noted, on an average of three replications was used to analyse the data. During the observation of wilting the other visible physiological changes are also noted. Leaf chlorosis is the visible physiological change which is observed and the data is collected and analysed.

Wilt observation and Leaf chlorosis

Whole plant wilting is observed after the application of ABA. Wilting is graded on a scale of 1 to 5, with 5 representing fully turgid, 4 representing soft to touch, 3 representing the beginning of wilting, 2 representing complete loss of turgor, and 1 representing wilting to the point where leaves are dry and brittle (Waterland *et al.,* 2010).

During the observation of wilting, leaf chlorosis is also analysed and visual observations are noted on a scale of 1 to 11 and the degree of leaf chlorosis is evaluated. With a rating of 11 being completely green with no signs of chlorosis, 10 with 10% or less leaf chlorosis, 9 with 11% to 20%, 8 with 21% to 30%, 7 with 31% to 40%, 6 with 41% to 50%, 5 with 51% to 60%, 4 with 61% to 70%, 3 with 71% to 80%, 2 with 81% to 90% and 1 with 91% to 100% chlorosis (Waterland *et al.,* 2010).

Statistical analysis

The experimental values are the mean values of selected three plant species. Values obtained are taken by visual observations which were analyzed by using ANOVA (One-way analysis) by SPSS (Software Package for Social Sciences) version 21.0.

Results

Effect of ABA is studied based on wilt status by visual observations. The shelf life of all the treated plants was extended from 2 to 5 days on application of ABA at 25 mg/l ($P \leq 0.05$) (Table 1) to drought-stressed plants. Wilting was delayed in ABA sprayed plants at 25 mg/l ($P \leq 0.05$) (Fig. 1) when compared to control plants. It was observed that Control plants wilted earlier than ABA sprayed plants. *B. pinnatum* maintained higher wiltstatus starting on 10th day when compared to the control plants wilting starting on 6th day (Fig. 4a) than *T. paniculatum,* wilting started on 8th day when compared to the control plants, wilting starting on 4th day (Fig. 4b) and *S. androgynus* wilting started on 6th

Table 1. Wilt status in control and ABA sprayed selected three plant species and shelf life extension

Species	Concentration of ABA (mg /L) Time before wilt (Days)		
	0	25 mg / l	Shelflife extension
T. paniculatum	4.5	8.3	3.8
B. pinnatum	6.6	10.6	4
S. androgynus	4.4	6.6	2.2

Values are an average of the number of days from Day 0 to visible symptoms of wilt status rating from 1 to 5. Values are mean of three replications (n=3).

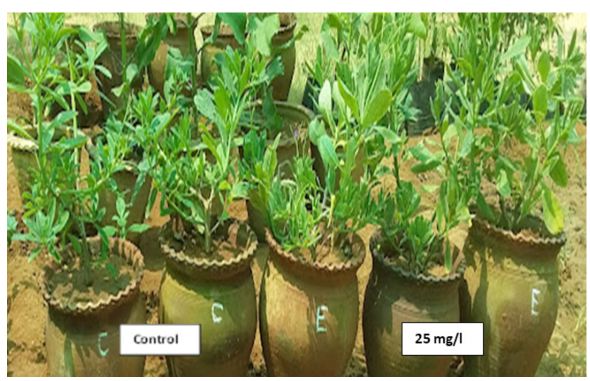


Fig. 1. *Talinum paniculatum*

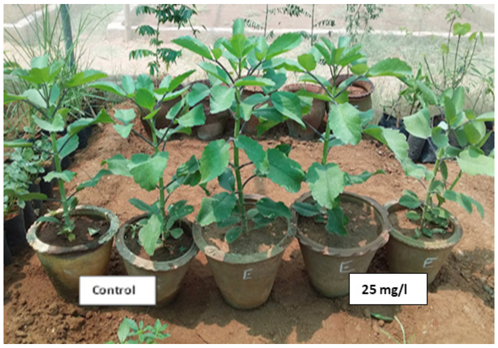


Fig. 2. *Bryophyllum pinnatum*

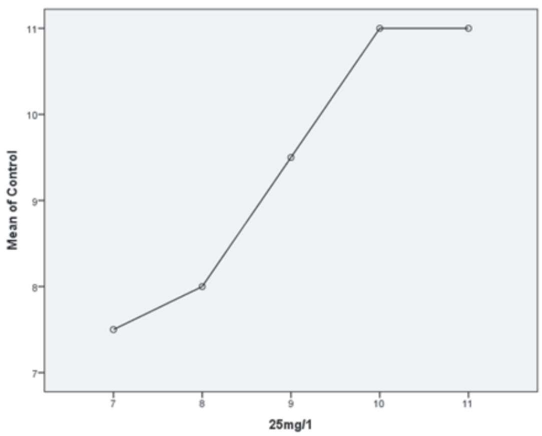
day when compared to the control plants wilting starting on 4th day (Fig 4c).

The Means plot is a visual representation of comparing means output. The means plot shows the average values of a) *T. paniculatum* b) *B. pinnatum* and c) *S. androgynus* data. It analyzes the way in which the mean varies across the different groups of data.

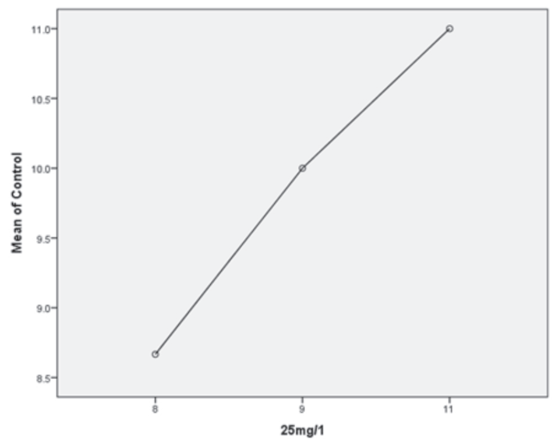
Fig. 3. *Sauropus androgynus*

Leaf chlorosis observation

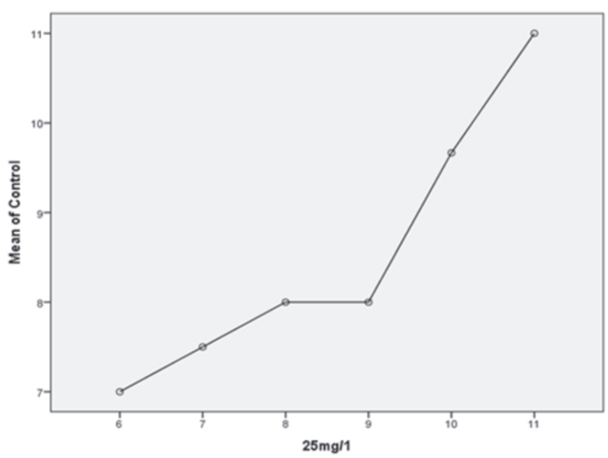
On application of ABA wilting is delayed in all the selected species but visual observations showed leaf chlorosis (yellowing of leaves). In *T. panniculatum*



A) *T. paniculatum*



b) *B. pinnatum*



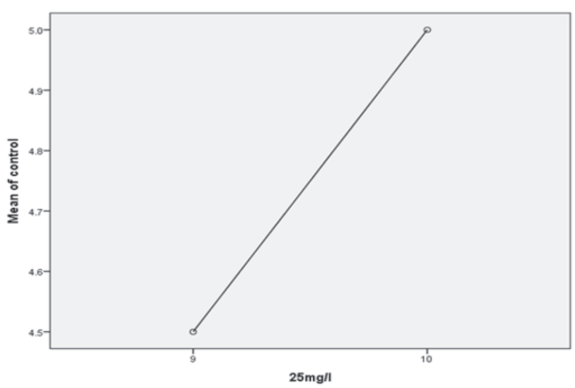
S. androgynus

Fig. 4. Wilt status of three species which are sprayed with 0 to 25 mg/L s-ABA.

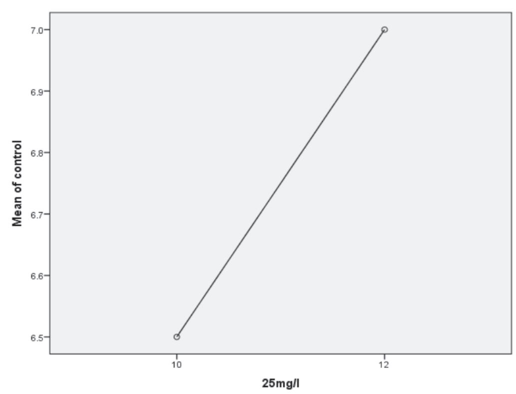
there is no much difference in the control and experimental plants, leaf chlorosis started on 8th day with 21% to 30%($P \leq 0.05$) (Fig. 5a). *B. pinnatum* delayed leaf chlorosis than *T. panniculatum* and chlorosis started on 10th day in ABA applied plants (25 mg/l) ($P \leq 0.05$) than in control plants i.e., on 8th day with 11 to 20 % chlorosis (Fig. 5b). *S. androgynus* showed early chlorosis on 6th day in control plants when compared to experimental plants which showed delayed chlorosis i.e., on 8th day with 31 to 41% Chlorosis (Fig. 5c).

Discussion

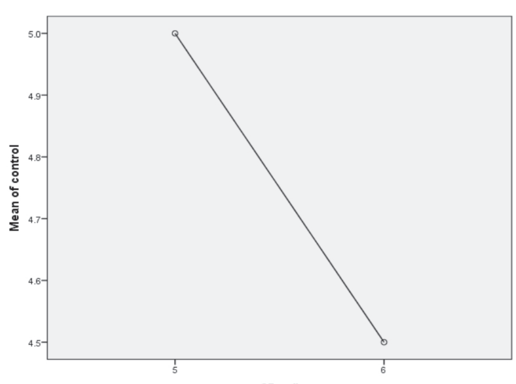
ABA applications reduced water loss in all plants treated during severe drought stress. Due to the decrease in water loss, *T. paniculatum*, *B. pinnatum* and *S. androgynus* were able to withstand brief drought stress. Depending on the species, ABA produced a delay in the onset of the visual wilting symptoms, which led to shelf life extension of 2 to 5 days.ABA treatment delayed wilting in *B. pinnatum* and *T. paniculatum*, the treated plants after rewatering were similar to continuously irrigated plants. It has been demonstrated that the responses to ABA treatments varies from species to species (Blanchard *et al*., 2007; Sharma *et al*., 2006). ABA is more effective when foliar application is done than when compared to the application to the roots (Leskovar and Cantliffe, 1992). Drought stress is first noticed at the roots, where ABA is synthesized and transported through xylem to the leaves. Stomatal closure and reduced water loss in the leaves are caused by free ABA. Recently, it was revealed that plants can also produce ABA in their leaves when there is a severe drought (Malladi and Burns, 2007). ABA application to the leaves result in stomatal closure that is quicker and more efficient than ABA applications to the roots. And the response could differ depending on the species. In the present study application of ABA delayed wilting symptoms in drought induced plants with extended shelf life period from 2 to 5 days. *B.*



a) *T. paniculatum*



b) *B. pinnatum*



c) *S. androgynus*

Fig. 5. Leaf chlorosis analysis of three species of medicinal plants sprayed with 0 to 25 mg/l ABA.

pinnatum is having higher drought tolerance than *T. paniculatum* and *S. androgynus.* Leaf chlorosis is also extended in *B. pinnatum* in ABA treated plants. ABA application to the plants can avoid drought during transport and also can increase the yield.

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