Leaf epidermal features as taxonomic characters in some *Lannea* spieces (*Anacardiaceae*) from Nigeria

Abdullahi Alanamu AbdulRahaman, Opeyemi Saheed Kolawole & Felix Ayotunde Oladele

[1] Applied Plant Anatomy and Wood Technology Laboratory, Department of Plant Biology, University of Ilorin, Ilorin, Nigeria, e-mail: abdulrahamanaa@unilorin.edu.ng (corresponding author)

[2] Department of Biological Sciences, Federal University, Kashere, Gombe State, Nigeria

Received: April 22, 2014 ▷ Accepted: October 21, 2014

Abstract. Leaf epidermal morphology of seven species of *Lannea* in the family *Anacardiaceae* were studied, namely: *L. kerstingii, L. welwitschii, L. schimperii, L. egregia, L. acida, L. microcarpa,* and *L. fruticosa*. The study revealed that several interesting characters, such as anticlinal cell wall pattern (*ACWP*), epidermal cell wall shape (*ECWS*), epidermal cell size (*ECS*), stomatal complex type (*SCT*), stomatal density (*SD*), stomatal size (*SS*), stomatal index (*SI*), and trichomes are constant and variable within and between some genera. *ACWP*s are straight, curved, round and undulate in the studied species. *ECWS* are found to be polygonal on both leaf surfaces, except in *L. fruticosa*. Stomata occur only on the abaxial leaf surface in all studied species; cyclocytic and anomocytic stomatal complex types are the only two types found, the anomocytic type occurring in five species: *Lannea kerstingii, L. welwitschii, L. schimperii, L. egregia,* and *L. acida.* Stomata are smaller in size in all species. Another important fact is that *SD* and *SI* vary from species to species. Out of the seven *Lannea* species, only three possess trichomes, namely *L. kerstingii, L. schimperii* and *L. fruticosa*. Thus the above-mentioned leaf features are of great taxonomic significance.

Key words: *Anacardiaceae, Lannea*, leaf anatomy, Nigeria, taxonomy

Introduction

Plant taxonomy as a concept denotes classification, naming and identification of numerous plants species. Therefore, plant taxonomy employs many methods in achieving this aim, namely morphology, chemistry, DNA markers, and anatomy. Leaf epidermis anatomy has been used repeatedly to classify and reclassify plants. Epidermal cells, such as trichomes, stomatal features and anticlinal cell wall patterns, are used. The epidermal cells and other appendages are less subjected to modification, so they serve as good taxonomic characters. Several taxonomists have used these epidermal features to delimit plants within genera and families (AbdulRahaman & Oladele 2003; Watson 2006; Van Wyk & al. 2008; Ren & al. 2007; Hardie 2009; AbdulRahaman & Oladele 2010a; Saheed & Illoh 2010).

Species of *Lannea* A. Rich. in Guillem are native to tropical Africa. Some *Lannea* species yield timber that is used locally; others are employed for a variety of purposes in indigenous medicine. The roots and bark are recorded to be used against diarrhea and for treatment of rachitic children and strained muscles. In the Côte d'Ivoire, the bark is used for treatment of diarrhea, oedema, paralysis, epilepsy, and insanity. In Mali, *L. velutina* is used in treatments of chest pain, gastric ulcer, wounds, skin diseases, respiratory tract diseases, and fever (Paulsen & Malterud 2014).

Some species of the genus *Lannea* in the family *Anacardiaceae* are selected for anatomical studies of their leaves. *Anacardiaceae*, the cashew family, includes approximately 800 species in 82 genera (Mitchell & Mori 1987). A family with such a large number of species should be subjected to rigorous taxonomic studies, so

as to ascertain their proper classification into groups and to avoid adulteration of samples. Members of the family are cultivated for their fresh leaves, seeds and fruits which are of tremendous importance.

Leaves of seven species of genus *Lannea* in the family *Anacardiaceae* were studied anatomically to elucidate their taxonomic potential by using such features like stomata, epidermal cells and trichomes to delimit the species within the genus and genera in the family.

Material and methods

Leaves of seven species of genus *Lannea* (Table 1) were collected for anatomical study. The plants were identified and deposited at the Herbarium of the Department of Plant Biology, University of Ilorin, Ilorin, Nigeria. Leaf cuticles were macerated in concentrated nitric acid or trioxonitrate (v) acid, rinsed in distilled water, stained in 1 % aqueous safranin solution and mounted in diluted glycerine (Dutta 2003). Parameters of the determined stomata were stomatal density, stomatal index and stomatal size (Franco 1939; Dilcher 1974), and frequency of each complex type was given as percentage occurrence of such complex type based on all occurrences (Obiremi & Oladele 2001). The epidermal cell size was determined as product of length and width of the cell on the basis of sample size of 35 samples. The index of similarity of the leaf surface followed the formula of Philips (1959). Statistical analysis of the data consisted of Analysis of Variance and Duncan Multiple Range Test (Bailey 1976; Duncan 1955).

Table 1. List of the studied plant species in the family *Anacardiaceae*.

Species	Place of collection
Lannea kerstingii **Engl. & K. Krause**	**Moro Local Government Area, Kwara State, Nigeria**
Lannea welwitschii **(Hiem) Engl.**	**Moro Local Government Area, Kwara State, Nigeria**
Lannea schimperi **(Hochst ex A. Rich) Engl.**	**Ila Orangun, Osun State, Nigeria**
Lannea egregia **Engl. & K. Krause**	**Ila Orangun, Osun State, Nigeria**
Lannea acida **A. Rich.**	**Ila Orangun, Osun State, Nigeria**
Lannea microcarpa **Engl. & K. Krause**	**Ila-Orangun, Osun State, Nigeria**
Lannea fruticosa **(Hochst ex A. Rich) Engl.**	**Ila-Orangun, Osun State, Nigeria**

Results

All seven studied species were hypostomatic. Two types of stomatal complexes were observed in the studied species – cyclocytic and anomocytic – which occurred with a frequency of 100. Anomocytic stomata were seen only in four species of *Lannea,* namely *L. kerstingii, L. welwitschii, L. scimperii* and *L. acida*. Stomatal density, stomatal index and stomatal size varied from one species to the other in the seven species of *Lannea*. Anatomical analysis of the leaf epidermal features revealed that *ACWP*s were round and straight in all studied *Lannea* species. *ECS*s were polygonal in all *Lannea* species, except in *L. fruticosa,* where they were isodiametric. ECWS also revealed certain variations between the *Lannea* species. The cells were smaller in the seven species. Trichomes occurred only in three *Lannea* species: *L. kerstingii, L. schimperii* and *L. fruticosa* (Tables 1 and 2; Fig. 1).

An indented dichotomous taxonomic key is provided for the purpose of delimiting the seven studied species on the basis of variations in their epidermal features:

1a Stomatal complex type cyclocytic 2

2a Trichomes absent; epidermal cell wall shape irregular or polygonal 3

3a Stomatal density 159.86 mm^{-2}; stomatal size 5.90 µm ..... *L. egragia*

3b Stomatal density 46.05 mm^{-2}; stomatal size 14.86 µm ..... *L. microcarpa*

2b Trichomes present; epidermal cell wall shape isodiametric ..... *L. fruticosa*

1b Stomatal complex types anomocytic 7

4a Trichomes present 8

5a Trichomes present on abaxial leaf surface only; stomatal density 72.56 mm^{-2}; stomatal size 12.00 µm; stomatal index 6.25 % .... *L. kerstingii*

5b Trichome present on both abaxial and adaxial leaf surfaces; stomatal density 114.60 mm^{-2}; stomatal density 8.96 µm; stomatal index 11.20 % ..... *L. schimperii*

4b Trichomes absent 6

6a Stomatal density 127.56 mm^{-2}; stomatal size 62.00 µm ..... *L. welwitschii*

6b Stomatal density 121.78 mm^{-2}; stomatal size 9.20 µm ..... *L. acida*

Table 2: Stomatal features and trichomes in the studied species.

		Stomata					
Species	**Leaf surface**	**Type**	**Frequency (%)**	**Density (mm-2)**	**Size (µm2)**	**Index (%)**	**Trichome**
Lannea kerstingii	Adaxial	–	–	–	–	–	Absent
	Abaxial	Anomocytic	100.00	72.56^{e}	12.00^{b}	6.25^{c}	Present
Lannea welwitschii	Adaxial	–	–	–	–	–	Absent
	Abaxial	Anomocytic	100.00	127.56^{c}	62.00^{a}	14.08^{a}	Absent
Lannea schimperii	Adaxial	–	–	–	–	–	Present
	Abaxial	Anomocytic	100.00	114.60^{d}	8.96^{c}	11.20^{b}	Present
Lannea egragia	Adaxial	–	–	–	–	–	Absent
	Abaxial	Cyclocytic	100.00	159.86^{a}	5.97^{d}	5.21^{c}	Absent
Lannea acida	Adaxial	–	–	–	–	–	Absent
	Abaxial	Anomocytic	100.00	121.78^{c}	9.20^{c}	12.00^{b}	Absent
Lannea microcarpa	Adaxial	–	–	–	–	–	Absent
	Abaxial	Cyclocytic	100.00	46.05^{f}	14.86^{b}	5.21^{c}	Absent
Lannea fruticosa	Adaxial	–	–	–	–	–	Absent
	Abaxial	Cyclocytic	100.00	148.09^{b}	9.16^{c}	13.19^{a}	Present

Mean values with the same letters do not differ significantly at $p < 0.05$.

Table 3: Leaf epidermal features in the studied species.

Species	**Leaf surface**	**Anticlinal cell wall pattern (*ACWP*)**	**Epidermal cell wall shape (*ECWS*)**	**Epidermal cell size (µm) (*ECS*)**
Lannea kerstingii	Adaxial	Round	Polygonal	8.22^{b}
	Abaxial	Round	Polygonal	5.36^{c}
Lannea welwitschii	Adaxial	Round	Polygonal	7.70^{bc}
	Abaxial	Straight	polygonal	7.43^{bc}
Lannea schimperii	Adaxial	Round	Polygonal	7.63^{bc}
	Abaxial	Round	Polygonal	6.52^{c}
Lannea egragia	Adaxial	Straight	Polygonal	9.23^{ab}
	Abaxial	Straight	Polygonal	8.65^{b}
Lannea acida	Adaxial	Straight	Polygonal	8.50^{b}
	Abaxial	Round	Polygonal	10.65^{a}
Lannea microcarpa	Adaxial	Straight	Polygonal	8.88^{b}
	Abaxial	Round	Polygonal	8.53^{b}
Lannea fruticosa	Adaxial	Straight	Isodiametric	10.73^{a}
	Abaxial	Straight	Isodiametric	9.69^{a}

Mean values with the same letters do not differ significantly at $p < 0.05$.

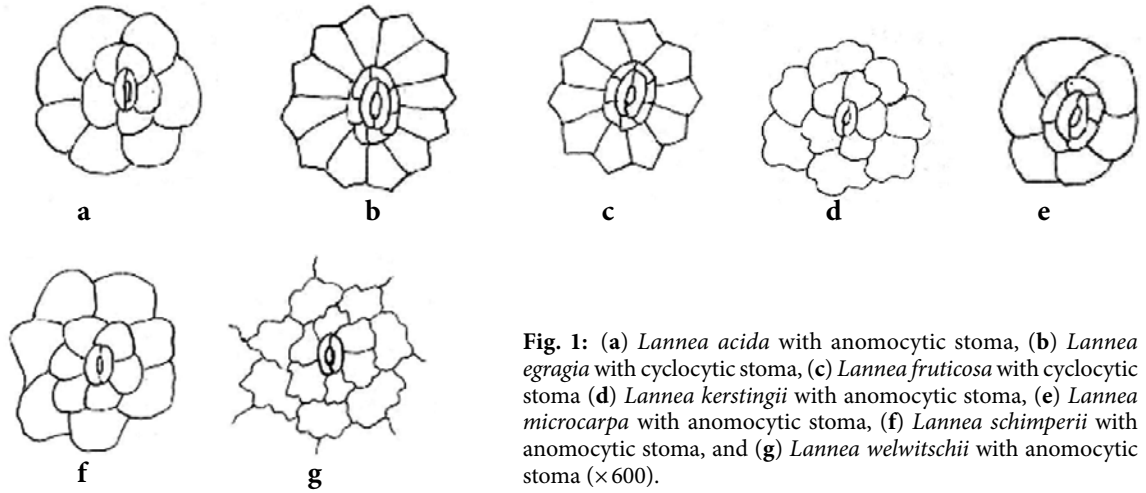

Fig. 1: (**a**) *Lannea acida* with anomocytic stoma, (**b**) *Lannea egragia* with cyclocytic stoma, (**c**) *Lannea fruticosa* with cyclocytic stoma (**d**) *Lannea kerstingii* with anomocytic stoma, (**e**) *Lannea microcarpa* with anomocytic stoma, (**f**) *Lannea schimperii* with anomocytic stoma, and (**g**) *Lannea welwitschii* with anomocytic stoma (×600).

Plant anatomy has been found to be very essential in plant taxonomy; the purpose is to develop a system of plants classification in a way that all the differences and similarities are set out in an ordered manner (Olorode 1984). In spite of the fact that vegetative and floral characters are markedly modified in relation to the habitat and pollination mechanisms, the summaries of character variation in Tables 2 and 3 indicate that the taxonomic application of diversity of epidermal morphology in the *Lannea* species cannot be overemphasized. The decision to choose epidermal characters for this study was prompted by the earlier studies that these characters represented genetic variations and have been used to solve some taxonomic problems in some other plant groups by the taxonomists (Srivastava 1978; Oladele 1990; Adegbite 1995; Nwockeocha 1996; Ogunkunle & Oladele 1997; Ogunkunle & Oladele 2008; AbdulRahaman & Oladele 2010a,b).

The results showed that the leaves of the studied species were hypostomatic (Figure 1). A similar phenomenon was observed earlier by AbdulRahaman (2009) in *E. milii*. *SCT* is exclusively cyclocytic in six species of *Anacardiaceae*, except in *L. kerstingii, L. welwitschii, L. schimpertii,* and *L. acida,* where it is anomocytic. Stomatal frequency varies from leaf to leaf and that was observed among the species in the three studied families. The homogeneous nature of *STC* on the leaves of some species of *Anacardiaceae* is responsible for 100 % occurrence of *SCT*s, where a single *SCT* was present on a leaf surface. *SD* was found to vary from one plant to another (Esau 1977). *SD* among the ten studied species revealed a significant difference: it was 46–148.09 mm^2 in seven *Lannea* species, and ranged from 177.63–320 mm^2 in the other three species (Table 3). *SD* was generally found to be higher on the abaxial surface of the leaves than on the adaxial. This pattern is in agreement with some earlier works (Oyeleke & al. 2004; AbdulRahaman 2009; AbdulRahaman & Oladele 2009; Saadu & al. 2009). According to Esau (1965), the stomata index varied in different parts of the leaves or in different leaves of same plants. *SI* and *SD* were markedly lower (ranging from 5.21–14.08 %) and smaller (ranging from 5.97–14.86µm in the *Lannea* species, except in *L. welwitschii,* where it was 62 µm), respectively (Table 3). Davies and Heywood (1963) considered stomatal size to be too variable as a diagnostic feature, while other researchers like Wilkinson (1971) had a contrary view. The latter indicated that stomatal size shows a much wider range in some taxa than in others, and that occasionally it could be useful as a diagnostic character, when dealing with taxa in which size ranges are restricted. The present study is in agreement with this observation of Wilkinson (1971).

Trichome is another significant taxonomic feature in the species of the genus *Lannea*. Trichomes are found in some species and absent in others. They are present in *L. kerstingii* and *L. fruticosa* only on the abaxial surface, and in *L. schimperii* on both leaf surfaces.

The above leaf epidermal features could be said to be taxonomically significant because of discontinuities that occurred within and between species and genera, as shown in Tables 2 and 3 and Fig. 1. This study clearly shows that micromorphological features are useful for identification of plants. Leaf epidermal features are of taxonomic significance in the species of genus *Lannea*; therefore, the species within this genus can be separated and distinguished on the basis of their stomata, epidermal cells and trichomes.

References

AbdulRahaman, A.A. 2009. Morphological and epidermal adaptations to water stress in some ornamental plant species. Ph.D. Thesis, University of Ilorin, Ilorin, Nigeria. (Unpubl.).

AbdulRahaman, A.A. & Oladele, F. A. 2003. Stomatal complex types, stomatal size, density and index in some vegetable species in Nigeria. – Nigerian J. Bot., **16**: 144-150.

AbdulRahaman, A.A. & Oladele, F.A. 2005. Stomata, trichomes and epidermal cells as diagnostic features in six species of the genus *Ocimum* L. (*Lamiaceae*). – Nigerian J. Bot., **18**: 214-223.

AbdulRahaman, A.A. & Oladele, F. A. 2009. Stomatal features and humidification potentials of *Borassus aethiopum, Oreodoxa regia* and *Cocos nucifera.* – African J. Pl. Sci., **3**(2): 059-063.

AbdulRahaman, A.A. & Oladele, F.A. 2010a. Leaf micromorphology of some *Amaranthus*. – Nigerian J. Pure Appl. Sci., **23**: 2136-2143.

AbdulRahaman, A.A. & Oladele, F.A. 2010b. Stomatal complex types and epidermal cells in *Jatropha* species L. (*Euphorbiaceae*). – Nigerian J. Pure Appl. Sci., **23**: 2160-2163.

Adegbite, A.E. 1995. Leaf epidermal studies in three Nigerian species of *Aspilia* (*Heliantheae – Asteraceae*) and two hybrids. – Nigerian J. Bot., **8:** 25-33.

Althawadi, A.M. & Grace, J. 1986. Water use by the desert cucurbit, *Citrullus colocynthus*. – Schrad De Cologia, **70**(3), 475-480.

Baily, N.T.J. 1976. Statistical Method in Biology. Cambridge Univ. Press, Cambridge.

Davis, P.A. & Heywood, V.H. 1963. Principle of Angiosperm Taxonomy. Oliver and Boyd, Edinburgh. pp. 210-230.

Dilcher, D.L. 1974. Approaches to the identification of angiosperm leaf remains. – Bot. Rev., **40**(1): 1-57.

Duncan, D.B. 1955. Multiple range and multiple t-test. – Biometrics, **77**: 1-42.

Dutta, A.C. 1979. Botany for Degree Students. 5th ed. Satiburiroa Gauhati. pp. 756-757.

Esau, K. 1965. Plant Anatomy. 2nd edition. John Wiley and Sons, Inc., New York PP 49-64.

Esau, K. 1977. Anatomy of Seed Plants (3rd ed). John Wiley and Sons, Inc., New York, pp 44-88.

Franco, C. 1939. Relation between chromosome number and stomata in *Coffea*. – Bot. Gaz., **100**: 817-823.

Hardie, J. 2009. Autumn leaves seen through herbivour eye. – Proc. Royal Soc. Biol. Sci., **276**: 121, doi: 1098/rspb.

Jeffrey, C. 1990. A new system of *Cucurbitaceae.* – Bot. Zhurn., **90**: 332-335.

Metcalfe, C.R. & Chalk, L. 1988. Anatomy of Dicotyledon (2nd ed). Oxford Unive. Press, Oxford. pp. 97-117.

Mitchell, J.D., & Mori, S.A. 1987. The cashew and its relatives (*Anacardium*: *Anacardiaceae*). –Mem. NY Bot. Gard., **42**: 1-76.

Nwokeocha, C.C. 1996. Foliar epidermal studies in *Oryza punctata*. – Nigerian J. Bot., **9**: 49-58.

Obiremi, E.O. & Oladele, F.A. (2001). Water conserving stomatal system in selected *Citrus* species. – S. African J. Bot., **67**: 258-260.

Ogunkunle, A.T.J. & Oladele, F A. 1997. Stomatal complex types in some Nigerian species of *Ocimum, Hyptis* and *Tinnea*. – Biosci. Res. Communic., **9**: 93-100.

Ogunkunle, A.T.J. & Oladele, F.A. 2008. Leaf epidermal studies in some Nigerian species of *Ficus* L. (*Moraceae*). – Pl. Syst. Evol., **274**: 209-221.

Oladele, F.A. 1990. Leaf epidermal features in *Vernonia amygdalina* and *V. cinerea.* – Nigerian J. Bot., **3**: 71-77.

Olorode, O. 1984. Taxonomy of West African Flowering Plants. Longman Publishing Company, New York. pp. 25-32.

Oyeleke, M.O., AbdulRahaman, A.A. & Oladele, F.A. 2004. Stomatal anatomy and transpiration rate in some afforestation tree species. – NISEB Journal, **4**(2): 83-90.

Paulsen, B.S. & Malterud, K.E. 2014. *Lannea velutina* A. Rich (*Anacardiaceae*). The Falculty of Mathematics and Natural Sciences, School of Pharmacy, Universitetet I Oslo. www.mn.uio.no/farmasi/english/projects/maliplants/medicinalplants. Retrieved on 19/04/2014.

Philips, E.A. 1959. Methods of Vegetation Study. Holt Publ. Company, New York. 44p.

Ren, B., Zhu, X-Y. & Jlang, Y-C. 2007. Systematic significance of leaf epidermal features in *Apios* and *Cochlianthus* (*Leguminosae*). – Acta Phytotax. Sin., **45**(3): 274-284.

Saadu, R.O., AbdulRahaman, A.A. & Oladele, F.A. 2009. Stomatal complex types and humidification potential of some root tuber species. – African J. Pl. Sci., **3**(5): 107-112.

Saheed, S.F. & Illoh, H.C. 2010. A taxonomic study of some species in *Cassiinae* (*Leguminosae*) using leaf epidermal characters. – Not. Bot. Hort. Agrobot., **38**(1): 21-27.

Shields, L.M. 1950. Leaf xeromorphy as related to physiological influences. – Bot. Rev., **16**: 399-447.

Stace, C.A. 1965. Cuticular studies as an aid to plant taxonomy. – Bull. Brit. Mus. (Nat. Hist.), Bot., **4**: 3-78.

Straburger, E. 1975. Textbook of Botany. Longman, London. pp. 101-185.

Srivastava, A. K. 1978. Study of leaf epidermis in the genus *Digitaria* Rich. (*Gramineae*). – J. Indian Bot. Soc., **57:** 155-160.

Van Wyk, A. E., Robbertse, P. J. & Kok, P. D. 2008. The genus *Eugenia* L. (*Myrtaceae*) in South Africa: the structure and taxonomic value of stomata. – Bot. J. Linn. Soc., **84**(1): 41-56.

Watson, L. 2006. The taxonomic significance of certain anatomical observation on *Ericaceae*. – New Phytol., **63**(3): 274-280.

Wilkinson, H. P. 1971. Leaf anatomy of various *Anacardiaceae* with reference to the epidermis. Ph.D. Thesis, University of London, London. (Unpublished).