Akinlabi AA and Oladipo OT (2021)
Notulae Scientia Biologicae
Volume 13, Issue 2, Article number 10760
DOI:10.15835/nsb13210760
Research Article

Venation studies of some species in the genus *Ficus* Linn. in Southwestern Nigeria

Adebisi A. AKINLABI*, Olaniran T. OLADIPO

Obafemi Awolowo University, Department of Botany, Ile – Ife, Osun State, Nigeria; akinlabiadebisi@yahoo.com (corresponding author); niranoladipo.to@gmail.com

Abstract

The present study investigates the venation of ten species of the genus *Ficus* collected from Obafemi Awolowo University Ile-Ife (latitude 7° 31' 14.7612" N and longitude 4° 31' 49.1340" E) and the NACGRAB, Ibadan, Nigeria (latitude 7°23′4″N and longitude 3°50′31″E). The leaf venations of the species were carried out using standard methods. All photomicrographs of the features were taken with the aid of Amscope digital camera mounted on a celesterone binocular microscope. All data were subjected of analysis of variance using SAS software. The result revealed the Leaf venation pattern based on areole shape, length and width, veinlets ending and trichomes. The leaf venation patterns of the species show that they are significant in identifying and delimiting studied species within the genus with respect to qualitative and quantitative data. Species specific variation were recorded for the venation patterns as areole shape, length and width, veinlets ending and trichomes and these features are either genetically fixed or as a result of environmental extremes. Presence of cystolith cells, trichomes and no veinlets ending is diagnostic of *Ficus mucuso*. The study concluded that venation patterns are therefore significant in delimitation of species in the genus *Ficus* and these characters can be employed as additional information in the existing taxonomical keys of the genus.

Keywords: areole; cystolith cells; leaf; shape; venation

Introduction

The genus *Ficus* belongs to the family Moraceae and also amongst the largest genera of higher plants (Scott, 1996; Frodin, 2004). *Ficus* is one of the most diverse woody plant genera globally (Berg, 1989; Chaudhary *et al*., 2012).

Ficus consist of about 1000 species distributed widely from pan-tropical and subtropical origins all over the world with great diversity in South East Asia, Malaysia and tropical South America (Bercu and Popoviciu, 2014; Teleb and Salah-El-Din, 2014). Berg (1989) discovered about 105 species in the African floristic region, out of which about 60 species of *Ficus* are found in West Tropical Africa (Burkill, 1998) and at least 44 species are seen in Nigeria (Keay, 1989).

The habits of the genus include deciduous and evergreen trees, shrubs, herbs, climbers and creepers and also life forms including free standing tree, epiphytes, semi-epiphytes in the crevices, rheophytes and lithophytes (Chaudhary *et al*., 2012; Rahman and Khanom, 2013; Mawa *et al*., 2013). Members of *Ficus* are

Received: 18 Jun 2020. Received in revised form: 02 Jun 2021. Accepted: 07 Jun 2021. Published online: 23 Jun 2021.
From Volume 13, Issue 1, 2021, Notulae Scientia Biologicae journal uses article numbers in place of the traditional method of continuous pagination through the volume. The journal will continue to appear quarterly, as before, with four annual numbers.

recognised by highly characterized inflorescence, the syconium or hypanthodium, which are hollow bags of numerous male and female flowers with fleshy receptacle, the male flowers are arranged towards an ostiole located at the free end of the receptacle, the remaining part of the cavity are occupied by female flowers. The female flowers are located close to the orifice or opening and are usually protected by sterile scales (Olorode 1984; Sharma, 1993; Clement and Wieblen, 2009).

During the last decades, plant anatomical study and its data are often used in characterization of species within a genus and in determining evolutionary relationships. Several authors have referred to plant taxonomy and application as a remarkable evolution in vascular plants studies (Stant, 1973; Metcalfe and Chalk, 1979; Forbes, 1980; Abubakar and Yunusa, 1998; Ogunkunle and Oladele, 2000; Ahmad *et al*., 2010; Odedeji and Adedeji, 2015; Talebi *et al*., 2017).

According to Sehgal and Paliwal (2008) and Ummu *et al*. (2014), anatomical studies of leaf venation have been proven to be useful for the identification of various species. Ummu *et al*. (2014) described the variation in the leaf venation of twenty-one species of *Ficus* in Peninsular Malaysia and observed eight leaf venation patterns. They also concluded that tracheid, cystolith cells and trichomes are anatomical characters that assist in delimiting the species. Siti-Khaulah and Noraini (2016) studied eight species of *Ficus*, the presence of complex veinlet, complete ultimate marginal and opened venation were observed. The report concluded that anatomical features of venation are of taxonomic significance in differentiating and identifying the studied species. Hickey (1973) indicated that the venations of leaves are associated with plant evolution and the significant role systematically in plant identification and classification. This study is to investigate the leaves venation patterns of ten species of *Ficus* identifying features that may be of taxonomic value in delimiting the species.

Materials and Methods

Herbarium Survey

A preliminary study of herbarium materials of the selected species of *Ficus* was carried out in Obafemi Awolowo University (IFE) herbarium and relevant literature were reviewed. The flora of West Tropical Africa by Hutchinson and Dalziel (1958) was also consulted.

Ten species of *Ficus* were collected at various locations in Southwestern Nigeria as shown in Table 1. The species of *Ficus* were authenticated at IFE herbarium, Obafemi Awolowo University, Ile-Ife. The Flora of West Tropical Africa by Hutchinson and Dalziel (1954-72) was also consulted. Fresh specimens collected at different collection sites were preserved in 50% ethanol.

For the venation studies, sizeable portions of the matured leaves of the species were obtained from the median parts of well expanded leaves. The leaves were decolourised by boiling in 90% ethanol (to remove chlorophyll) at 20 °C for about 10-15 minutes, washed in 3-4 changes of water to remove all traces of alcohol. The leaves were later boiled in 5% sodium hydroxide for 15 minutes to enhance further clearing of leaves.

The leaves were washed thoroughly to remove alkaline solution. The partially cleared leaves were further cleared in 5% domestic bleach (parozone) for 20-30 minutes. The cleared leaves were rewashed in 3-4 changes of water, and stored in 50% ethanol as described by Olatunji (1983). These were stained in 1% aqueous solution of Safranin O and mounted on a clean slide in 25% glycerol for venation studies. Slides examinations were made under the light microscope. Photomicrographs of veins and areoles were made using Amscope digital camera mounted on a celesterone binocular microscope.

Table 1. The sites of collection and the coordinates of the *Ficus* species studied

S/N	Species	Voucher Number	GPS coordinates	Description of collection sites
1.	*Ficus exasperata* Vahl	IFE- 17752	7°31′11″N4°31′38″E	Adjacent Central Science Laboratory, OAU, Ile-Ife
2.	*Ficus recurvata* De Wild	IFE- 17757	7°31′11″N4°31′34″E	Biological Garden, Behind Botany car park, OAU, Ile-Ife
3.	*Ficus mucuso* Welw. Ex Ficalho	IFE- 17755	7°31′13″N4°31′38″E	Biological Garden, Behind Botany car Park, OAU, Ile-Ife
4.	*Ficus sur* Forssk	IFE- 17758	7°31′4″N4°31′35″E	OAU Bus stop, Ile-Ife.
5.	*Ficus leprieurii* (Miq.) CC, Berg	1FE-17753	7°31′13″N4°31′38″E	Reforestation Garden, OAU, Ile-Ife
6.	*Ficus elastica* Roxb. Ex Hornen	IFE- 17751	7°31′21″N4°31′47″E	Parks and Garden, OAU, Ile-Ife.
7.	*Ficus benjamina* Vahl	IFE-17750	7°23′7″N3°50′28″E7° 31′16″N4°31′29″E	1.Moor Plantation, Apata, Ibadan 2.Opposite First bank lecture theatre, OAU
8.	*Ficus lutea* Vahl	IFE- 17754	7°31′11″N4°31′34″E	Biological Garden, Behind Botany car park, OAU, Ile- Ife
9.	*Ficus polita* Vahl	IFE- 17756	7°31′11″N4°31′34″E 7°23′4″N3°50′31″E 7°31′10″N4°31′29″E	1.Biological Garden, Behind Botany car park, OAU, Ile-Ife 2.Moor plantation, Apata, Ibadan 3.Beside OAU Library, Ile-Ife.
10.	*Ficus thonningii* Blume	IFE- 17759	7°31′11″N4°31′34″E	Biological Garden, Behind Botany car park, OAU, Ile-Ife

*Collectors' names: Akinlabi Adebisi A.

Statistical analysis

Data collected were subjected to analysis of variance based on completely randomized design to test for significant difference among the accessions of the ten species of *Ficus* studied. The means were separated using Duncan's Multiple Range Test (DMRT) and photographs of some of the morphological characters of the taxa were also taken. The data were analyzed using SAS software (2003).

Results

The leaf morphology of the ten *Ficus* species studied showed brochidodromous venation which is a type of camptodromous venation. The secondary veins of the leaf do not terminate at the margin but were joined together forming series of prominent arches (Table 2 and Figure 1).

Ficus leprieurii (Miq) CC. Berg

Areoles well developed. The shape of the areole is polygonal. Size ranges from 92.00 - 180.00 µm long and 76.00 - 116.00 µm wide. Veinlets ending is simple and are mostly branched, ranges from 0 - 6 in number.

Ficus thonningii Blume

Areoles are well developed and their shape ranges from rectangular to pentagonal, size ranges from 88.00 ± 3.20 µm wide and 116.80 ± 5.61 µm long. Veinlets ending are mostly linear and occasionally branched or forked, 0 - 3 per areole.

Ficus mucuso Welw. Ex Ficalho

Areole are well developed, shape varies from triangular to rectangular, 84.00 - 160.00 µm long and 60.00 - 120.00 µm wide. Silicified bodies with cellulose skeleton (Cystolith cells) and trichomes were present. It has no veinlets ending.

Ficus lutea Vahl

Areoles are developed with shape ranging from triangular to polygonal, about 88.00 - 148.00 µm long and 68.00 - 112.00 µm wide. Veinlets ending are simple, linear and occasionally branched and 0 - 3 veinlets per areole.

Ficus polita Vahl

Areoles are well developed, rectangular to triangular in shape. Shape ranges from 112.00 - 180.00 µm long to 64.00 - 128.00 µm wide. Veinlets ending are simple, linear and branched, 0 - 1 per areole.

Ficus sur Forssk

Areoles are well developed with shape largely triangular to polygonal. Areole size ranges from 56.00 - 120.00 µm long to 52.00 - 96.00 µm wide. Veinlets ending are simple and forked, 0 - 2 per areole.

Ficus benjamina Vahl

Areoles are well developed, shape vary from triangular, rectangular and quadrangular, about 60.00 - 100.00 µm long to 40.00 - 60.00 µm wide. Veinlets ending are simple, linear and mostly bifurcated. Veinlet ending ranges from 0 - 3 per areole.

Ficus exasperata Vahl

Areoles are well developed, largely rectangular to triangular. Areole length varies from 92.00 - 160.00 µm and areole width ranges from 60.00 - 116.00 µm. Veinlets ending are linear to branched and ranges from 0 - 3 per areole.

Ficus recurvata De Wild

Areoles are well developed; shape vary from rectangular to polygonal and size from 44.00 - 100.00µm long to 40.00 - 84.00 µm wide. Veinlets ending are simple, linear and branched and with about 0 - 3 per areole.

Ficus elastica Roxb. Ex Hornem

Areoles are well developed with shape ranging from triangular to pentagonal to polygonal. Areole size varies 84.00 - 192.00µm long and 64.00 - 180.00 µm wide. The veinlets ending are singly divided and ranges between 0 - 3 per areole.

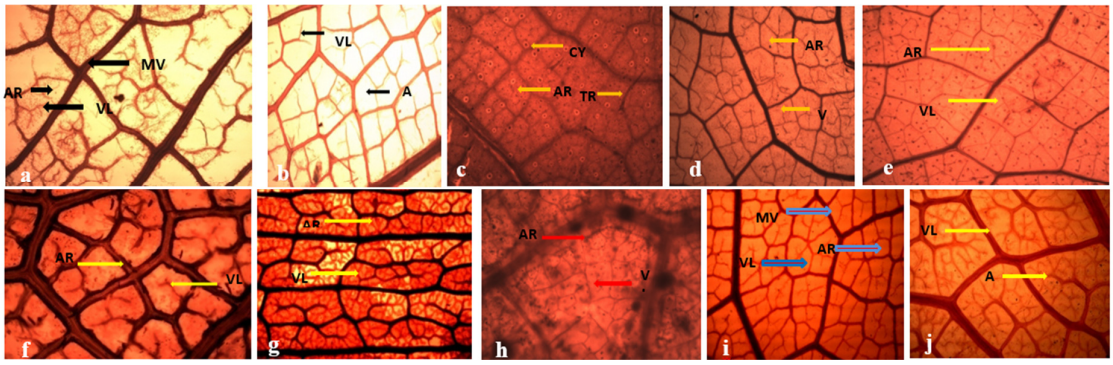

Figure 1. The venation patterns of the species studied
a) *F. leprieurii*; b) *F. thonningii*; c) *F. mucuso*; d) *F. lutea* e) *F. polita*; f) *F. sur*; g) *F. benjamina*; h) *F. exasparata;* i) *F. recurvata*; j) *F. elastica*
Legend: MV= Main Vein, VL=Veinlet, AR= Areole, CY=Cystolith, TR = Trichome

Table 2. Quantitative parameter of the venation pattern of ten *Ficus* species studied

Plant species	Areole width (µm)	Areole length (µm)	Number of veinlets ending
Ficus lutea	92.27±7.54^{ab}	120.00±4.84^{a}	0 - 3
F. thonningii	88.00±3.22^{b}	116.80±5.61^{a}	0 - 3
F. exasperata	87.47±3.84^{b}	120.00±3.84^{a}	0 - 3
F. mucuso	84.27±4.05^{b}	116.27±5.16^{a}	-
F.recurvata	54.40±4.15^{c}	70.40±4.38^{b}	0 - 3
F. leprieurii	107.00± 4.70^{a}	132.80±4.83^{a}	0 - 6
F. polita	96.53±3.84^{a}	130.67±3.88^{ab}	0 - 1
F. sur	68.53±3.29^{c}	82.67±5.20^{b}	0 - 2
F. elastica	103.2±9.54^{a}	131.47±8.76^{a}	0 - 3
F. benjamina	55.73±3.47^{b}	80.8±2.80^{c}	0 - 3

*Means with the same letter along columns are not significantly different at P ≤ 0.05

Discussion

Variations in patterns of leaf venation are not just useful in identifying taxon, but can also be used in differentiating between species of a genus. Rich diversity of venation patterns was observed in monocotyledons by Inamdar *et al.* (1983) and dicotyledonous plants by Hickey (1973). Areole shape in the *Ficus* species studied ranges from triangular, rectangular, and pentagonal to polygonal. Ogundipe and Wujek (2004) identified the significance of veinlet termination endings in the family Bignoniaceae. Characters of venation such as well-developed areole are classificatory for the species of *Ficus* studied. 0 - 3 veinlets ending were classificatory for *F. exasperata, F. benjamina, F. thonningii* and *F. lutea, F. elastica* and *F. recurvata. Ficus sur* has 0 - 2 veinlets ending, *Ficus polita* had 0 - 1 veinlets ending and *Ficus mucuso* had no veinlets ending. The 0 - 6 veinlets ending in *Ficus leprieurii* is diagnostic of the species.

Cystolith cells are silicified bodies with cellulose skeleton and trichomes were present only in *F. mucuso* and are diagnostic of the species. Siti-Khaulah and Noraini (2016) also reported the presence of cystolith cells in *F. pubigera* and opened areolar venation in most *Ficus* species studied but concluded that leaf venation has taxonomic significance in differentiation and identification of species. The cystolith cells are made of calcium carbonate located in lithocysts and occur in either papillate or hair-like form, usually found in the epidermis of leaves (Mauseth 1988; Ummu *et al.*, 2014). Also, as far back as 1950, Metcalfe and Chalk have reported the presence true cystoliths in some genera of Moraceae, such as *Broussonetia, Chlorophora, Conocephalus, Ficus*

and *Morus*. Consequently, the presence of cystoliths in *F. mucuso* is a common characteristic of the genus *Ficus* which is unique to certain species.

Also, the importance of trichomes have been emphasized in literatures (Ramayya and Rao, 1976; Rao and Ramayya, 1977; Adedeji *et al*., 2007). Among the ten *Ficus* species studied, trichomes were only present in *F. mucuso.* Various type of trichomes, such as straight and long, short and peltate have been observed on leaves in *Ficus* taxa according to Klimko and Truchan (2006). Ummu *et al.* (2014) also reported, simple and unicellular trichomes in the leaf venation of some species, such as in *F. aurantiacea* var. *aurantiacea, F. aurata, F. benghalensis, F. fulva, F. hispida, F. lepicarpa, F. sagittata* and *F. superba*.

The venation pattern revealed intraspecific and interspecific differences among the *Ficus* species studied based on quantitative attributes i.e the areole width and areole length. However, characters like long areole length is classificatory for species such as *F. polita, F. elastica, F. lutea, F. exasperata, F. mucuso, F. thonningii* and *F. leprieurii* having significantly longer areole length. This agrees with current sectional grouping of the species in Galoglychia excluding *F. exasperata* and *F. mucuso* in section Sycidium and Sycomorous respectively. *F. sur, F. benjamina and F. recurvata* have short areole length. The significantly wide areole in *F. leprieurii* and *F. elastica* with (107.00 ± 4.70μm and 103.2 ± 9.54μm) respectively is also classificatory of the species and this distinguishes them from other *Ficus* species and they are both in section galoglychia and it agrees with the sectional classification of Berg (1989).

Conclusions

The venation patterns in all the ten *Ficus* species studied are unique with respect to the areole shape, width and length as well as the veinlets ending. The study emphasized the taxonomic importance of leaf venation and its pattern usefulness in delimiting the species of the taxon. The study therefore concluded that the information will be useful in further taxonomic study of the genus *Ficus*.

Authors' Contributions

Both authors read and approved the final manuscript.

Acknowledgements

The authors are grateful to Mr. MS Akinropo, Dr. AJ Akinloye and Mr. A Omole of the Department of Botany, O.A.U Ile-Ife, for the kind gestures provided during the laboratory work.

Conflict of Interests

The authors declare that there are no conflicts of interest related to this article.

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