

ANATOMICAL STUDIES ON LEAVES AND SHOOTS OF SOME URBAN TREES

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Abstract

The present study was conducted on eleven urban tree species i.e., *Acacia nilotica* L., *Alstonia scholaris* R. Br., *Cassia fistula* L., *Conocarpus erectus* L., *Dalbergia sissoo* Roxb., *Eucalyptus citriodora* Hook., *Melia azedarach* L., *Mimusops elengi* L., *Pongamia pinnata* L., *Putranjiva roxburghii* Wall. and *Syzygium cumini* L. The anatomical studies were carried out on one and a half year old saplings of these species, which were planted in Botanic Garden Government College University Lahore. Stomata and xylem anatomy was carried out using light microscope. The results showed that the size of stomata ranged from 5.50 to 11.93 μm , the maximum size was found in *Mimusops elengi* and the minimum was found in *Cassia fistula*. The number of stomata ranged from 22.5 to 212, maximum was found in *Alstonia scholaris* and minimum in *Mimusops elengi*. The size of stem xylem vessel ranged from 12.21 to 25.52 μm , maximum was found in *Cassia fistula* and minimum in *Mimusops elengi*. The number of stem xylem vessel ranged from 8.75 to 45.5, maximum was found in *Alstonia scholaris* and minimum in *Acacia nilotica*. The size of root xylem vessel ranged from 18.07 to 80.61 μm , maximum was found in *Dalbergia sissoo* and minimum in *Mimusops elengi*. The number of root xylem vessel was ranged from 3 to 14, maximum was found in *Alstonia scholaris* and minimum in *Dalbergia sissoo*. The theoretical hydraulic conductivity (k_i) for stem ranged from $1.37 \times 10^{-11} \text{ Kg } \mu\text{m}^{-1} \text{ Mpa}^{-1}\text{s}^{-1}$ to $6.01 \times 10^{-11} \text{ Kg } \mu\text{m}^{-1} \text{ Mpa}^{-1}\text{s}^{-1}$, maximum in *Acacia nilotica* and minimum in *Cassia fistula*. The theoretical hydraulic conductivity value for root xylem vessel ranged from $1.01 \times 10^{-11} \text{ Kg } \mu\text{m}^{-1} \text{ Mpa}^{-1}\text{s}^{-1}$ to $6.69 \times 10^{-11} \text{ Kg } \mu\text{m}^{-1} \text{ Mpa}^{-1}\text{s}^{-1}$, maximum in *Acacia nilotica* and minimum in *Syzygium cumini*. It is concluded that species with maximum k_i value consumed more water and on the basis of water conduction per xylem vessels and the species with small k_i conduct less water per xylem vessel. Such data can be useful to plan the plantation for the urban ecosystems.

Introduction

The morphological and anatomical features of different species determine the performance and productivity (Wright *et al.*, 2004; Sack *et al.*, 2013). Functions of leaves in the ecosystem play fundamental roles as they control the exchange of carbon and water (Wright *et al.*, 2004; Reich, 2014). Under the different environmental conditions variations in the anatomical components and plant trait have been observed which lead to affect the performance of plants (Witkowski & Lamont, 1991). Species of stressful environment or long life span might have low maximum rate of photosynthesis and low concentration of nutrients. The survival of plants is limited by environmental stress factors at different global levels. All levels of organizations in the plants are effect by water stress. Opening of stomata causes the diffusion of CO_2 in leaves, as photosynthetic rate is positively linked with stomatal conductance (Farquhar & Sharkey, 1982). Structurally safe xylem is characterized by thick walls which are narrow, hence resistant to cavitation but with potentially slow the gain of photosynthetic carbon (Hubbard *et al.*, 1999; Hacke & Sperry 2001).

Environment where evaporation is high and water supply is limited, the species with hydraulically safe xylem tend to dominate. In leaves, rate of gas exchange coordinated with water stress causes leaf venation system to change its hydraulic efficiency. Structures of the stomata along with its functional properties are also changed under stress conditions (Aasamaa & Sober, 2001). Changes that occur during drought conditions are irreversible. Stomatal size and density are determined by water status and stomatal density linked with parameter of gas exchange (Xu & Zhou, 2008). Leaves expand more than other anatomical structures during non-stress conditions. When water is limited stomatal pattern and hydraulic traits consume water in such way that their survival would be possible. With the passage of time requirements of plants vary depending on ontogenetic stage as well as local environment and regional environment (Poorter & Kitajima, 2007; Wimmer, 2002).

The study of functional characteristics helps to understand the effect of environment (Baraloto *et al.*, 2010). Relationship of coordination of leaf, stem, and root are related to the growth of plants and their

survival (Reich *et al.*, 2003; Cavender-Bares *et al.*, 2004). Such environment where growth of trees is determined by the availability of water, the size of xylem vessels, their arrangement and frequency can play a major role (Tyree & Zimmermann, 2002). In plants water moving through xylem path is influenced by diameter and frequency of xylem vessels (Sperry, 2003).

Need of woody species varies for support, water transport and storage. Capacity of growth in woody plant is constrained by xylem hydraulics. Plant growth is also related to stem traits. As age of the tree progresses, growth is limited by a factor called hydraulic resistance. Mechanical support is provided by dense wood and resulting in slow growth rate. In woody species fine roots determine the stem length and branches (McCully, 1999; Draye, 2000). The ability of perennial plants to draw upon stored nutrient reserves reduces the need for high nutrient uptake rates to occur simultaneously with new leaf production. In fact, root and leaf production are frequently asynchronous (Wells & Eissenstat, 2000).

Root traits are less dependent and less supportive when sources are deficient. They get nutrients from the soil independently (Eissenstat, 1992; Brundrett, 2002; Menge *et al.*, 2008; Comas *et al.*, 2014). Previous increase in growth of species is observed under functional traits (Schnitzer and Bongers 2011). An economic spectrum develops which describes the leaves amount for fine roots (Wahl and Ryser, 2000; Reich *et al.*, 2003; Lavorel *et al.*, 2007). Root traits belong to particular group rapidly use the belowground resources as a result poor structural development and lead to excessive turnover of root (Eissenstat & Yanai 1997; Eissenstat *et al.*, 2000; Cranie *et al.*, 2003). Among plant groups functional character of leaf and roots are linked with one another. It is the trait of leaf to provide poor substitution for function of root. For understanding of plant strategies of resources, it is important to observe belowground and aboveground traits (Eissenstat 1992).

Whole functioning of the plant is linked with vascular system because it has functional properties and developmental coordination of hydraulic traits and photosynthesis (Brodribb, 2009; Beerling & Franks, 2010). When plant under water stress, different changes take place at different level in whole plant which balance the initial negative effect of water (Boyer, 1982; Chaves *et al.*, 2003; Flexas *et al.*, 2006). Changes take place due to water stress is because all the functions in the plants are related with each other (Heckenberger *et al.*, 1998; Niinemets and sack, 2006). Cell expansion and growth of plant is inhibited by low

availability of the water (Hsiao and Xu, 2000). Leaves which adapt themselves in stress conditions, they develop small size leaves as compared to normal leaves growing in normal conditions. Cytological structure of leaves also may change (Karamanos *et al.*, 1982; Chaves *et al.*, 2003; Kramer & Boyer 1995).

Photosynthesis and gas exchange negatively change with water stress. When water stress applied for short period it is observed that its only effects are functional consequences but in natural environment water stress occurs after many days and weeks (Flexas *et al.*, 2006).

Growth and aboveground biomass accumulation follow a common pattern as tree size increases, with productivity peaking when leaf area reaches its maximum and then declining as tree age and size increase (Ryan & Waring, 1992). Age- and size-related declines in forest productivity are major considerations in setting the rotational age of commercial forests, and relate to issues of carbon storage, since changes in forest structure can influence large-scale biomass accumulation. Despite the ecological and practical significance of the growth and aboveground biomass accumulation follow a common pattern as tree size increases, with productivity peaking when leaf area reaches its maximum and then declining as tree age and size increase (Ryan & Waring, 1992). Age and size-related declines in forest productivity are major considerations in setting the rotational age of commercial forests, and relate to issues of carbon storage, since changes in forest structure can influence large-scale biomass accumulation.

When amount of water in the soil is gradually low various functions of the trees are affected (Hsiao & Xu 2000). Leaf, root and stem show different response in drought conditions because they are sensitive to drought conditions, growth of tree is affected in arid as well as in humid climate. During stress conditions chemical and hydraulic signals exist between roots and shoots develop communication. Drought, change the physiological conditions of the tree but cell extension is more sensitive process. An individual tree can bear the drought conditions but leaf canopy area, growth rate and reproductive success, all of these related through a number of anatomical and physiological processes (Boyer 1982; Chaves *et al.*, 2003). The objectives of present study were to; 1) investigate the vessel and stomatal anatomy and their comparison across the species 2) to understand the tradeoff between the anatomical traits.

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comparison across the species 2) to understand the tradeoff between the anatomical traits.

Materials and Methods

Study site: This study was carried out at Botanical Garden Government College University, Lahore situated at Mall road. Annual temperature of Botanical Garden is 30°C, mean annual temperature in summer is 36°C and annual mean temperature in winter is 25°C. Range of annual precipitation is 250-500mm (Khan, 1996).

Plant species and sample collection: Eleven tree species belonging to seven families were studied (Table 3.1). These plant species were one and half year old and planted in experimental plot of Botanical Garden. Four replicates of each specie used for the anatomical study. Samples were collected in the zipper bags and labeled. Leaves were selected for study of stomata while branch and root samples were collected for the study of xylem. For these foliar and branch anatomical investigations, for individual trees of each species were selected and for each tree 5-6 leaves and 3-5 branches of equal size were selected.

Study of stomata: Impression method was used for the study of stomatal numbers (Zhao *et al.*, 2017). Before microscopic study of leaves, dust particles were removed carefully with the help of camel hair brush. Randomly selected leaves of four replicates placed on the smooth surface then applied nail polish varnish on the abaxial surface of leaves. After 3 minutes, nail polish was removed with the help of tweezers. Mounted the layer of nail polish on the glass slide and added a drop of water with the help of dropper and placed cover slip carefully and observed the glass slide under 40X power of the light microscope.

Section cutting for study of xylem vessels: Randomly selected bra etnches and fine roots of each species were used. Selected branches and roots of each replicates were washed. Peel-off the epidermis of the branch. Sections were prepared by free hand-sectioning of roots and branches. Free-hand sectioning was done by using blade. Sections were placed in the petri plate with some water. To observe the section, it was mounted on the glass slide. Added a drop of water and placed a cover slip on it. Placed glass slide under 40X of light microscope for observation.

Photography: Images of stomata and vessels of roots and branches were taken with the help of cell phone camera. Stomatal density was estimated by counting the total number of stomata. Stomatal length was calculated by using image J and 10 randomly selected stomata were

used for measurement for each image of stomata and vessel. Size of vessels and stomata was estimated by using image J (Fan *et al.*, 2011).

Data Analysis: The data was analyzed using Sigma Plot version 14.5. The mean values of studied traits were compared across species using t-test.

Theoretical hydraulic conductance of xylem vessel: Theoretical hydraulic conductance of vessel was calculated (Fan *et al.*, 2011).

Table 1: Species scientific names, codes, family and number of replicates

Sr. no.	Species name	Species code	Family
1	<i>Acacia nilotica</i> L.	An	Fabaceae
2	<i>Alstonia scholaris</i> R.Br.	As	Apocynaceae
3	<i>Cassia fistula</i> L.	Cf	Fabaceae
4	<i>Conocarpus erectus</i> L.	Ce	Combretaceae
5	<i>Dalbergia sisso</i> Roxb.	Ds	Fabaceae
6	<i>Eucalyptus citriodora</i> Hook.	Ec	Myrtaceae
7	<i>Melia azedarach</i> L.	Ma	Meliaceae
8	<i>Mimusops elengi</i> L.	Me	Sapotaceae
9	<i>Pongamia pinnata</i> L.	Pp	Fabaceae
10	<i>Putranjiva roxburghii</i> Wall.	Pr	Euporbiaceae
11	<i>Syzygium cumini</i> L.	Sc	Myrtaceae

Results

Stem vessel size across the species studied: The stem vessel size ranged from 12.2µm to 25.5µm, the maximum was found in *Cassia fistula* while the lowest in *Mimusops elengi*. The individual species mean values are shown in figure 1 and the mean comparison across species are shown in Table 2.

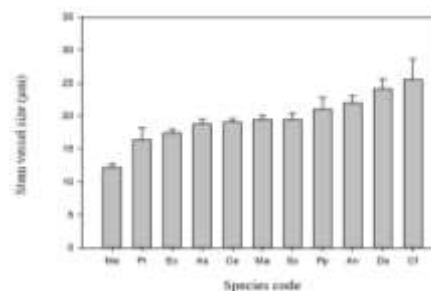


Fig. 1. Stem vessel size across species studied. Species codes are listed in Table 3.1.

It was found that *Acacia nilotica* showed the significant difference of vessel size with *Melia azedarach* and *Eucalyptus citriodora*, while no significant differences were found for *Acacia nilotica* with the rest of species. *Alstonia scholaris* showed significant difference with *Dalbergia sisso*, and *Melia azedarach*. While *Cassia fistula* showed significant

difference with *Melia azedarach* only. *Conocarpus erectus* showed the significant difference with *Dalbergia sisso* and *Melia azedarach*. *Melia azedarach* showed significant difference with *Mimusops elengi*. *Mimusops elengi* showed significant difference with *Pongamia pinnata*, *Syzygium cumini* and *Eucalyptus citriodora*

Table 2. Differences of stem vessels size across the species studied.

Species names	As	An	Cf	Ce	Ds	Ma	Me	Pp	Pr	Sc	Ec
An	-----	21.999 18.784 0.057	21.999 25.526 0.358	21.999 19.089 0.069	21.999 24.134 0.290	21.999 19.422 0.101	21.999 12.219 <0.001	21.999 21.030 0.670	21.999 16.983 0.071	21.999 19.443 0.134	21.999 17.412 0.017
As	-----	-----	18.784 25.526 0.112	18.784 19.089 0.745	18.784 24.134 0.026	18.784 19.422 0.551	18.784 12.219 <0.001	18.784 21.030 0.322	18.784 16.983 0.425	18.784 19.443 0.617	18.784 17.412 0.194
Cf	-----	-----	-----	25.526 19.089 0.135	25.526 24.134 0.710	25.526 19.422 0.149	25.526 12.219 0.023	25.526 21.030 0.278	25.526 16.983 0.070	25.526 19.443 0.150	25.526 17.412 0.082
Ce	-----	-----	-----	-----	19.089 24.134 0.035	19.089 19.422 0.703	19.089 12.219 <0.001	19.089 21.030 0.374	19.089 16.983 0.350	19.089 19.443 0.763	19.089 17.412 0.0620
Ds	-----	-----	-----	-----	-----	24.134 19.422 0.040	24.134 12.219 0.002	24.134 21.030 0.237	24.134 16.983 0.026	24.134 19.443 0.043	24.134 17.412 0.014
Ma	-----	-----	-----	-----	-----	-----	19.422 12.219 <0.001	19.422 21.030 0.460	19.422 16.983 0.294	19.422 19.443 0.987	19.422 17.412 0.062
Me	-----	-----	-----	-----	-----	-----	-----	12.219 21.030 0.014	12.219 16.983 0.081	12.219 19.443 0.002	12.219 17.412 <0.001
Pp	-----	-----	-----	-----	-----	-----	-----	-----	21.030 16.983 0.175	21.030 19.443 0.484	21.030 17.412 0.142
Pr	-----	-----	-----	-----	-----	-----	-----	-----	-----	16.983	16.983

										19.443	17.412
										0.305	0.839
Sc	----	----	----	----	----	----	----	----	----	----	19.443
											17.412
											0.137
Ec	----	----	----	----	----	----	----	----	----	----	----

Note: Bold number indicates the significant differences in the mean values $p < 0.05$.

Stem vessel number across the species studied: The stem vessel number ranged from 8.75 to 45.5, the maximum was found in *Alstonia scholaris* while the lowest in *Acacia nilotica*. The individual species mean value mean are shown in figure 2 and the comparison across species are shown in Table 3

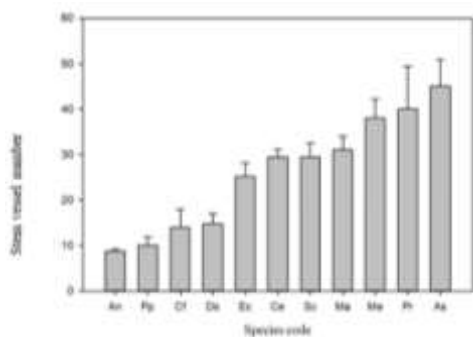


Fig. 2. Stem vessel number across species studied. Species codes are listed in Table 1.

It was found that *Acacia nilotica* showed the significant difference with *Alstonia scholaris*, *Conocarpus erectus*, *Melia azedarach*, *Mimusops elengi*, *Putranjiva roxburghii*, *Syzygium cumini*. *Alstonia scholaris* showed the significant differences with *Cassia fistula*, *Dalbergia sissio*, *Pongamia pinnata*, *Eucalyptus citriodora*. *Cassia fistula* showed the significant differences with *Conocarpus erectus*, *Melia azedarach*, *Mimusops elengi*, *Putranjiva roxburghii*, *Syzygium cumini*. *Conocarpus erectus* showed the significant differences with *Dalbergia sissio*, *Pongamia pinnata*. *Dalbergia sissio* showed the significant differences with *Melia azedarach*, *Mimusops elengi*, *Putranjiva roxburghii*, *Syzygium cumini*, *Eucalyptus citriodora*. *Melia azedarach* showed the significant differences with *Pongamia pinnata*. *Mimusops elengi* showed the significant differences with *Pongamia pinnata*. *Pongamia pinnata* showed the significant differences with *Putranjiva roxburghii*, *Syzygium cumini*, *Eucalyptus citriodora*

Table 2. Differences of stem vessel number across the species studied.

Species names	An	As	Cf	Ce	Ds	Ma	Me	Pp	Pr	Sc	Ec
An	----	8.750 45.500 0.008	8.750 14.00 0.275	8.750 29.500 0.0154	8.750 14.750 0.079	8.750 31.000 0.005	8.750 38.000 0.005	8.750 10.000 0.559	8.750 40.000 0.045	8.750 29.500 <0.001	8.750 25.50 0.010
As	----	----	45.500 14.00 0.006	45.500 29.500 0.074	45.500 14.750 0.009	45.500 31.000 0.087	45.500 38.000 0.342	45.500 10.000 0.006	45.500 40.000 0.641	45.500 29.500 0.068	45.500 25.5 0.033
Cf	----	----	----	14.00 29.500 0.036	14.00 14.750 0.876	14.00 31.000 0.016	14.00 38.000 0.006	14.00 10.000 0.407	14.00 40.000 0.063	14.00 29.500 0.022	14.00 25.50 0.066

Ce	----	----	----	----	29.500 14.750 0.031	29.500 31.000 0.784	29.500 38.000 0.199	29.500 10.000 0.012	29.500 40.000 0.364	29.500 29.500 1.000	29.500 25.50 0.444
Ds	----	----	----	----	----	14.750 31.000 0.007	14.750 38.000 0.007	14.750 10.000 0.165	14.750 40.000 0.003	14.750 29.500 0.003	14.750 25.50 0.033
Ma	----	----	----	----	----	----	31.000 38.000 0.226	31.000 10.000 0.002	31.000 40.000 0.420	31.000 29.500 0.688	31.000 25.50 0.227
Me	----	----	----	----	----	----	----	38.000 10.000 0.003	38.000 40.000 0.855	38.000 29.500 0.129	38.000 25.50 0.050
Pp	----	----	----	----	----	----	----	----	10.00 40.00 0.047	10.000 29.500 <0.001	10.00 25.50 0.007
Pr	----	----	----	----	----	----	----	----	----	40.000 29.500 0.349	40.000 25.50 0.217
Sc	----	----	----	----	----	----	----	----	----	----	29.500 25.50 0.268
Ec	----	----	----	----	----	----	----	----	----	----	----

Note: Bold number indicates the significant differences in the mean values $p < 0.05$.

Root vessel size across the species studied: The root vessel size ranged from 18.07um to 80.61um, the maximum root vessel size was found in *Dalbergia sisso* while the lowest in *Mimusops elengi*. The individual species mean values are shown in figure 3 and the comparison across species are shown in Table 3.

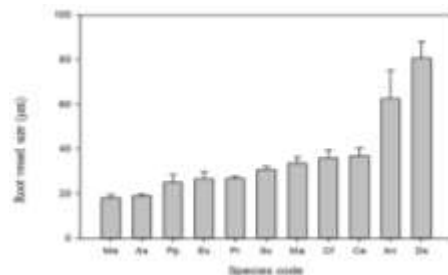


Fig. 3. Root vessel size across species studied. Species codes are listed in Table 1

It was found that *Acacia nilotica* showed significant differences with *Alstonia scholaris*, *Melia azedarach*. *Alstonia scholaris* showed significant differences with

Cassia fistula, *Conocarpus erectus*, *Dalbergia sisso*, *Melia azedarach*, *Pongamia pinnata*, *Putranjiva roxburghii*, *Syzygium cumini*. *Cassia fistula* showed significant differences with *Dalbergia sisso*, *Melia azedarach*. *Conocarpus erectus* showed significant differences with *Melia azedarach*, *Syzygium cumini*.

Dalbergia sisso showed significant differences with *Mimusops eleng*, *Pongamia pinnata*, *Syzygium cumini*. *Melia azedarach* showed significant differences with *Mimusops elengi*. *Mimusops elengi* *Syzygium cumini*

Table 4. Differences of root vessel size across the species studied.

Species names	An	As	Cf	Ce	Ds	Ma	Me	Pp	Pr	Sc	Ec
An	--	62.540 18.953 0.040	62.540 35.826 0.120	62.540 36.783 0.130	62.540 80.615 0.272	62.540 33.466 0.101	62.540 18.077 0.037	62.540 30.669 0.081	62.540 26.709 0.064	62.540 24.944 0.057	62.540 26.586 0.060
As	----	---	18.953 35.826 0.017	18.953 36.783 0.013	18.953 80.615 0.003	18.953 33.466 0.011	18.953 18.077 0.598	18.953 30.669 0.037	18.953 26.709 0.001	18.953 24.944 0.018	18.953 26.586 0.095
Cf	-----	----	----	35.826 36.783 0.859	35.826 80.615 0.004	35.826 33.466 0.634	35.826 18.077 0.013	35.826 30.669 0.346	35.826 26.709 0.088	35.826 24.944 0.054	35.826 26.586 0.110
Ce	-----	-----	-----	----	36.783 80.615 0.005	36.783 33.466 0.496	36.783 18.077 0.009	36.783 30.669 0.261	36.783 26.709 0.062	36.783 24.944 0.037	36.783 26.586 0.079
Ds	-----	-----	-----	-----	-----	80.615 33.466 0.005	80.615 18.077 0.003	80.615 30.669 0.003	80.615 26.709 0.005	80.615 24.944 0.004	80.615 26.586 0.002
Ma	-----	-----	-----	-----	-----	----	33.466 18.077 0.007	33.466 30.669 0.552	33.466 26.709 0.094	33.466 24.944 0.050	33.466 26.586 0.162
Me	-----	-----	-----	-----	-----	-----	----	18.077 30.669 0.027	18.077 26.709 0.002	18.077 24.944 0.013	18.077 26.586 0.071
Pp	-----	-----	-----	-----	-----	-----	-----	----	30.669 26.709 0.335	30.669 24.944 0.196	30.669 26.586 0.417
Pr	-----	-----	-----	-----	-----	-----	-----	-----	----	26.709 24.944 0.370	26.709 26.586 0.973
Sc	-----	-----	-----	-----	-----	-----	-----	-----	-----	----	24.944 26.586

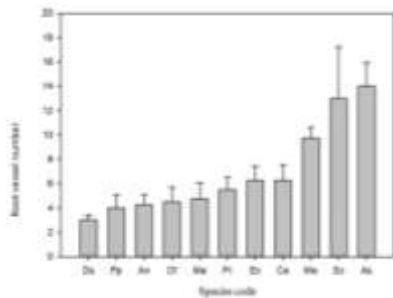
											0.666
Ec	----	----	----	----	----	----	----	----	----	----	----

Note: Bold number indicates the significant differences in the mean values $p < 0.05$.

Fig. 4. Root vessel number across species studied. Species codes are listed in Table 3.1.

Root vessel number across the species studied:

The root vessel number ranged from 3 to 14, the maximum was found in *Alstonia scholaris* while the lowest in *Dalbergia sisso*. The individual species mean values are shown in figure 4.4 and the comparison across species are shown in Table 4.4



It was found that *Acacia nilotica* showed significant differences with *Alstonia scholaris* and *Melia azedarach*. *Alstonia scholaris* showed significant differences with *Cassia fistula*, *Conocarpus erectus*, *Dalbergia sisso*, *Mimusops elengi*, *Pongamia pinnata*, *Putranjiva roxburghii* and *Eucalyptus citriodora*. *Cassia fistula* showed significant differences with *Mimusops elengi*. *Dalbergia sisso* showed significant differences with *Mimusops elengi*. *Melia azedarach* showed significant differences with *Mimusops elengi*. *Mimusops elengi* showed significant differences with *Pongamia pinnata*, *Putranjiva roxburghii*.

Table 5. Differences of root vessel number across the species studied.

Species names	An	As	Cf	Ce	Ds	Ma	Me	Pp	Pr	Sc	Ec
An	---	4.250 14.000 0.009	4.250 4.500 0.871	4.250 6.250 0.241	4.250 3.000 0.252	4.250 4.750 0.762	4.250 9.750 0.004	4.250 4.000 0.862	4.250 5.500 0.390	4.250 13.000 0.128	4.250 6.250 0.224
As	----	----	14.000 4.500 0.008	14.000 6.250 0.019	14.000 3.000 0.009	14.000 4.750 0.009	14.000 9.750 0.110	14.000 4.000 0.007	14.000 5.500 0.013	14.000 13.000 0.839	14.000 6.250 0.018
Cf	----	----	----	4.500 6.250 0.350	4.500 3.000 0.304	4.500 4.750 0.893	4.500 9.750 0.014	4.500 4.000 0.766	4.500 5.500 0.551	4.500 13.000 0.135	4.500 6.250 0.337
Ce	----	----	----	----	6.250 3.000 0.075	6.250 4.750 0.440	6.250 9.750 0.066	6.250 4.000 0.223	6.250 5.500 0.661	6.250 13.000 0.210	6.250 6.250 1.00
Ds	----	----	----	----	----	3.000 4.750	3.000 9.750	3.000 4.000	3.000 5.500	3.000 13.000	3.000

						0.280	0.002	0.437	0.091	0.098	6.250
											0.065
Ma	----	----	----	----	----	----	4.750 9.750	4.750 4.000	4.750 5.500	4.750 13.000	4.750 6.250 0.429
							0.023	0.675	0.671	0.144	
Me	----	----	----	----	----	----	----	9.750 4.000	9.750 5.500	9.750 13.000	9.750 6.250 0.057
								0.007	0.021	0.502	
Pp	----	----	----	----	----	----	----	----	4.000 5.500	4.000 13.000	4.000 6.250 0.210
									0.356	0.120	
Pr	----	----	----	----	----	----	----	----	----	5.500 13.000	5.500 6.250 0.651
										0.173	
Sc	----	----	----	----	----	----	----	----	----	----	13.000 6.250 0.209
Ec	----	----	----	----	----	----	----	----	----	----	----

Note: Bold number indicates the significant differences in the mean values $p < 0.05$.

stomata size across species studied: Stomata size ranged from 5.50µm to 11.93µm, the maximum was found in *Mimusops elengi* while the lowest in *Cassia fistula*. The individual species mean values are shown in figure 4.5 and the comparison across species are shown in Table 5.

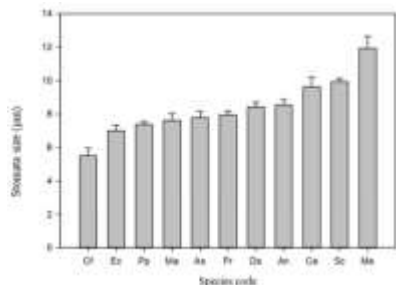


Fig. 5: Stomata size across species studied. Species codes are listed in Table 1.

It was found that *Acacia nilotica* showed significant differences with *Melia azedarach*,

pongamia pinnata, *Syzygium cumini* and *Eucalyptus citriodora*.

Alstonia scholaris showed significant differences with *Conocarpus erectus*, *Melia azedarach* and *Syzygium cumini*. *Cassia fistula* showed significant differences with *Conocarpus erectus* and *Melia azedarach*. *Conocarpus erectus* showed significant differences with *Melia azedarach*, *Mimusops elengi*, *Pongamia pinnata*, *Putranjiva roxburghii* and *Eucalyptus citriodora*. *Dalbergia sisso* showed significant differences with *Mimusops elengi*, *Pongamia pinnata*, *Syzygium cumini* and *Eucalyptus citriodora*. *Melia azedarach* showed significant differences with *Mimusops elengi* and *Syzygium cumini*. *Mimusops elengi* showed significant differences with *Pongamia pinnata*, *Putranjiva roxburghii* and *Eucalyptus citriodora* Table 4.5 showing the differences of stomata size across the species studied. Note: Bold number indicates the significant differences in the mean values $p < 0.05$.

Pongamia pinnata showed significant differences with *Syzygium cumini*. *Putranjiva roxburghii* showed significant differences with *Syzygium cumini*.

Syzygium cumini showed significant differences with *Eucalyptus citriodora*.

4.6 Stomata number across species studied: Stomata number ranged from 22.5 to 212, the maximum was found in *Alstonia scholaris* while the lowest in *Mimusops elengi*. The individual species mean values are shown in figure 4.6 and comparison across species are shown in Table 4.6

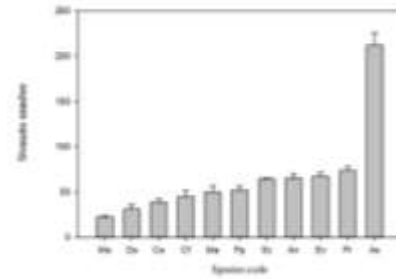


Fig. 4.6 Stomata number across species studied. Species codes are listed in Table 3.1.

It was found that *Acacia nilotica* showed significant differences with *Cassia fistula*, *Conocarpus erectus*, *Dalbergia sisso* and *Mimusops elengi*. *Cassia fistula* showed significant differences with *Melia azedarach*, with *Putranjiva roxburghii*, *Syzygium cumini* and *Eucalyptus citriodora*. *Conocarpus erectus* showed significant differences with *Melia azedarach*, with *Putranjiva roxburghii*, *Syzygium cumini* and *Eucalyptus citriodora*. *Dalbergia sisso* showed significant differences with *Putranjiva roxxburghii*.

Table 4.6 Showing the differences of stomata number across the species studied.

Species names	An	As	Cf	Ce	Ds	Ma	Me	Pp	Pr	Sc	Ec
An	--	65.500 212.00 0.306	65.500 44.750 0.037	65.500 38.500 0.005	65.500 31.000 0.003	65.500 49.500 0.105	65.500 22.500 0.002	65.500 52.000 0.250	65.500 73.250 0.289	65.500 64.250 0.814	65.500 67.000 0.837
As	----	---	212.00 44.750 0.294	212.00 38.500 0.291	212.00 31.000 0.287	212.00 49.500 0.297	212.00 22.500 0.282	212.00 52.000 0.298	212.00 73.250 0.310	212.00 64.250 0.305	212.00 67.000 0.306
Cf	-----	----	----	44.750 38.500 0.422	44.750 31.000 0.132	44.750 49.500 0.617	44.750 22.500 0.032	44.750 52.000 0.533	44.750 73.250 0.011	44.750 64.250 0.045	44.750 67.000 0.031
Ce	---	---	---	---	38.500 31.000 0.290	38.500 49.500 0.218	38.500 22.500 0.022	38.500 52.000 0.242	38.500 73.250 0.001	38.500 64.250 0.005	38.500 67.000 0.005
Ds	---	---	---	---	---	31.000 49.500 0.074	31.000 22.500 0.193	31.000 52.000 0.103	31.000 73.250 <0.001	31.000 64.250 0.006	31.000 67.000 0.002
Ma	---	---	---	---	---	---	49.500	49.500	49.500	49.500	49.500

							22.500	52.000	73.250	64.250	67.000
							0.0257	0.833	0.0310	0.114	0.087
Me	---	---	---	---	---	---	---	22.500	22.500	22.500	22.500
								52.000	76.250	64.250	67.000
								0.046	0.001	<0.001	0.002
Pp	---	---	---	---	---	---	---	---	52.000	52.000	22.500
									73.250	64.250	67.000
									0.098	0.270	0.212
Pr	---	---	---	---	---	---	---	---	---	73.250	73.250
										64.250	67.000
										0.146	0.401
Sc	---	---	---	---	---	---	---	---	---	---	64.250
											67.000
											0.634
Ec	---	---	---	---	---	---	---	---	---	---	---

Note: Bold number indicates the significant differences in the mean values $p < 0.05$

roxburghii, *Syzygium cumini* and *Eucalyptus citriodora*. *Melia azedarach* showed significant differences with *Mimusops elengi* and *Putranjiva roxburghii*. *Mimusops elengi* showed significant differences with *Pongamia pinnata*, *Putranjiva roxburghii*, *Syzygium cumini* and *Eucalyptus citriodora*.

4.7 Xylem theoretical hydraulic conductance (ki) for root and stem across species studied.

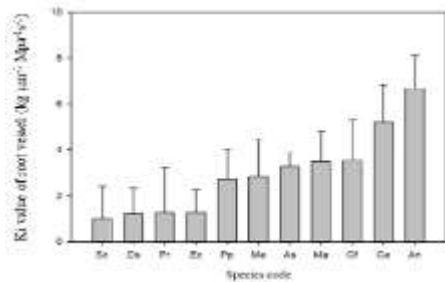


Fig 4.7 Species code and theoretical hydraulic conductance of root vessels ($\text{Kg } \mu\text{m}^{-1} \text{Mpa}^{-1} \text{s}^{-1}$). All values on Y-axis are with 10^{-11} power.

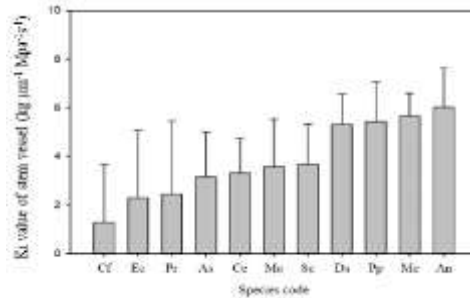


Fig 4.8 Species code and theoretical hydraulic conductance of stem vessels in ($\text{Kg } \mu\text{m}^{-1} \text{Mpa}^{-1} \text{s}^{-1}$). All values on Y-axis are with 10^{-11} power.

4.8 Relationship of stem xylem vessel with root xylem vessel: A positive relationship ($R^2 = 0.41$) was found between stem vessel number and root vessel number as shown in figure 4.10

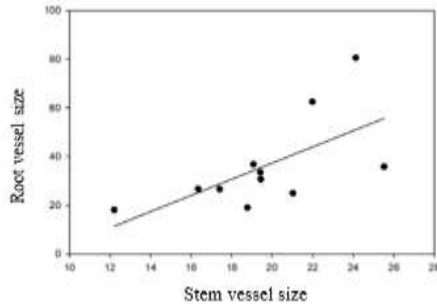


Fig 4.9: Relationship of stem xylem vessel with root xylem vessel. Each dot represents the individual specie.

4.9 Relationship of stem xylem vessel with root xylem vessel: A positive relationship ($R^2 = 0.23$) was found between stem vessel number and root vessel number as shown in figure 4.11.

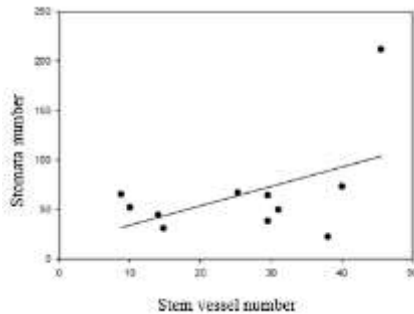


Fig 4.10 Showing linear relationship between stem vessel number and root vessel number.

4.9 Relationship between stem vessel number and root vessel number: A positive relationship ($R^2 = 0.48$) was found between stem vessel number and root vessel number as shown in figure 4.11

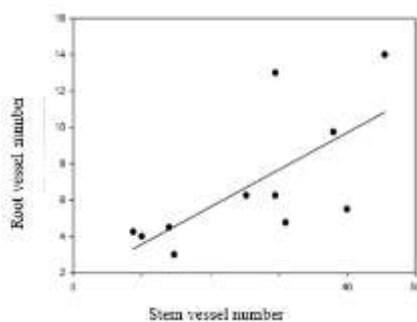


Fig 4.11 Showing linear relationship between stem vessel number and root vessel number.

Discussion

The study conducted on the 11 trees seedlings has provided some useful insights into the foliar, branch and

root anatomical features. The studied traits broadly reflect the hydraulic performance of species, which is reflected in terms of water supply through the xylem vessels from roots, to the stem xylem and finally at the leaf level (stomata) to release water and fix the carbon (Bussotti *et al.*, 2018). These traits are also useful to understand the hydraulic safety and efficiency among the trees (Johnson *et al.*, 2011; Meinzer *et al.*, 2009). As the trees with large diameter of vessel potentially have large theoretical conductivity per xylem vessels, but such vessels are more susceptible to embolism, such species need to maintain the constant supply of water through the roots which can help to avoid the embolism and will be requiring moist habitat or may reduce the path length. In our study the species with larger vessel diameter are exhibiting the larger xylem hydraulic conductivity supporting the previous studies (Choat *et al.*, 2008; Apgaua *et al.*, 2015). Further, we found the positive relationships between the stem vessel size and root vessel size, indicating the balance among the development of organs both above ground and below ground. But this finding contradicting with Tyree and Zimmermann (2002), reporting the more vulnerability of root vessels to drought as compared to stem vessels due to significantly large diameter of root vessels than the stem vessels. The species with small vessels and having more parenchyma cells also have the adaptation to avoid the drought induced embolism by utilizing the starch to release sugars which can move into the vessels helping the transportation of water due to osmotic potential (Carlquist, 2001). Such arrangement of cells also help the species for the recovery of embolism.

It was found that in some species the root xylem was wider than the stem. The resistance to the conductivity of water in the xylem is determined by the size of xylem conduit, as the conductivity in the xylem is proportional to the fourth power of conduit radius. The increase in xylem area leads to the increase in xylem hydraulic conductance. The large difference in the species xylem hydraulic conductance is because of the differences in their xylem sizes and also the length of xylem conduit length, although the vessel length can be correlated with the xylem diameter (Alder *et al.*, 1996; Kavanaugh *et al.*, 1999; Martínez-Vilalta & Pockman 2002). Such changes in xylem affect the efficiency of water transport. As the water absorbed by the deep roots have to be travelled from a long distance to reach the canopy leaves, than the water of shallow roots, the comparison of xylem anatomy at the different intervals of water path length may reflect some insights on the species differences and their adaptations to transport water to the canopy. The differences in the xylem size at the deep roots, surface roots and at the stem will be necessary to create the gradient for conduction of water.

The number of stomata and the size differed significantly across the study species mainly because the species respond differently even under the similar climatic conditions. As each species has evolved with the specific morphological traits through time. The meteorological conditions have the influence on plants as they force to adjust foliar and xylem plant traits (Tian *et al.*, 2016).

In our study the xylem hydraulic conductance was as: *Acacia nilotica* > *Mimusops elengi* > *Pongamia pinnata* > *Delbergia sisso* > *Syzygium cumini* > *Melia azadarach* > *Cnocarpus erectus* > *Alstonia scholaris* > *Putranjiva roxburghii* > *Eucalyptus citriodora* > *Cassia fistula*. It is important to note that the results reported

here for the xylem conductance and foliar anatomy are on the unit area basis, not on the whole tree xylem area or as some species can have low conductance but larger conducting area, and can have the high value of conductance when compared with the whole tree basis.

Conclusion:

The foliar, branch and root anatomical features can provide useful insights about the conductance of water in trees. Such kinds of traits provide approximate about the water conducting capacity and can help to plan the species for plantation in the specific areas.

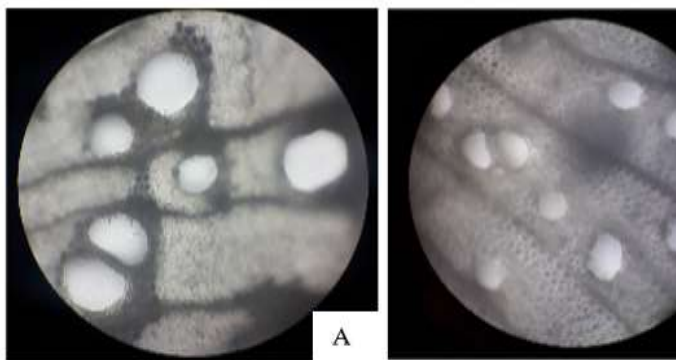


Plate 1: *Acacia nilotica*, A (Stem xylem vessels), B (Root xylem vessels), C (stomata)

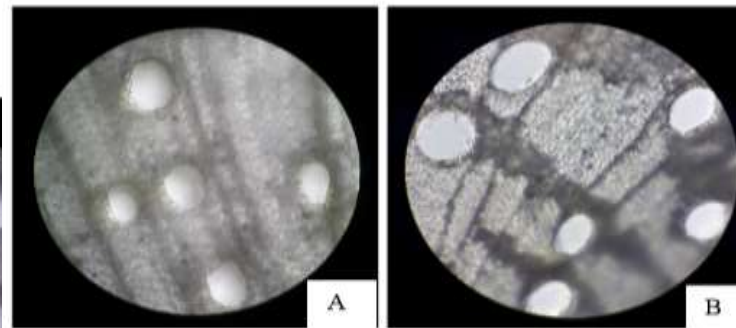


Plate 3: *Cassia fistula*, A (Stem xylem vessels), B (stomata)

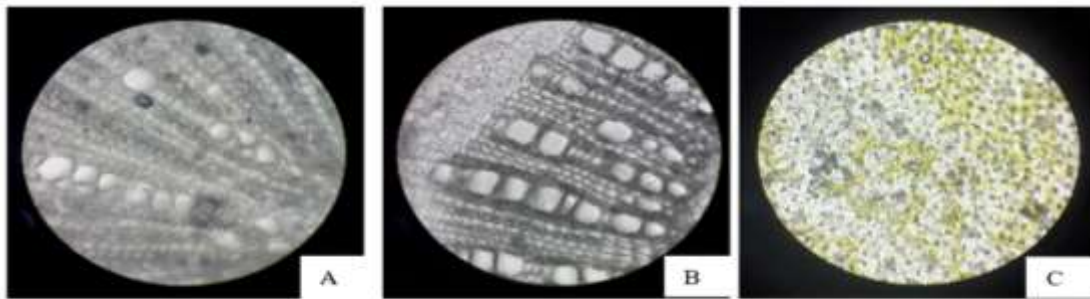


Plate 2: *Alstonia scholaris*, A (Stem xylem vessels), B (Root xylem vessels), C (stomata)

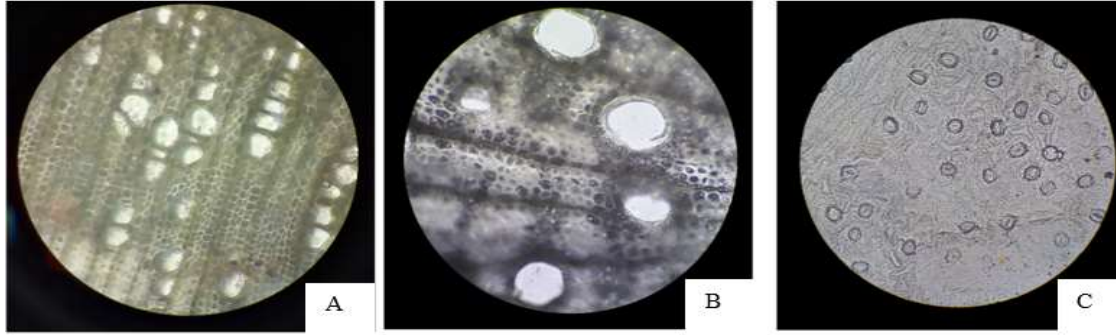


Plate 4: *Conocarpus erectus*, A (Stem xylem vessels), B (Root xylem vessels),
C (stomata)

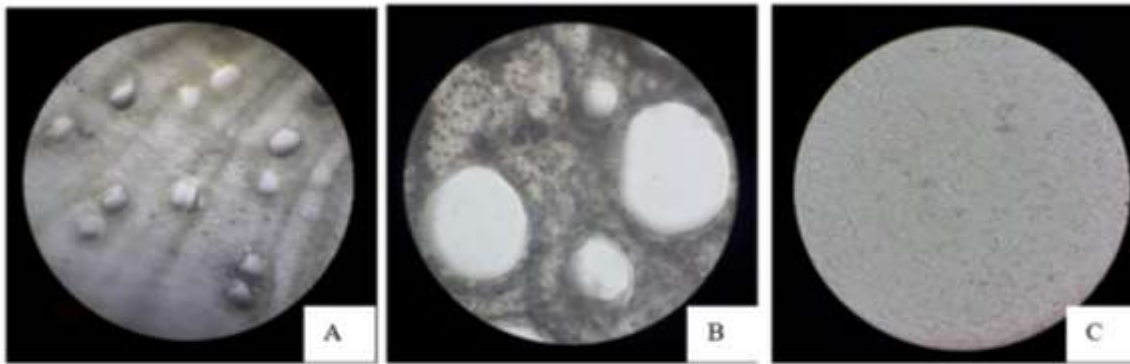


Plate 5: *Dalbergia sisso*, A (Stem xylem vessels), B (Root xylem vessels),
C (stomata)

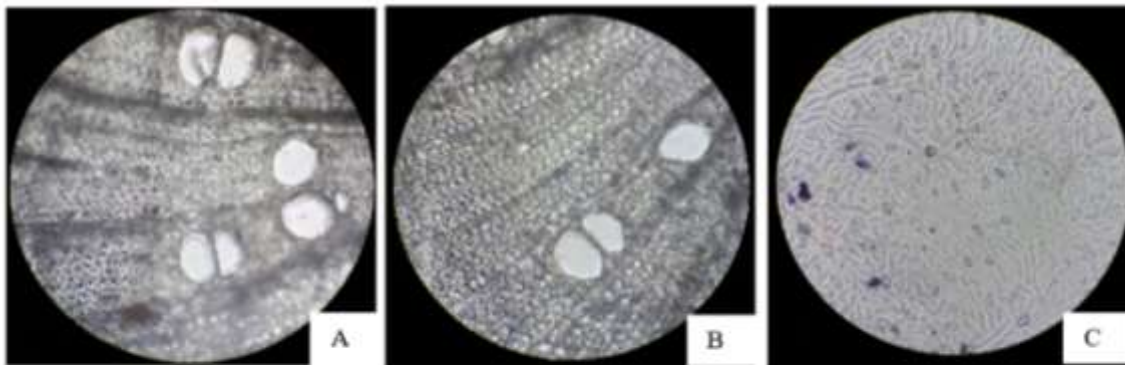


Plate 6: *Melia azedarach*, A (Stem xylem vessels), B (Root xylem vessels),
C (stomata)

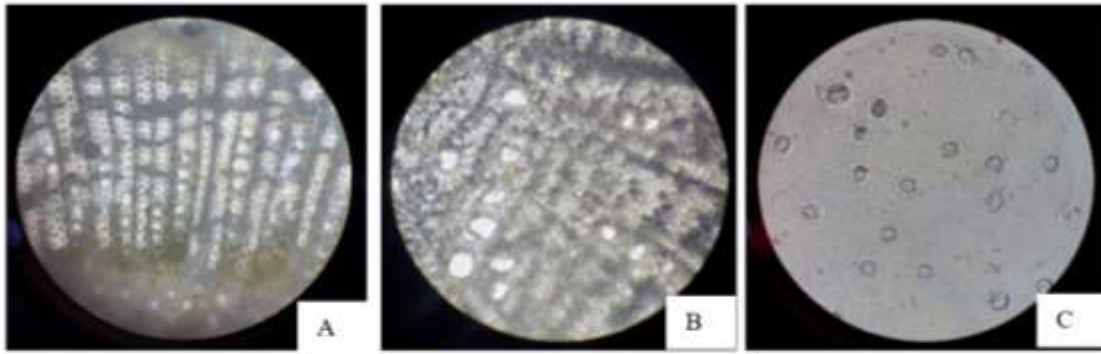


Plate 7: *Mimusops elengi*, A (Stem xylem vessels), B (Root xylem vessels), C (stomata)

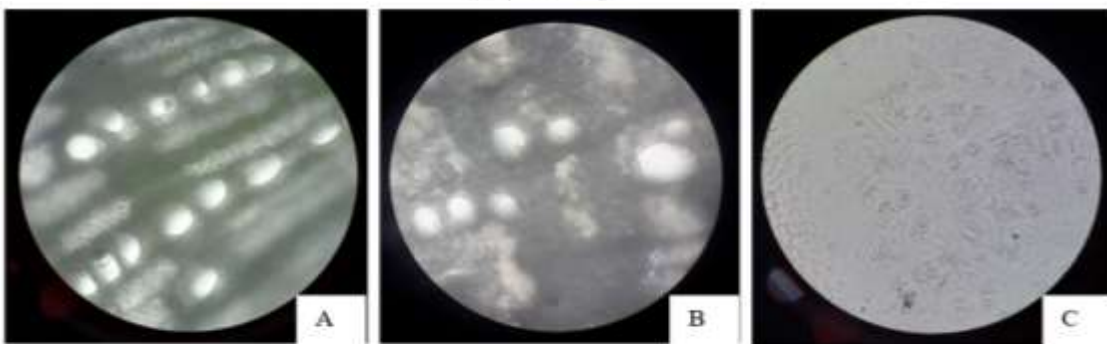


Plate 8: *Pongamia pinnata*, A (Stem xylem vessels), B (Root xylem vessels), C (stomata)

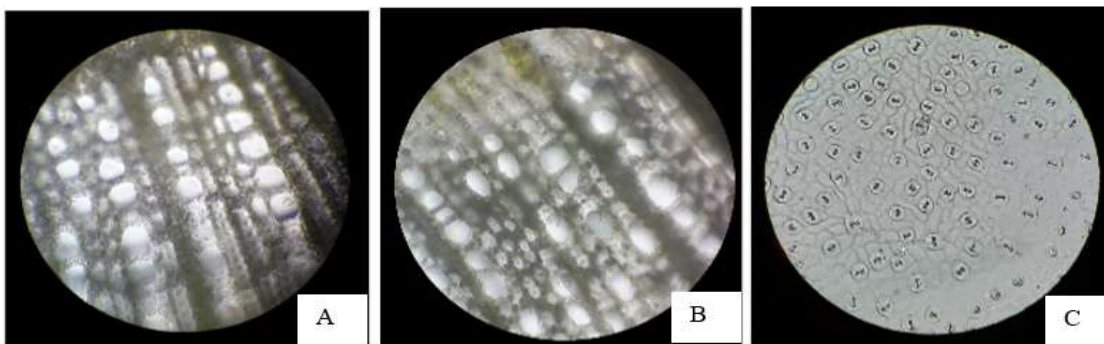


Plate 9: *Putranjiva roxburghii*, A (Stem xylem vessels), B (Root xylem vessels), C (stomata)

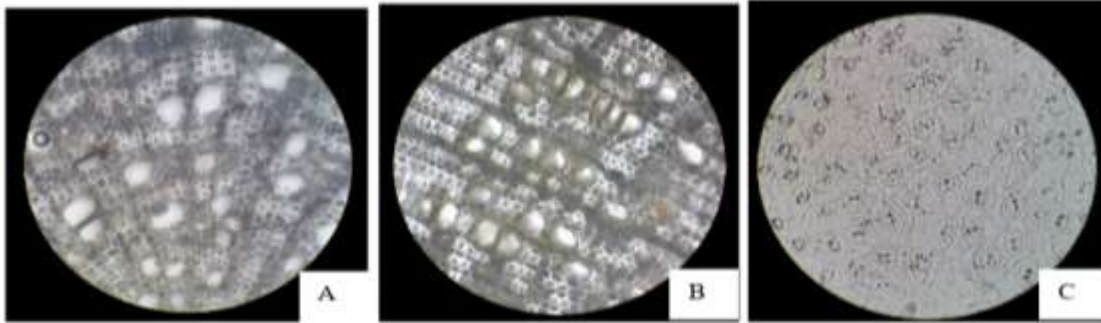


Plate 10: *Syzygium cumini*, A (Stem xylem vessels), B (Root xylem vessels), C (stomata)

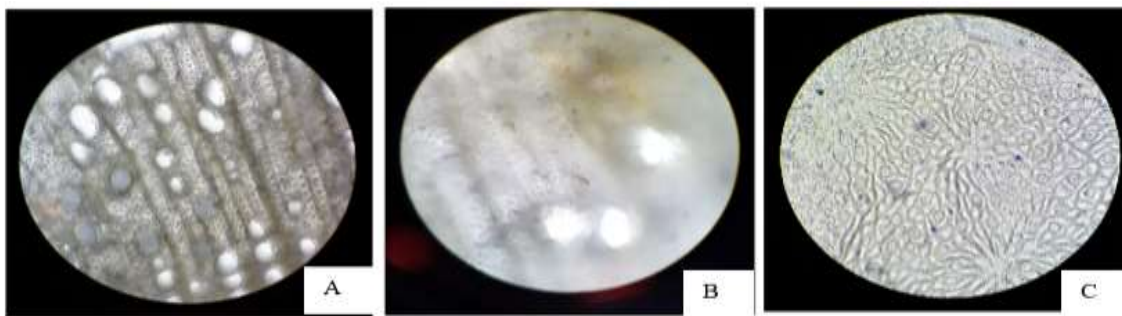


Plate 11: *Eucalyptus citriodora*, A (Stem xylem vessels), B (Root xylem vessels), C (stomata)

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