

A NOTE ON PROLINE CONTENT IN EUCALYPTUS HYBRID LEAVES

T.C. POKHRIYAL AND A.S. RATURI

Forest Influences Branch,
Forest Research Institute & Colleges, Dehra Dun (India)

Introduction

Tree species growing in the forest usually live under fluctuating water regime i.e., abundant water supply to extreme drought. The extent of growth loss resulting due to water stress is generally not known, because of the difficulty of showing how much more growth would occur if internal water balance of forest trees were retained at favourable level throughout the entire growing season (Kozlowski, 1968).

Several plant species are known to accumulate free amino acids especially proline, when they experience moisture stress conditions (Stewart *et al.* 1966, Barnite and Naylor 1966, Kemble and Macpherson, 1954, Waldern *et al.* 1974, Singh *et al.* 1972) and decline on the release of stress (Singh *et al.* 1973, Sinha and Raj gopal, 1975). The free proline content of the leaves of many species also increased with a decrease in the leaf water potential (Singh *et al.* 1973, Palfi *et al.* 1974).

Singh *et al.* (1972) reported a strong correlation in proline accumulation and high stability index and suggested as a selection criterion for drought resistance in some of the crop plants. It would be rewarding, to examine whether such a correlation also

exists in the tree species like *Eucalyptus* which is controversial about its water consumptive behaviour. An attempt was therefore made to study the changes in the proline contents of different leaflets of *Eucalyptus* i.e., young, developed and mature, under different soil moisture conditions. The present communication deals with the variations in the proline contents among the different leaf blades, so that standard procedure can be developed for the selection of the plant material during the course of sampling in the future programme.

Material and Method

The leaf samples were collected from the *Eucalyptus* hybrid trees from the potculture area of the Forest Influences Branch, F.R.I., Dehra Dun, U.P. The sampling procedure was similar as earlier described by Pokhriyal and Raturi (1984)

The method for the estimation of proline was followed as described by Bates *et al.* (1973). All the estimates were carried out atleast six times and data was statistically analysed. The standard error (\pm) and L.S.D. values were calculated wherever required.

Results

Maximum proline contents

(608.77 $\mu\text{g/g}$ dry wt.) was noticed in the first and minimum (24.8 $\mu\text{g/g}$ fr. wt.) in the tenth leaf blade of *Eucalyptus hybrid*. Proline content decreased with the increase in the age of leaf blades, however, differences only in the upper leaf-blades were found significant. In case of total proline contents per leaf blade, an increase was noticed upto 4th leaf-blade thereafter it decreased in the subsequent leaf-blades. A significantly higher values (29.78 μg of proline contents per leaf blade) was noticed in the 4th leaf blade and lowest (6.85 μg) in the first from the tip (Fig. 1). Fresh and dry weights of individual leaf-blades increased with the growth of the plants but the differences between successive leaves were not found significant in most of the cases (Fig. 2).

Discussion

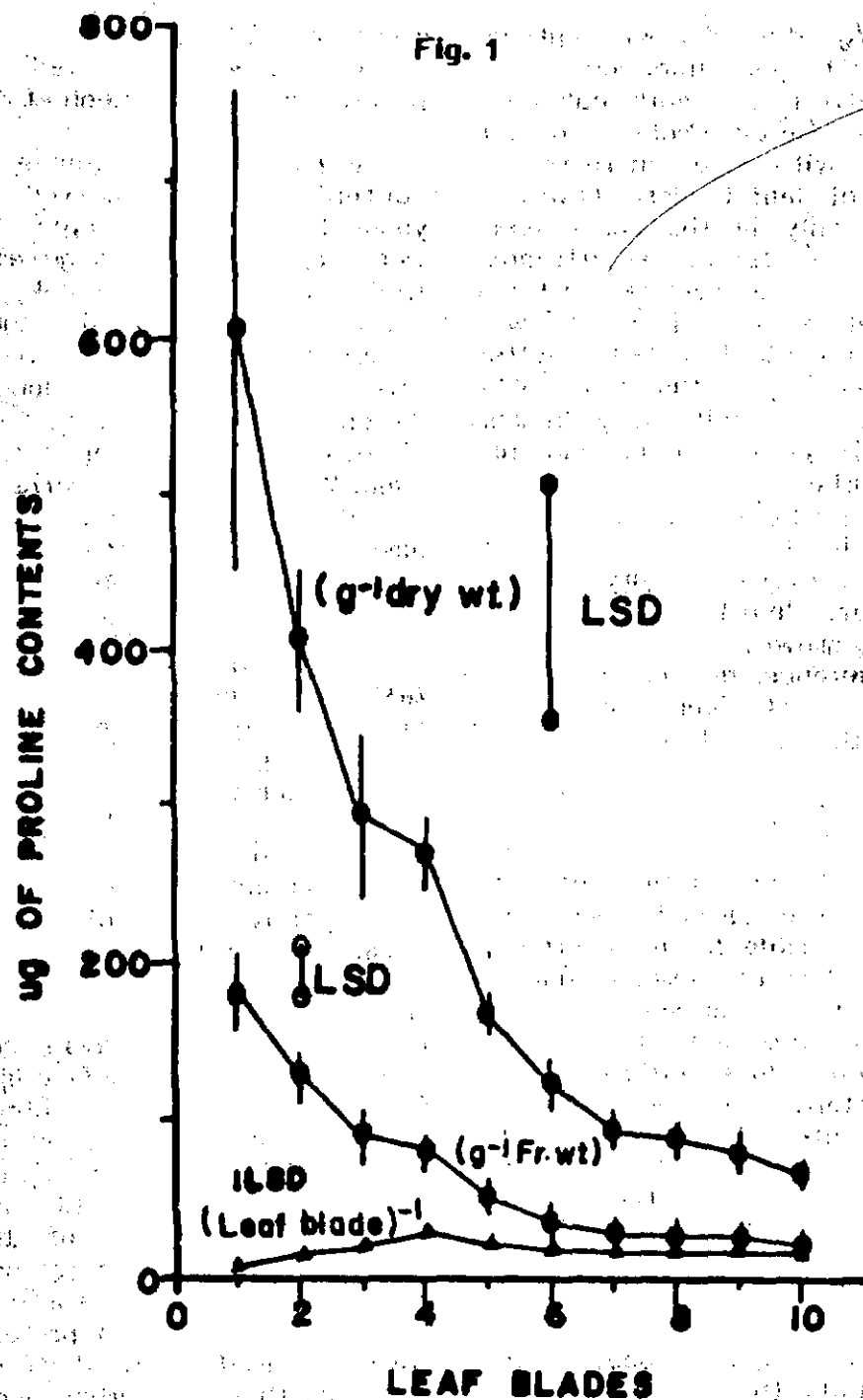
Water deficit influences all phases of tree growth and are probably responsible for more growth losses than all other causes combined (Kramer, 1980). It is very difficult to estimate precisely how much water deficit alone influence the growth patterns because of the confounding effect of water supply with those of other environmental factors i.e., light, temperature and humidity etc. The effect of temperature and drought on the tree growth are closely related. High temperature increased transpiration thereby inducing shoot water deficits and they also amount for depletion of reserve carbohydrates. Whereas, amino acids were continuously synthesised during the water

stressed treatments, but protein synthesis was inhibited and so its level decreased (Singh *et al.*, 1973).

In *Eucalyptus*, maximum proline contents were observed in the young leaves. Similar type of results were reported for orange and grape fruit trees by Yelenosky (1979). The mature leaves of the older plants can also accumulate proline after experiencing a long water deficit period in the field and potculture conditions (Routley, 1966, Waldren *et al.* 1974 and Jones *et al.* 1980) and proline content also varied with the position of leaves in the canopy (Boggs and Stewart, 1976).

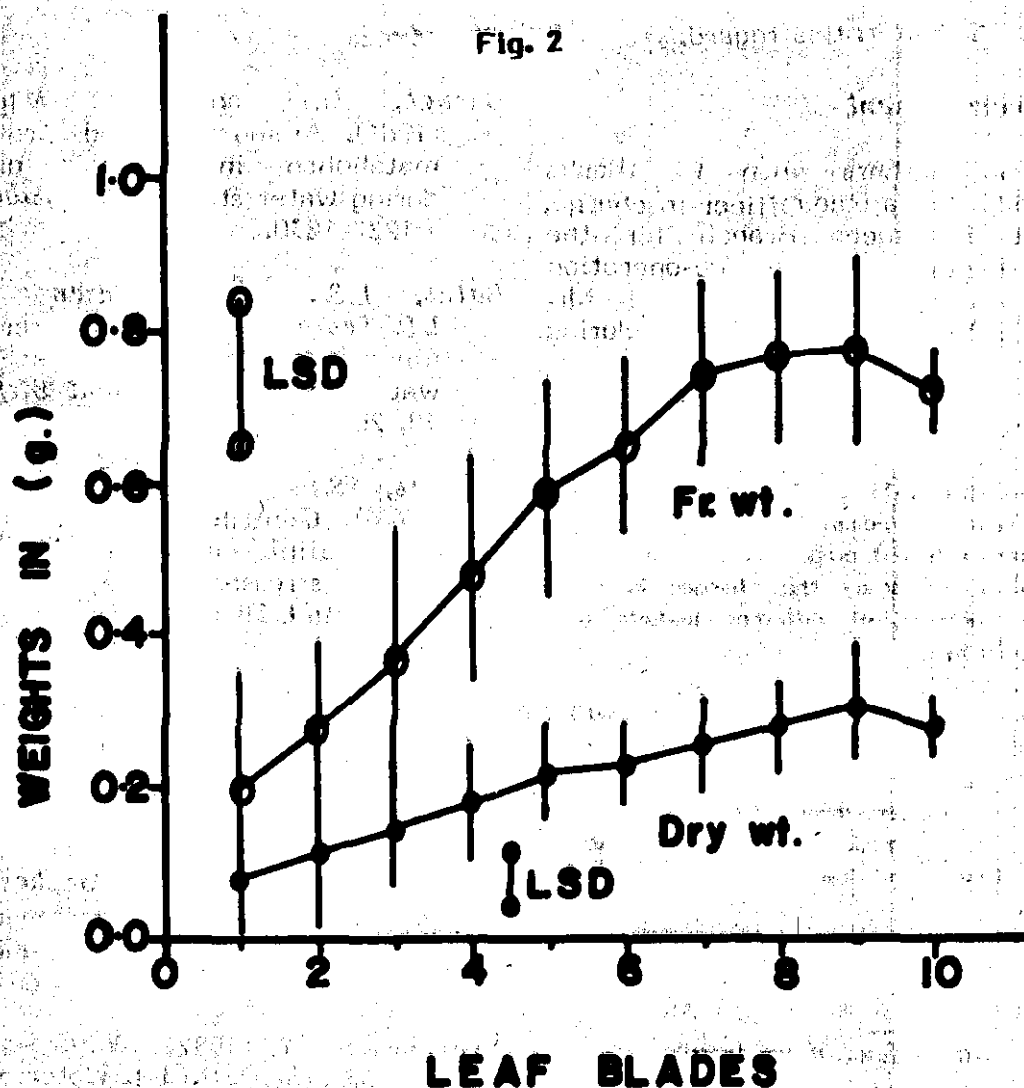
The young leaf blades of *Eucalyptus* synthesised more proline and are also capable of assimilating more NO_3^- as compared to the old ones already reported by Pokhriyal and Raturi (1984). Raj gopal (1981) reported an increase in the nitrate reductase activity by the exogenous application of proline. Whether these changes are related with the water stress or not, is yet to be established in *Eucalyptus*, because most of the available literature is related to only crop plants. During the process of water stress, in some plants proline accumulates because of increased proline synthesis and inhibition of its oxidation. The proline content of the leaf oxidise faster when the carbohydrate content is low. An increased incorporation of proline into protein along with increased carbohydrate contents indicates that the carbon and energy limit protein synthesis (Stewart, 1972). The reason seems to be related with the increase and decrease

Fig. 1



Changes in the Proline Contents g^{-1} fr.wt., g^{-1} dry wt.
and per Leaf blades of *Eucalyptus hybrid*

Fig. 2



Changes in Fresh and Dry weights of different Leaf blades of *Eucalyptus hybrid*

In the weights and proline contents of *Eucalyptus* leaf blades in this case.

The young *Eucalyptus* leaf blades are capable of synthesising more proline as compared to the old

ones. Whether these leaflets can be suitable indicator of water stress symptoms in the fast growing tree species like *Eucalyptus* is yet to be standardized. An elaborate study is required before drawing

any conclusion in this regard.

Acknowledgement

The authors wish to thanks: Shri B.K. Subba Rao, Officer-in-charge, Forest Influences Branch for the encouragement and co-operation and Mr. H.C. Nautiyal and Mr. S.R. Joshi for technical help during the course of this work.

Summary

Accumulation of proline, a free amino acid is a common feature in some of the stress affected crop plants. An attempt was made to study the changes in the proline contents of different leaflets of Eucalyptus hybrid.

Maximum (608.77 µg/g dry wt.) and minimum (24.81 µg/g fr. wt.) proline contents was estimated in the first and tenth leaf blades respectively. But the total proline per leaf blade was highest in the fourth and lowest in the first leaf blade.

संकर युकेलिप्टस की पत्तियों के प्रोलीन
तत्व के विषय में एक टिप्पणी
टी. सी. पोन्नरियाल व ए. एस. रतूड़ी
बारांछा

प्रतिबल प्रभावित फसल-पौधों में एमिनो प्रोलीन का जमाव हो जाना एक सामान्य घटना है। प्रस्तुत अध्ययन में संकर युकेलिप्टस के विभिन्न पर्णकों के प्रोलीन तत्व की पड़ताल की गई है। शुष्क मार अथवा ताजे भार की प्रति इकाई में बंताई प्रोलीन की अधिकतम मात्रा प्रथम पर्णक में तथा न्यूनतम सिरे से बारीक करके दसवें पर्णक तक में पाई गई। इसके विपरीत प्रति पर्णक कुल प्रोलीन मात्रा चौथे पर्णक में अधिकतम और पहले पर्णक में न्यूनतम पाई गई।

References

- Barnet, M.M. and A.W. Naylor (1966). Amino acid and protein metabolism in Bermuda grass during water stress. *Pl. Physiol.* 41 : 1222-1230.
- Bates, L.S., R.P. Waldren and I.D. Teare (1973). Rapid determination of free proline for water stress studies. *Plant & Soil* 39, 205-207.
- Bogges, S.F. and C.R. Stewart (1976). Contribution of arginine to proline accumulation in water stressed barley leaves. *Pl. Physiol.* 58 : 796-97.
- Jones, M.M., C.B.O. Osmand & N.C. Turner (1980). Accumulation of solutes in leaves of sorghum and sunflower in response to water deficits. *Aust. J. Pl. Phys.* 7: 193-205.
- Kemble, A.R. and H.T. Macpherson (1954). Liberation of aminoacids in perennial ryegrass during wilting *Biochem J.* 58: 146-49.
- Kozłowski, T.T. (1982). Water supply & tree growth Part I. Water deficits. *Forestry Abstracts* 43; 57-95.
- Kramer, P.J. (1980). Drought, Stress and the origin of adaptations. In "Adaptations of plants to water and high temperature stress" (N.C. Turner and P.J. Kramer editors) pp 7-20 New York, Wiley.
- Palfi, G., E. Koves, M. Bito and R. Sebestyen (1974). The role

of aminoacids during water-stress in species accumulating proline. *Phyton*, 32 : 121-27.

Pokhriyal, T.C. and A.S. Raturi (1984). Nitrate assimilation in leaf-blades of *Eucalyptus*. *Indian For.* 110 : 202-208.

Rajgopal, U. (1981). The Influences of exogenous proline on stomatal resistance in *Vicia faba*. *Pl. Physiol.* 52 : 292-296.

Routley, D.G. (1966). Proline accumulation in wilted ladino clover leaves. *Crop Sci.* 6 : 358-361.

Singh, T.N., D. Aspinall and L.G. Paleg (1972). Proline accumulation and varietal adaptability to drought in barley: a potential metabolic measure of drought resistance. *Nat. New Biol.* 236: 188-190.

.....(1973). Nitrogen metabolism and growth in the barley plant during water stress. *Aust. J. Biol. Sci.* 26 : 5-56.

Singh, T.N., L.G. Paleg and D. Aspinall (1973). Stress metabolism. I. Nitrogen metabolism and growth in the barley plant during water stress. *Aust. J. Biol. Sci.* 26: 45-56.

Sinha, S.K. & U. Rajgopal (1975). Effect of moisture stress on nitrate reductase activity and accumulation of proline in *Sorghum*. Proc-Sym. on Crop plant responses to Environmental stress, Almora, India. pp. 36-44.

Stewart, C.R., C.L. Morris and J.F. Thompson (1966). Changes in amino acid content of excised leaves during incubation. II. Role of sugar in the accumulation of proline in wilted leaves. *Pl. Physiol.* 41: 585.

Stewart, C.R. (1972). Effects of proline and carbohydrates on the metabolism of Exogenous proline by excised Bean leaves in the dark. *Pl. Physiol.* 50: 551-555.

Waldern, R.P., L.D. Teare and S.W. Ehler (1974). Changes in proline clover concentration in *Sorghum* and Soybean plants under field condition, *Crop-Sci.* 14: 447-50.

Yelenosky, G. (1979). Accumulation of free proline in citrus leaves during cold hardening of young trees in controlled temperature regimes. *Pl. Physiol.* 64: 425-27.