

Study of UVB radiation effect on plant water relations and Electrolyte leakage in medicinally important plant *Simarouba glauca*DC.

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Abstract-

The effects of UVB radiation on medicinally important plant Simarouba glauca leaf water relation as well as electrolyte leakage characteristics were investigated. Seedlings of Simarouba glauca were grown under UVB chamber and experimental results were compared with those are not UVB irradiated as a control. UV-B radiations was artificially supplied by UV-B tubes (Philips TL20 W/16, NV, Holland). The UV-B irradiance was provided for 10h (08:00am-18:00pm). For different days (4, 8, 12 and 16 days). In present study it is observed that RWC was slightly decreased or remains stable but osmotic potential was increased by 10-15%. In electrolyte leakage study it is resulted that slight elevation in membrane leakage in 8 days treated plants from over all study it is noticed that, RWC slightly decreased or remains constant which indicates water relations under UVB stress might be remains stable to maintain optimum balance where as increased osmotic potential under UVB irradiated condition will helps to participate the ions freely in the growth and development process during stress, which is beneficial to survive the S.glauca plants under UVB stress. Where as in electrolyte leakage slight elevation and further recovery which shows recovery of some membrane proteins in response to UVB stress which might be helpful for development of UVB stress tolerance.

Introduction-

Irradiated ultraviolet-B (UV-B) radiation plays a vital role in plant-water relations by signalling changes in constitutive and inducible as a plant defenses. In point of view the use of UV-B radiations as a tool to work as plant shield in practice has gained raising interest. An alternation in the environmental situations might, however, get the UV-B -induced plant responses. While, in some research study plant responses to UV-B which can induces adaptation to alternations in certain abiotic factors.

A minor component of sunlight is UVB wavelengths (280-320 nm) have a major impact on terrestrial ecosystems because of their high energy levels (Mac kenzie, *et al.*, 2003 and Caldwell *et al.*, 2007). On the basis of RWC the physical and chemical properties as well as the components of the chemical potential of water can be determined (Cowan and Miltrope 1968). Osmotic adjustment it divert the solutes from vital processes like protein and cell wall synthesis but itself can not promote growth (Munns 1988). Thus electrolyte leakage is considered as Hallmark of stress response in intact plant cells. Therefore this phenomenon is widely used for determining stress induced injury of plant tissue as well as measure of plant tolerance against stress (Levitt, 1972.; Blum and Ebercom, 1981; Bajji *et al.*, 2002; Lee and Zhu 2010). Electrolyte leakage is a detector of membrane permeability.



Materials and methods-

A. Plant material

Simarouba glauca edible oil tree is commonly planted along wastelands or dry land forest areas by Department of forest in Maharashtra, Karnataka and Andhra Pradesh as well as in agricultural Universities of these states. Freshly harvested seeds of *S. glauca* were purchased from Sri Sri Institute of Agriculture, Bangalore.

B. Methods

1. Supplementary UV-B radiation treatments-

One year old seedlings of *S. glauca* were purchased from social forestry Kagal. Seedlings with plastic bags were kept in polyhouse under minimum and maximum air temperature at 21 to 31°C respectively with relative humidity of air up to 55%.

In early April seedlings were exposed to UV-B treatments. UV-B radiations was artificially supplied by UV-B tubes (Philips TL20 W/16,NV,Holland).The UV-B irradiance was provided for 10h(08:00am-18:00pm)for different days (4,8,12 and 16 days) as per the method described by Lydon *et al.* ,(1986). The tubes were installed 15cm above perpendicular to the seedlings and oriented in an east-west direction. Tubes were wrapped with 13 mm cellulose diacetate (CA) film to remove out UVC radiation shorter than 290 nm. CA paper was changed per week to avoid photo degradation. Control seedlings were exposed to normal day light.

1. Leaf-Water relations-

a. Relative water content (RWC)

Relative water content (RWC) was determined according to the method of Slatyer (1955). The one gram leaf discs were taken from control and treated plants, then transferred to petridish containing 20ml distilled water. They were immersed for three hours in petridish as they become fully turgid and then water saturated leaf discs were removed and weight in turgid condition was recorded, then the discs were allowed to dry in oven at 80 °C for constant weight up to 4 days and dry weight was recorded.

b. Osmotic Potential:

It was determined as per the method described by Janardhan and Krisnamurthy (1975). One gram of fresh leaf material was thoroughly washed, blotted and crushed in mortar with pestle in little amount of distilled water and filtered through a four layered muslin cloth and volume was adjusted to 20 ml with distilled water. The electrical conductivity of this leaf extract was measured on the conductivity meter (ELICO MODEL PE-133). Simultaneously one gram fresh leaf material was kept in an oven at 60° C for drying. From the dry weight of material moisture content was calculated.

2. Electrolyte leakage

Electrolyte leakage or Ion leakage was measured in terms of electrical conductivity (EC) of solution according to method described by Yan *et al.*,(1996). The washed leaf samples were punched with punching machine. The one gram discs were placed in a glass beaker containing 10 ml deionised water. The beaker was kept at 30°C for 3 hours and the conductivity of the solution was measured with the help of conductivity bridge model Elico (M. 82T). The samples were boiled in boiling water bath for 5 minutes and cooled to room temperature.

Result And Discussion-

a) Relative water content-

Effect of UV-B radiation on relative water content in a *S. glauca* is shown in fig. 1. It is noticed from fig. that the RWC is slightly decreased in response to UV-B radiation and this decrease is more significant in 16 days UV-B irradiated plants.

Relative water content (RWC) is the appropriate measure of plant water status in terms of physiological consequences of cellular water deficit (Barr and Weatherly 1962). Thus it also measures water deficit in the leaf. Normal RWC value ranges between 98% in turgid and transpiring leaves to about 40% in gradually drying and desiccated leaves. After a fully turgid leaf the amount of water content is relative to what the plant organ or tissue holds.

b) Osmotic potential –

The effect of UV-B radiation on the osmotic potential in *Simarouba glauca* plant is as shown in fig.2. It is observed from fig. that osmotic potential is more negative with increasing days of UV-B irradiations.

Osmoregulation helps in maintaining turgor and cell volume thus it is generally considered as an important adaptation to environmental stress. Osmotic adjustment usually defined as induction in osmotic pressure of cell sap resulting from several solute molecules per cell rather than from a lower cell volume (Munns 1988). The significance of lower osmotic potential is that, weaker binding forces exerted by the solute due to higher partial molar free energy of water helps to participate ions at a greater freedom (Levitt 1972).

Electrolyte leakage –

The effect of UV-B radiation on the activity of electrolyte leakage in *Simarouba glauca* is shown in fig.3. It is observed from fig. that the electrolyte leakage is slightly increased with increasing days of exposure of UV-B radiation. This increase is 3 to 15% higher over the control.

Electrolyte leakage directly related with plant stresses like UV stress, salinity, pathogen attack, draught, heavy metal etc. From plant cells K⁺ efflux is mainly correlated with electrolyte leakage, which is mediated by plasma membrane cation conductance. An ion imbalance that developed due to intercellular and extracellular osmotic instability under a stress condition is known as membrane permeability. Membrane disintegration accompanied with the chain reactions induced by free radicals (Mazliak 1983). One of the chain reactions is a lipid peroxidation is considered as the main cause of membrane destruction (Merzlyak 1989; Smirnov 1993 and Sairam *et al.*, 2005). In the twice ambient UV-B treatment the electrolyte leakage was significantly increased in *Populus cathayana* but not in *Populus kangdingensis* (Ren *et al.* 2006).

Summary and Conclusion-

After a UV-B radiation stress there are no significant changes are found in RWC in wheat and pea plant (Alexieva *et al.*, 2001). The plant of *Psidium sativum* when irradiated with UV-B light no significant measure changes were found in RWC when irradiated more than 15 days but after 24 days RWC had decreased to up to 70% (Nogues *et al.*, 1998). In the present study the RWC was slightly decreased or remains stable. Thus the water relations under UV-B stress might be remain stable to maintain optimum balance.

In the present investigation the osmotic potential was increased by 10-15% which helps to maintain energetic status of water inside leaf cell as indicated by Slatyer and Taylor (1960). Further this will help to participate the ions freely in the growth and development process during stress. This might be beneficial to survive the *S. glauca* plants under UVB stress.

In the present study 3-15% increase in electrolyte leakage was noticed in *Simarouba glauca* leaves. This slight elevation in membrane leakage in 8 days treated plants was further recovered by 3-5% in 12 and 16 day UV-B irradiated plants. Thus, there might be recovery of some membrane proteins in response to UV-B stress which might be helpful for development of UV-B stress tolerance.

Table: 1. Effect of UV-B radiation on Relative water content of leaves of *S. glauca*.

Treatments	RWC content
Control	65.67
4 Days	64.19 (-2.25)
8 Days	58.59 (-10.78)
12 Days	63.67 (-3.04)
16 Days	57.01 (-13.18)

- Each value is mean of three determinations.
- Values are expressed as percent (%).
- Values in parenthesis indicate percent increase (+) or decrease (-) over the control.

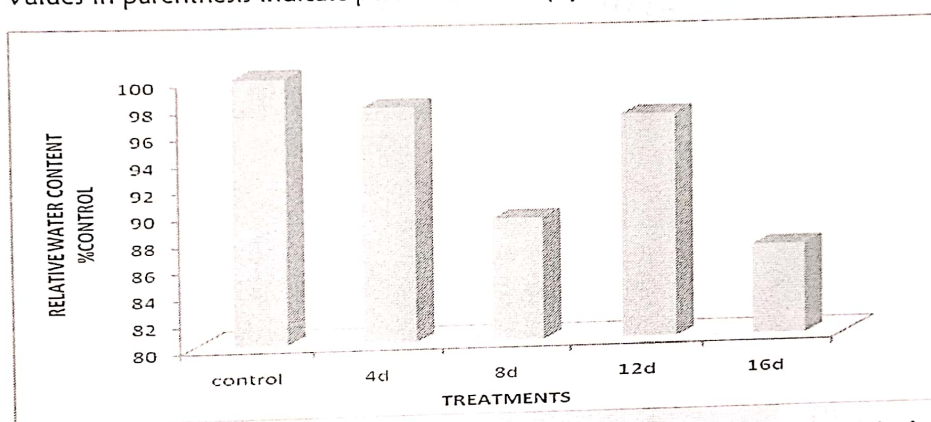


Figure: 1 Effect of UV-B radiation on Relative water content of leaves of *S. glauca*.

Table: 2 Effect of UV-B radiation on osmotic potential of leaves of *S. glauca*.

Treatments	Osmotic potential
Control	1.54
4 Days	1.59 (+3.24)
8 Days	1.69 (+9.74)
12 Days	1.71 (+11.03)
16 Day	1.71 (+11.03)

Each value is mean of three determinations.

Values are expressed as bar.

Values in parenthesis indicate percent increase (+) or decrease (-) over the control.

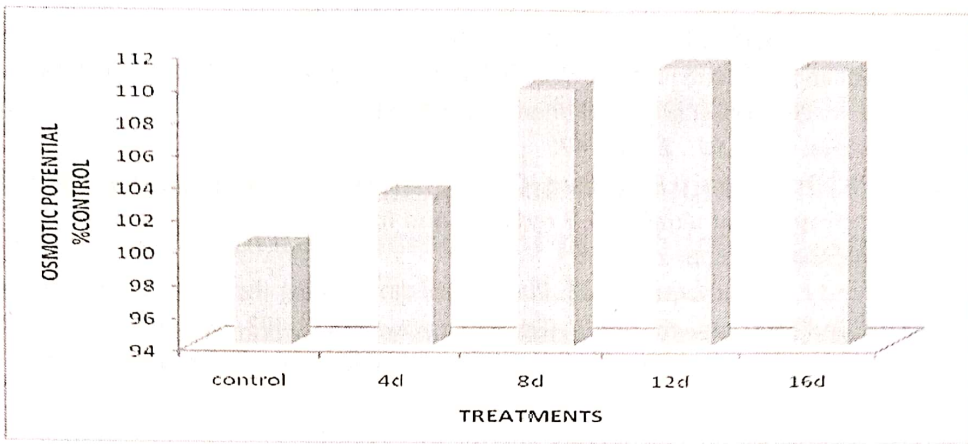


Figure: 2 Effect of UV-B radiations on osmotic potential of leaves of *S. glauca*.

Table: 3 Effect of UVB radiation on electrolyte leakage of leaves of *S. glauca*

Treatments	Electrolyte leakage
Control	23.00
4 Days	25.00 (+8.69)
8 Days	37.00 (+60.86)
12 Days	28.00 (+21.73)
16 Days	33.00 (+43.47)

Each value is mean of three determinations.

Values are expressed as percent (%).

Values in parenthesis indicate percent increase (+) or decrease (-) over the control.

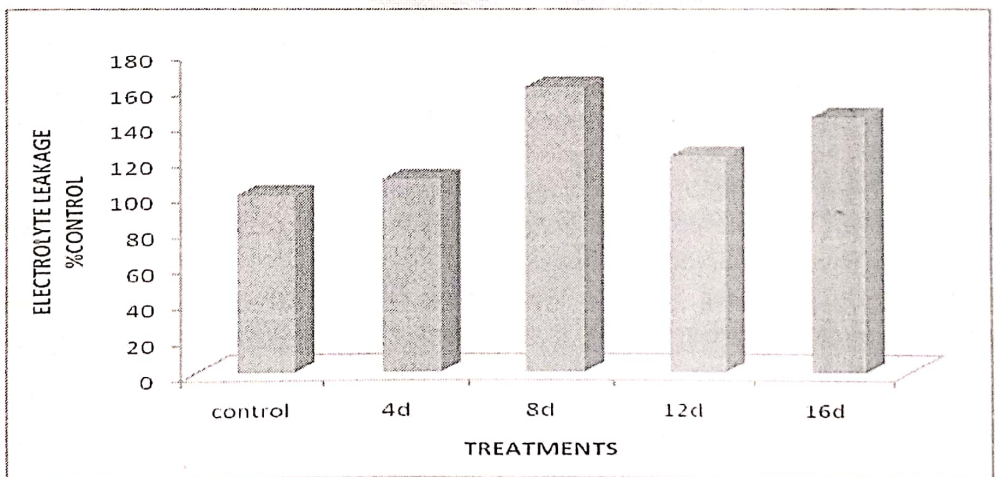


Figure: 3 Effect of UVB radiations on electrolyte leakage of leaves of *S. glauca*.

Bibliography-

1. Alexieva, V., Sergiev I., Mapelli S., and E. Karanov (2001) The effect of drought and ultraviolet radiation on growth and stress markers in pea and wheat. *Plant, Cell & Environment*, 24: 1337-134.

2. Bajji, M., Kinet J.M. and S. Lutts. (2002). Osmotic and ionic effects of NaCl on germination, early seedling growth and ion content of *Atriplex halimus* (Chenopodiaceae). *Can. J. Bot.*, **80**: 297- 304.
3. Blum, A. and A. Ebercon (1981). Cell membrane stability as a measurement of drought and heat tolerance in wheat. *Crop Sci.*, **21** :43-47.
4. Caldwell, M.M., Bornmann, J.F., Ballare, C.L., Flink, S.D., & Kulandaivelu, G., (2007). Terrestrial ecosystem, increased solar ultraviolet-B radiation and interactions with other climate change factors. *Photochem. Photobiol. Sci.*, **6**: 252-266.
5. Cowan, I. R. and F. L.Milthorpe (1968). Plant factors influencing the water status of plant tissues. In 'Water deficits and plant growth. Vol. 1' (Ed. TT Kozlowski) 137-193. (Academic Press: New York).
6. Janardhan, K.V. and V. Krishnamoorthy (1975). A rapid method for determination of osmotic potential of plant cell sap. *Current Sci.*, **44(11)**: 390-391.
7. Lee, B. and J.K.Zhu (2010). phenotypic analysis of Arabidopsis mutants: electrolyte leakage after freezing stress. Cold Spring Harbour Protocols 2010, pdb.prot4970.
8. Levitt, J. (1972). Responses of Plants to Environmental Stresses. Academic Press New York.
9. Lydon, J.; Teramura, A.H. and E.G. Summers (1986). Effects of ultraviolet-B radiation on the growth and productivity of field grown soybean. In Stratospheric Ozone Reduction, Solar Ultraviolet Radiation and Plant Life, R.C. Worrest and M.M. Caldwell 313-325
10. Mac Kenzie, D.I.;Nichols, J.D.; Hines, J.E.; Knutson, M.G. and A.B.Franklin (2003) Estimating site occupancy, colonization and local extinction when a species is detected imperfectly. *Ecology*, **84**: 2200-2207
11. Mazliak, P. (1983). Plant membrane lipids: changes and alterations during aging and senescence. In: Post Harvest Physiology and Crop Preservation, Lieberman M (ed), Plenum Press, New s York, 123-140.
12. Merzlyak, M. N.(1989). Activated oxygen and oxidative processes in plant cell membranes. Itogi Nauki Tekhhiki, Ser. *Plant Physiology* **6**: 1-167.
13. Munns, R. (1988): Why measure osmotic adjustment? *Aust. J. Plant Physiol.*, **15** : 717-726.
14. Nogues, S.; Allen, D. J.; Morison, J.I.L. and N.R.Baker (1998). Ultraviolet-B radiation effects on water relations, leaf development and Photosynthesis in droughted pea plants. *Plant physiol.* **117**: 173-181.
15. Ren, Ji-Jun; Wang, Yan; Sun, Xiu-hua; Wang, Xue; Luo, Xiang-hua and He, Jing-Xin. (2006). Effect of PP (333), CCC and plucking hearts on the growth of French marigold. *Journal of Shenyang Agricultural University*.
16. Sairam, R. K.; Srivastava, G. C.; Agarwal, S. and R. C.Meena (2005). Differences in antioxidant activity in response to salinity stress in tolerant and susceptible wheat genotypes. *Biol. Plant.*, **49**: 85-91.
17. Slatyer, R. O. (1955). Studies on the water relations of crop plants grown under natural rainfall in northern Australia. *Australian Journal of Agricultural Research*, **6**: 365-367
18. Slatyer, R.O. & Taylor, S.A. (1960) Terminology in plant-soil-water relations. *Nature*, **187**:922-924
19. Smirnov N. (1993) The role of active oxygen in the response of plants to water deficit and desiccation. *New Phytol* **125**:27-58
20. Yan, B.; Dai, Q; Liu, X.; Huang, S. and Z. Wang (1996) Flooding induced membrane damage, lipid oxidation and activated oxygen generation in corn leaves. *Plant Soil* **197**:261-268

