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Stomatal studies in some afforestation tree species

[*1] Saralabai Viswanathan Chathlingathe, [2] Sakthivel Vellaikkannu, [3] Magadevan Subramani

[1] P. G. research Department of Botany, Government Arts College, (Autonomous) Coimbatore, Tamil Nadu, India
[2] Department of Biotechnology, P.G. Extension Centre, Bharathidasan University, Perambalur, Tamil Nadu. India
[3] Corporation Higher Secondary School, Peelamedu, Coimbatore, Tamil Nadu, India

Abstract
Trees form an important stabilizing agent in the forest and Savanna Ecosystem by protecting the soil against radiation and erosion. They conserve soil water by encouraging percolation and discouraging surface run off. They also play an important role in the water cycle as more than 98% of water absorbed by plants is lost to the atmosphere through stomatal, cuticular and lenticular transpirations. Over the years, exploitation of trees for domestic, agricultural and industrial purposes has been indiscriminate, thus resulting in deforestation, a progressive process of forest removal, which also causes climate change. Frequency of each stomatal complex type was determined as percentage occurrence in a total of 120 fields of view at 40 objective lens. Stomatal index was determined based on the number of stomata plus number of epidermal cells per square millimeter multiplied by 100. From both upper and lower surfaces, presence or absence of stomata was noted to determine the leaf type whether it is epistomatic, hypostomatic or amphistomatic. The index of similarity between the abaxial and adaxial surfaces of the leaf was determined as the number of stomatal complex types common to both surfaces.
Stomatal complex types are heterogeneous in all the four species studied. The heterogeneity varies from species to species. Four types in *Eucalptus globulus*, four types in *Polyalthia longifolia*, three types in *Ficus benghalensis* and three types in *Ficus religiosa*. The heterogeneity of the stomatal complex types is noteworthy. It is believed to be a factor in stomatal conductance. Those species with most heterogeneous stomata transpired more than others which are not. For instance, *E. torelliana* and P. longifolia which have four types each transpired at higher rate than *E. camaldulensis* and *P. pendula* with three types respectively. Moreover, the number of subsidiary cells having direct contact with the guard cells is relevant in determining the rate of transpiration. Presence of trichomes in this species may also be a factor of its lower transpiration rate than *E. Torelliana.*

Keywords: heterogenious stomatal types, stomatal index, stomatal frequency, afforestation

1. Introduction
Afforestation means massive program of social forestry to meet demands to local people for fuel, fodder, timber etc. Afforestation restores ecological balance of all ecosystems, maintain biological diversity, and act as catchments for all soil and water, conservation, prevent floods and future of tribal people. Our life is dependent upon trees. Trees and plants produce oxygen (O_2) through the process of photosynthesis. Men's experience of the last century have taught him that trees and plants are the key factors to strike a balance in the eco-system.
In the choice of trees species for the afforestation project, factors for consideration include rate of growth, stem morphology, foliage or canopy size, physiological and anatomical capacity for conservation of water and for humidification of the atmosphere [4]. The latter factor was considered in this work, because the anatomy of the stomatal complex is relevant to transpiration rate as demonstrated in some *Citrus* species [6] and in some vegetable species in Nigeria [1]. The present study, focussed on elucidation of the possible relationship between the stomatal complex types and the rate of transpiration in the selected tree species.

2. Materials and Methods
2.1 Collection of leaf specimens
The specimens were collected from the Government Arts College (Autonomous), Coimbatore, Tamil Nadu, India. The identification was authenticated at the Botanical Survey of India, Southern Regional Centre, Tamil Nadu Agricultural University Campus, Coimbatore, Tamil Nadu.

2.2 Isolation and determination of epidermal layer
Thin paradermal sections were cut using free hand method. The cut portions were placed on slides stained with Safranin and mounted in Glycerine. These sections were then studied under the Olympus Light Microscope.

2.3 Identification and determination of the stomatal complex types
Stomatal complex types were identified using the description made by [3, 5]. Frequency of each stomatal complex type was determined as percentage occurrence in a total of 120 fields of view at x 40 objective lens.

2.4 Determination of stomatal density and stomatal index
Stomatal index was determined based on the number of stomata plus, number of epidermal cells per square millimeter multiplied by 100 [3]. Sample size of 30 was used for each of the parameters.

2.5 Study of the structure, number, distribution and types of stomata
Stomata are minute pores of eliptical shape, consists of two specialized epidermal cell called guard cells.

2.6 Number of Stomata (Stomatal Frequency)

The number of stomata in a definite area of leaf varies from plant to plant. The number of stomta per unit area of leaf is called Stomatal Frequency. Stomata nearly occupy one to two percent of total leaf area when fully open. In isobilateral leaves (in monocots). Approximately the same number of stomata are found on upper surface (adaxial) and lower (abaxial) surface. But in dorsiventral leaves (in dicots) the number of stomata on the upper surface is much less in comparison to those found on the lower surface.

2.7 Distribution and Types of Stomata

Depending upon the distribution and arrangement of stomata in the leaves six categories of stomatal distribution have been recognized in plants. 1. Apple or mulberry (hypostomatic) type: Stomata are found distributed only on the lower surface of leaves, e.g., apple, peach, mulberry, walnut, etc. 2. Potato type: Stomata are found distributed more on the lower surface and less on its upper surface, e.g., potato, cabbage, bean, tomato, pea, etc. 3. Oat (amphistomatic) type: Stomata are found distributed equally upon the two surfaces, e.g. maize, oats, grasses, etc. 4. Water lily (epistomatic) type: Stomata are found distributed only on the upper surface of leaf, e.g., water lily, Nymphaea and many aquatic plants 5. Staurocytic type: Stomata surrounded by three to five similar subsidiary cells with anticlinal walls arranged cross-wise to the guard cells. 6. Potamogeton (astomatic) type: Stomata are altogether absent or if present they are vestigeal. e.g., Potamogeton and submerged aquatics.

Further Metacalf and Chalk recognized four types of stomata on the basis of their structure- a. Anomocytic type: In these stomata, accessory cells are absent. The guard cells are surrounded by ordinary epidermal cells, e.g., families Ranunculaceae, Cucurbitaceae, Papaveraceae and Malvaceae. b. Anisocytic type: In these stomata the guard cells are surrounded by three accessory cells. Of these two are larger whereas one is smaller in size.g., family Brassicaceae. c. Diacytic type: In these stomata the guard cells are surrounded by two accessory cells. Their common walls are at right angle to the walls of guard cells, families Caryophyllaceae, Acanthaceae. d. Paracytic type: In these stomata the guard cells are also surrounded by two accessory cells, but their common walls are parallel to guard cells, e.g., families Rubiaceae, Fabaceae etc.

On the basis of development, there are three types of stomata [7]: Mesogynous type: In this type of stomata guard cells as well as subsidiary or Accessory cells both are developed from one mother cell. e.g. Rubiaceace & Brassicaceae family. Perigynous type: In this type guard cells are formed from mother cell while subsidiary cells from nearby mother cells, eg.: Cucurbitaceae family. Mesoperigynous type: In this type guard cells & one subsidiary cells is formed from mother cell while other subsidiary cells develop Independently. e.g.: Ranunculaceae, Caryophyllaceae family.

3. Results and Discussion

Stomatal complex types are heterogeneous in all the four species studied (Table 1). The heterogeneity varies from species to species. Four types were found in *Eucalptus globulus*, four types in *Polyalthia longifolia*, three types in *Ficus benghalensis* and three types in *Ficus religiosa*. Six types of stomatal complex was identified including anomocytic which occurs in all the four species. Anomo tetracytic occurs in three species of *E. globulus*, and *P. longifolia*. Anisocytic occrs in the *E. globulus*, brachyparacytic and paratetracytic occurs in *P. longifolia*. Hemiparacytic occurs only in *Eucalyptus*. The distribution of the stomatal complex types on the leaf surface in three species is said to be hypostomatic except in *E. globulus* which has stomata on both surfaces hence leaves are amphistomatic.

Table 1: List of tree species studied

Species	Common name	Family
Eucalptus globulus	Eucalyptus	Myrtaceae
Polyalthia longifolia	Asoka	Annonaceae
Ficus benghalensis	Banian	Moraceae
Ficus religiosa	Bodhi tree	Moraceae

The heterogeneity of the stomatal complex types is noteworthy. It is believed to be a factor in stomatal conductance. Those species with most heterogeneous stomata transpired more than others which are not. For instance, *E. Globulus* and *P. longifolia* which have four types each transpired at higher rate than *F. Benghalensis* and *F. religiosa* with three types respectively (Table 2).

Table 2: Stomatal anatomy of some afforestation tree species.

Species	Surface	Stomatal types	Stomatal frequency (%)	Stomatal index (%)
Eucalptus globulus	Upper	Anisocytic Anamotetracytic Anomocytic	28.68 48.53 22.79	9.21
	Lower	Hemiparacytic Anisocytic Anamotetracyctic Anomocytic	1.10 28.53 47.79 22.52	1.35
Polyalthia Longifolia	Upper	-		
	Lower	Brachyparacytic Paratetracytic Anamotetracytic Anamocytic	18.79 19.39 33.94 27.88	12.15
Ficus benghalensis	Upper	-	-	
	Lower	Anomocytic Staurocytic Anomotetracytic	28.18 22.09 38.53	9.16
Ficus religiosa	Upper	-	-	
	Lower	Anomocytic Staurocytic Anomotetracytic	38.32 32.33 48.53	13.15

Moreover, the number of subsidiary cells having direct contact with the guard cells is relevant in determining the rate of transpiration. Carr and Carr (1990) proposed that large number of subsidiary cells may be responsible for rapid stomatal opening.

4. Conclusion

Stomatal complex types are heterogenous in all the four afforestation tree species studied. Those species with most hetergenous stomata transpired more than less heterogenous stomata. Moreover, the number of subsidiary cells and the presence of trichomes may also reduce transpiration rate. Hence, the tree species with homogenous stomata and with more number of subsidiary cells and tricomes are recommended for afforestation programme.

5. References

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