LEAF ANATOMICAL ADAPTATIONS IN SOME EXOTIC SPECIES OF *EUCALYPTUS* L'HÉR. (MYRTACEAE)

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Abstract

Studies were conducted to assess the diversity of anatomical adaptations of leaves in the genus *Eucalyptus* (family Myrtaceae) from the Faisalabad region. Thicker epidermis in *E. alba* Reinw. ex Blume and *E. maculata* Hook., are the adaptations to water limited environments like drought and salinity. Large cortical cell in *E. microtheca* F. Muell., and *E. botryoides* Sm., are the indication of their wide distribution in a variety of different environmental conditions. *Eucalyptus crebra* F. Muell., *E. maculata* Hook., and *E. microtheca* F. Muell., with significantly larger vascular tissue were the better adaptation to a variety of environment types. There was a great variation in leaf anatomical characteristics among *Eucalyptus* species collected from the Faisalabad region.

Introduction

Eucalyptus is a large genus containing more than 700 species, most of them are native to Australia, with a very small number found in adjacent parts of New Guinea and Indonesia and one as far north as the Philippines (Chen & Craven, 2007). It has been introduced in Pakistan, probably from Australia and many species have been found successfully adapted to a variety of habitat types in Pakistan (Ahmad, 1996).

This genus has very specific structural modification in its leaf anatomy. In certain species oil glands were reported in pith and midrib (Carr & Carr, 1969). The degree of ornamentation of waxes on leaf surfaces are frequently correlated with taxonomic groupings of this genus, which also proved useful as indicators of natural groupings of species (Hallam & Chambers, 1970).

Leaf anatomy is quite responsive to climatic conditions, for example *Eucalyptus camaldulensis* plants from more arid locations have thick leaves and high oil gland density (James & Bell, 1995). Similarly water deficit can cause disorganization and collapse of cells and tissues and alteration in mesophyll and epidermis thickness in this species (Souza *et al.*, 1999).

Species adapted to specific climatic conditions must have adapted some structural adaptations (Nawazish *et al.*, 2006) and leaf anatomical feature are the representative of such environmental adaptations. Low stomatal density and presence of two or more layers of palisade cells in leaves are the xeromorphic features (Cao, 2000). Trichome density is generally higher in plants from dry season than those from wet seasons and in plants from sun-exposed areas than in those from shaded area (Pérez-Estrada *et al.*, 2000).

Leaf thickness, amount of sclerification in leaves and other organs of plants, shape and orientation of stomata on both leaf surfaces and sclerification and size of vascular tissue, in particular vessels in metaxylem (Hameed *et al.*, 2009), and nature and density of salt hairs, trichomes and salt glands (Naz *et al.*, 2009) are good indicators of environmental stress like salinity and drought. The present studies were, therefore, focussed on the anatomical adaptations of leaves in the genus *Eucalyptus* (family Myrtaceae) from the Faisalabad region.

Materials and Methods

A survey was conducted in Faisalabad district and adjoining districts to collect leaf samples of different species of the genus *Eucalyptus* L., Hér. (Myrtaceae) for the comparative studies of leaf anatomy. One cm piece from the leaf centre of fully mature old leaf along the midrib was taken during February-March, 2005. For stomatal studies same leaf surface was scratched from both adaxial and abaxial surfaces to expose epidermis. The material was kept in FAA (formalin 5%, acetic acid 10%, ethyl alcohol 50% and distilled water 35%) solution for 48 h for fixation. The material was then readily transferred in Acetic Alcohol solution (one part acetic acid and three parts ethyl alcohol) for long-term preservation.

Free-hand sectioning was used for preparing permanent slide of leaf transverse sections by using double-stained standard technique (safranine and fast green) as suggested by Ruzin (1999). Anatomical characteristics including leaf thickness, size of dermal, parenchymatous, mechanical and vascular tissue and density and size of stomata were studied. The data were subjected to statistical analysis for comparison of means using six replications (Steel *et al.*, 1997).

Results

Eucalyptus crebra showed the thickest leaves in terms of both midrib and lamina thickness as compared to all other species studied in the experiment. This was followed by that recorded in *E. microtheca*, *E. citriodora*, *E. botryoides* and *E. camaldulensis* (Figs. 1, 2, 3, 4). Three species, *E. torelliana*, *E. robusta* and *E. cladocalyx* showed the minimum of this character. Epidermis cell area on both abaxial and adaxial side was the maximum in *E. alba* and *E. maculata* (Figs. 1, 2, 3, 4) and the minimum in *E. siderophloia*, *E. torelliana* and *E. melanophloia*. Sclerenchyma cell area was the maximum in *E. maculate* and the minimum in *E. siderophloia*.

Mesophyll (spongy and palisade) cell area was the maximum in *E. crebra* and *E. citriodora*. Three species, *E. melanophloia*, *E. torelliana* and *E. diversicolor* showed greatly reduced spongy cells, whereas *E. tereticornis* and *E. robusta* had reduced palisade cells (Figs. 2, 3, 4 & 5). Vascular bundle area was the maximum in *E. crebra* and *E. maculata* and the minimum in *E. longicornis*. Two species viz., *E. siderophloia* and *E. citriodora* also had very much reduced vascular bundles than those recorded in other species. Total xylem area was the maximum in *E. crebra*, *E. maculata* and *E. microthica* and *the minimum* in four species (*E. diversicolor*, *E. camaldulensis*, *E. longicornis* and *E. siderophloia*). Metaxylem area was the maximum in *E. microtheca* and the minimum in *E. falcate*, *E. cladocalyx*, *E. robusta*, *E. rudis* and *E. siderophloia*.

Stomatal area on abaxial leaf surface was the maximum in *E. cladocalyx, E. botryoids, E. camaldulensis, E. crebra* and *E. siderophloia* and the minimum in *E. torelliana* and *E. longicornis*. Stomatal area, on the other hand, was the maximum in *E. cladocalyx* and the minimum in *E. torelliana, E. crebra* and *E. siderophloia*. Stomatal density on abaxial leaf surface was the maximum in *E. falcate* and *E. robusta,* whereas minimum was recorded in *E. botryoids, E. alba, E. melanophloia* and *E. microthica* (Figs. 3, 4 5 & 6). Stomatal density on adaxial side was the maximum in *E. globulus, E. torelliana* and *E. falcata* and the minimum in *E. botryoids* and *E. cladocalyx*.

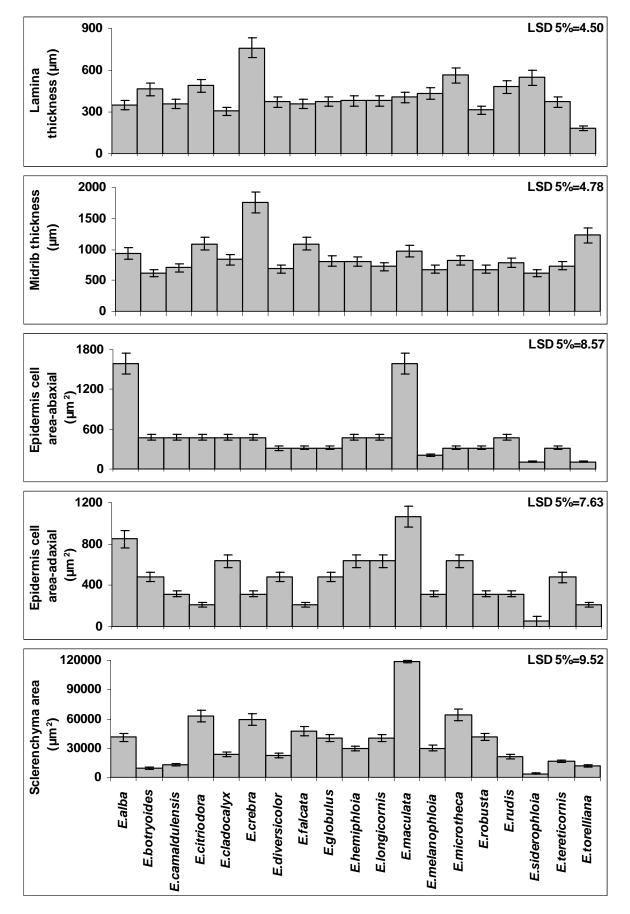


Fig. 1. Leaf anatomical characteristics (leaf thickness, epidermis, sclerenchyma) in some *Eucalyptus* species from the Faisalabad region

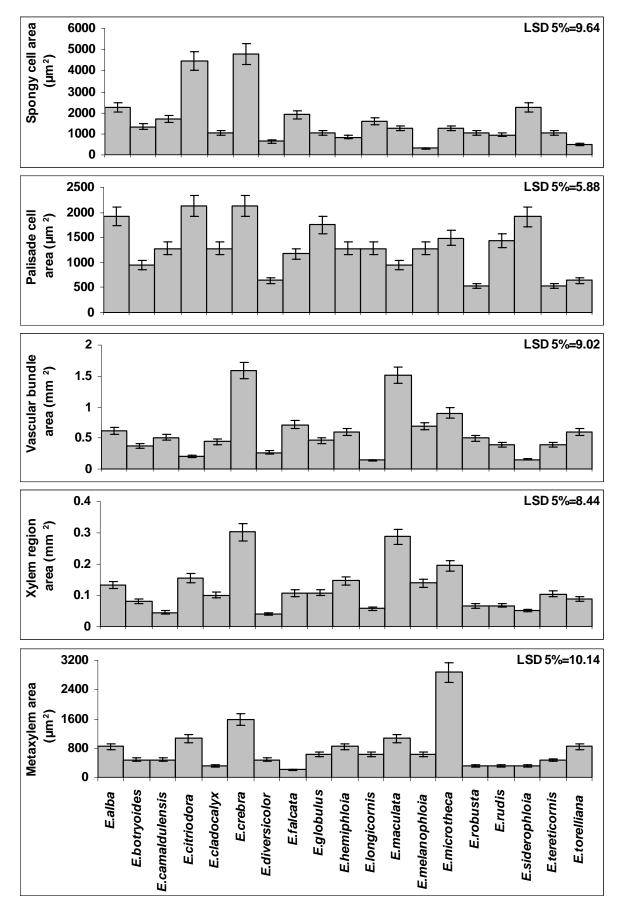


Fig. 2. Leaf anatomical characteristics (mesophyll, vascular tissue) in some *Eucalyptus* species from the Faisalabad region.

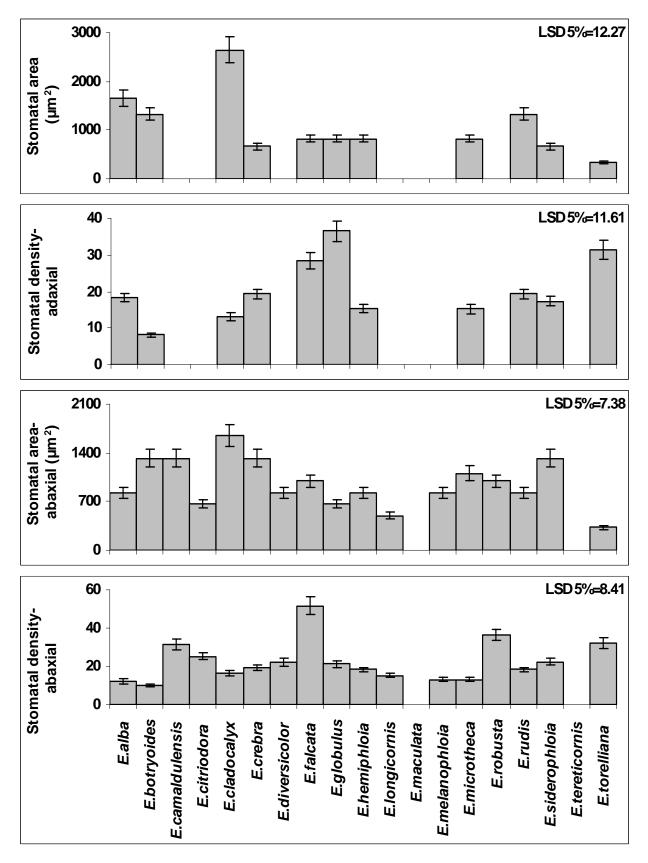
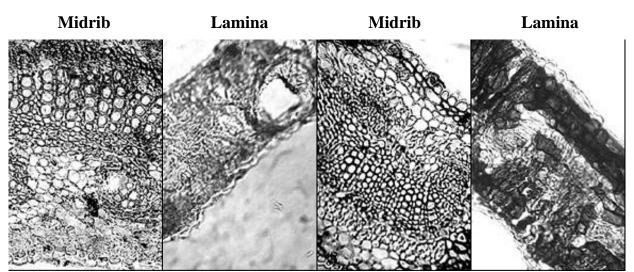
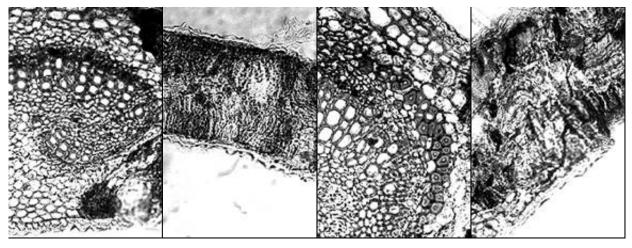


Fig. 3. Leaf anatomical characteristics (stomatal density and area) in some *Eucalyptus* species from the Faisalabad region.



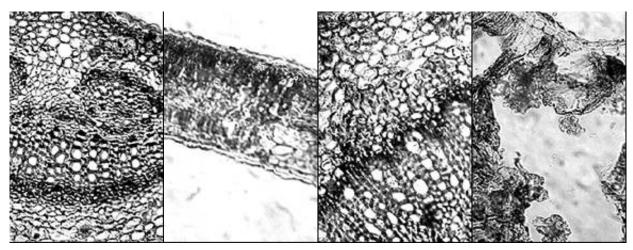
Eucalyptus alba

Eucalyptus botryoides



Eucalyptus camaldulensis

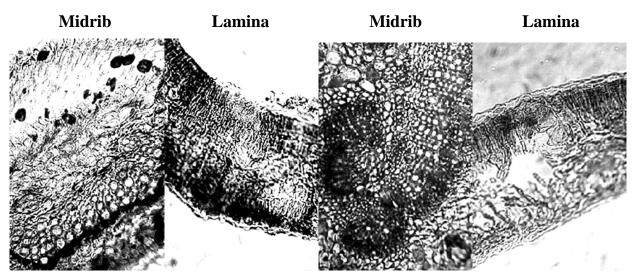
Eucalyptuscitriodora



Eucalyptus cladocalyx

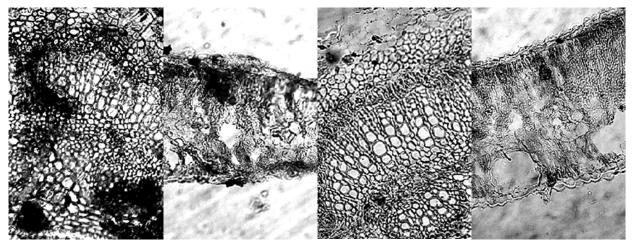
Eucalyptus crebra

Fig. 4. Leaf transverse sections of some *Eucalyptus* spp., from the Faisalabad region.



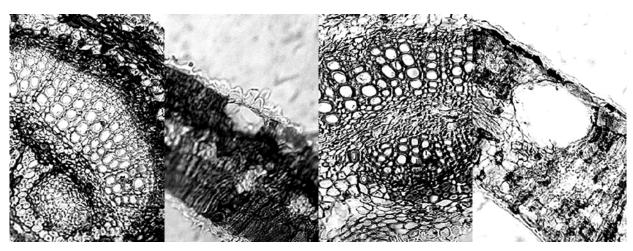
Eucalyptus diversicolor

Eucalyptus falcate



Eucalyptus globulus

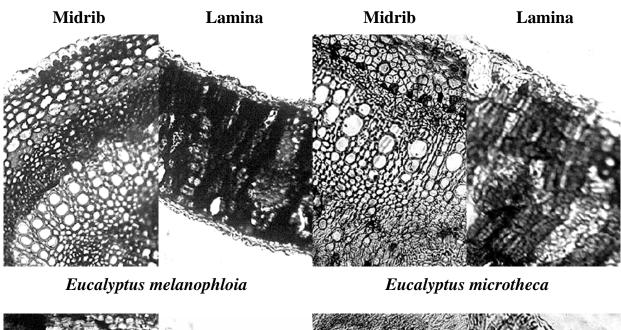
Eucalyptus hemiphloia

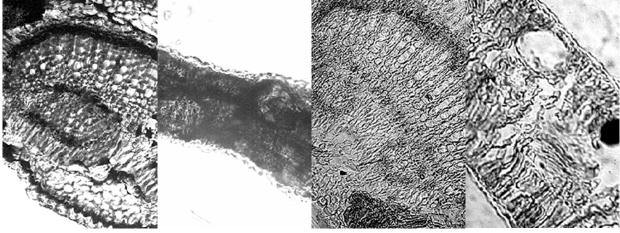


Eucalyptus longicornis

Eucalyptus maculata

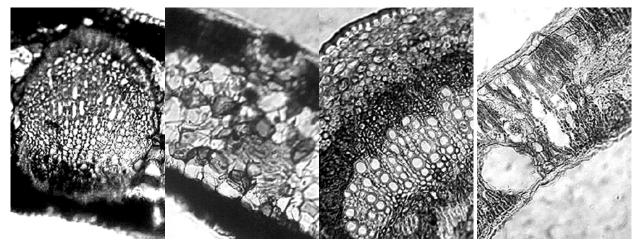
Fig. 5. Leaf transverse sections of some *Eucalyptus* spp. from the Faisalabad region.





Eucalyptus robusta

Eucalyptus rudis



Eucalyptus siderophloia

Eucalyptus tereticornis

Fig. 6. Leaf transverse sections of some Eucalyptus spp. from the Faisalabad region

Table 1. List of <i>Eucalyptus</i> species used for leaf anatomical studies from the Faisalabad region.		
Species	Vernacular name	Place of collection
Eucalyptus alba Reinw. ex Blume	White gum	PFRI, Gatwala
Eucalyptus botryoides Sm.	Bangalay	PFRI, Gatwala
Eucalyptus camaldulensis Dehn.	River Red Gum	UA, Faisalabad
Eucalyptus citriodora Hook.	Lemon-scented gum	UA, Faisalabad
Eucalyptus cladocalyx F. Muell.	Sugar gum	PFRI, Gatwala
Eucalyptus crebra F. Muell.	Narrow-leaved Red Ironbark	PFRI, Gatwala
Eucalyptus diversicolor F. Muell.	Karri	PFRI, Gatwala
Eucalyptus falcata Turcz	Silver mallee	Changa Manga Plantation
Eucalyptus globulus Labill.	Tasmanian blue gum	UA, Faisalabad
Eucalyptus hemiphloia F. Muell. ex Benth.	White Gum	Changa Manga Plantation
Eucalyptus longicornis (F.Muell.) F.Muell. ex Maiden	Red morel	PFRI, Gatwala
Eucalyptus maculata Hook.	Spotted gum	PFRI, Gatwala
Eucalyptus melanophloia F.Muell.	Silver-leaved ironbark	Changa Manga Plantation
Eucalyptus microtheca F. Muell.	Coolibah	Changa Manga Plantation
Eucalyptus robusta Sm.	Swamp Mahogany	UA, Faisalabad
Eucalyptus rudis Endl.	Desert Gum	PFRI, Gatwala
Eucalyptus siderophloia Benth.	Grey ironbark	UA, Faisalabad
Eucalyptus tereticornis Smith	Forest redgum	Changa Manga Plantation
Eucalyptus torelliana F. Muell.	Cadaghi	PFRI, Gatwala

Table 1. List of *Eucalyptus* species used for leaf anatomical studies from the Faisalabad region.

Punjab Forestry Research Institute (PFRI), Gatwala University of Agriculture (UA), Faisalabad

Discussion

Eucalyptus species, in general, have wide range of distribution in their native region and can tolerate a variety of environmental stresses (Merchant *et al.*, 2009), for example tolerant to salt stress as showing high survival percentage and stimulated growth at mild salinities (Cha-um & Ckirdmanee, 2008). These are the useful trees that can cause remediation of saline soil in terms of lowering saline water table by using underground water (Barrett-Lennard, 2002). Understanding the mechanisms of tree adaptation to environmental hazards like aridity and saline conditions in terms of morpho-anatomy and physiology is a major challenge in modern day science (Merchant *et al.*, 2009). Many *Eucalyptus* species have been found cultivated in Pakistan at different locations and quite a few of them are successfully adapted to the climates of Pakistan. A comprehensive list have been published by Ahmad (1996) in which habitat of most of the species have been presented in details. The anatomical studies of leaf were an attempt to correlate the successful survival of this genus in view of structural modifications to cope with a variety of abiotic factors.

Thick epidermis is known to be useful in checking water loss under limited moisture conditions along with the thickness of cuticle layers (Rashid *et al.*, 2001; Bahaji *et al.*, 2002). Thicker epidermis in *E. alba* and *E. maculata* are expected to be adapted to harsh water limited environments like drought and salinity.

Larger cortical cell area seems to be related to better storage of moisture that is essential for survival under harsh climates. Large cortical cells in *E. microtheca* and *E. botryoides* are the indication of their wide distribution in a variety of different environmental conditions, as reported by Zwieniecki & Newton (1995) and Baloch *et al.*, (1998).

Vascular bundle area seems to be directly related to efficient transport of water and nutrients from the soil, and these might be of greater importance under low availability of moisture. Greater vascular bundle size has been reported by Awasthi & Pathak (1999), in saline tolerant genotypes of *Ziziphus* species. *Eucalyptus crebra, E. maculata* and *E.*

microtheca with significantly larger vascular tissue perhaps the better adaptation to a variety of environment types, and hence, the wider distributional range as compared to other species. In conclusion, there was a great variation in leaf anatomical characteristics among *Eucalyptus* species collected from the Faisalabad region.

Leaf anatomical characteristics seemed to be not solely important in the distribution of the *Eucalyptus* species in the Punjab. The wider distribution of *E. citriodora* and *E. camaldulensis* in Central Punjab was apparently not correlated with structural modification and therefore, some physiological and biochemical aspects may also contribute in the successful adaptation if these species to a variety of habitat types throughout the country. Both these species, especially *E. camaldulensis*, have been tested for salt stress (Akilan *et al.*, 1997), drought (Prat & Fathi-Ettai, 1990), and waterlogging (Marcar, 1993).

Ecalyptus microtheca is well known for its high drought tolerance (Li & Wang, 2003). This is supported by specific structural adaptations like leaf thickness, increased sclerification in leaves, and total vascular region area and metaxylem area. *Eucalyptus maculata* is relatively tolerant to drought stress (Ahmad, 1996) and showed specific leaf structural adaptations such as thicker leaves, extensive sclerification and large xylem. On the whole, all the *Eucalyptus* species showed great amount of variation in their leaf anatomy and this may be the representative of their diverse ecological range.

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