



## TAXONOMIC STUDIES OF MEMBERS OF THE FAMILY MORACEAE IN SELECTED AREAS OF BENUE STATE, NIGERIA

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### Abstract

A study was carried out to determine the taxonomic spread of the family Moraceae in selected areas of Benue State between May, 2012 and December, 2014. The selected areas are namely: Agan forest Reserve in Makurdi Local Government Area, Ikwe wildlife Resort in Gwer-east Local Government Area, and Leke Forest Reserve in Konshisha Local Government Area. Also, the taxonomic characters of the members of the family found were analysed to find out how these species are related to each other phylogenetically. Flowering patterns such as flowering time, flowering frequency, and flowering duration, and other morphological characters were observed and noted fortnightly for one year. Chemical composition of the Moraceae plant species was determined by phytochemical screening using standard qualitative methods, while determination of chromosome number was carried out by root tip squashing technique. Cluster analysis was carried out using Euclidean test to place the identified plants into a number of different groups such that similar plants were placed in the same group on the basis of the set of measured variables. A stepwise discriminate elimination of set of characters was employed to find out which combination of the three sets of characters as measured gave the best fit of relationship between the eight species against the backdrop of the established classifications. The phytochemical constituents and the chromosomal numbers in this study are considered to be more true taxonomic characters to be considered in the classification of these plants. The combination revealed that *Ficus ingens* and *Ficus exasperata* are more similar to each other than they are to *Ficus platyphylla* and *Ficus sur*, i.e. their connections (clades) are the closest links to the bottom (Euclidean distance = 96% and 93% respectively). In the same vein, *Ficus ingens*, *Ficus exasperata*, *Ficus platyphylla* and *Ficus sur* in the next upper clade (Euclidean distance = 89%) are more similar to each other than they are to *Ficus polita*, *Ficus trichopoda* and *Ficus thonningii* in the next upper clade (Euclidean distance = 82.5%). *Artocarpus heterophyllus* is completely separate from all the other seven species and is said to be simplicifolious (one-leaved). In conclusion, a combination of phytochemical and chromosomal attributes gave a better indication of the relationship between the 8 species of Moraceae in line with the established classification scheme for the family. Phenological events in the eight species did not appear to be important in the classification of the species.

**Keywords:** Clade, Cluster analysis, Euclidean distance, Moraceae, Phenological, phylogenetically, simplicifolious

## 1. INTRODUCTION

The Moraceae, often called the mulberry family or fig family, is a family of flowering plants comprising of monoecious or dioecious trees, shrubs, lianas, or rarely herbs which comprise of 40 genera and 1,000 species, nearly all with milky sap (Gill, 1988). The leaves are simple and alternate or rarely opposite. The stipules are small and lateral or sometimes they form a cap over the bud and leave a cylindrical scar. The flowers are unisexual and minute, and are usually densely aggregated. These aggregations frequently take the form of pendulous aments or catkins. Usually, the perianth consists of 4 or 5 undifferentiated sepals, but sometimes fewer or no perianth segments are present. A typical male flower has four stamens, one opposite each perianth segment. The female flowers have a bicarpellate pistil, generally with two styles, although one may be suppressed. The ovary is superior or inferior and contains a single pendulous ovule in a solitary locule. Fruit types include drupes and achenes that are often coalesced or otherwise aggregated into a multiple accessory fruit (Gill, 1988). Most are widespread in the tropical and sub-tropical regions, less so in temperate climates. In West Africa, it is represented by 12 genera and 110 species (Zerega *et al.*, 2005). In Benue State, Nigeria, the Moraceae family is reported as the second largest family after Caesalpinioideae (Akesa, 2010 and Anyam *et al.*, 2010).

A large number of these plants provide edible fruits. These include *Artocarpus heterophyllus*, *Ficus carica*, *Ficus glomerata*, *Morus alba*, and *Treculia africana*. Others are of medicinal importance. These include, *Ficus sycamorus*, *Ficus polita* and *Ficus ingens* (Gill, 1988). The bark of *Antiaria toxicaria* is used for making garments and sacks. Many species yield good timber. *Morus australis* is grown for its leaves which are fed to silkworms. Many *Ficus* species are grown as shade trees. The bark

of *Ficus nekbudu* serves as source of inutshu cloth (Gill, 1988). The wood of *Maclura aurantiaca* is suitable for making bows (Gill, 1988).

The monophyletism of the family Moraceae has been well supported in different studies but relationships within the family at the tribal level (genera and species) is still not clear (Zerega *et al.*, 2005). The family Moraceae displays an amazing array of diversity in inflorescence structures, pollination syndromes, breeding systems, floral characters, and growth forms. This diversity makes it an excellent group for addressing many intriguing evolutionary questions (Zerega *et al.*, 2005). Classification or grouping of the plants in the past was focused mainly on the morphology of the plants without involving other parameters. For this reason, there is the need to include other means by which plants can be distinguished such as cytological and or phytochemical parameters etc. which can help in giving a more clear-cut evolutionary relationship between the species rather than using only the morphological traits. Wendy and Weiblen (2009), suggested evaluation of Moraceae classification based on morphological analysis in combination with other parameters such as the use of cytological and phytochemical parameters. Earlier, Stace (1980), suggested phylogenetic studies at different localities for the purpose of clarity in taxonomic systems of Moraceae. At present, comprehensive information on the classification of the Family Moraceae which embraces taxonomic characteristics such as morphological, chromosomal, and phytochemical characteristics are lacking. The aim of this study is to determine taxonomic relationships between members of the family Moraceae found in some parts of the Benue Savanna.

## 2. MATERIALS AND METHODS

### 2.1 Plant Identification and Grouping

A floristic survey of the study sites was made and sample plant species belonging to the Family Moraceae were collected. The field assessments of the taxa were based on their general morphology. This was in line with the methods used by Cronquist (1988), Hutchinson (1926; 1959; and 1973), and Keay *et al.* (1989), who also used diagnostic keys which do offer description of the plants concerned, and state the essential diagnostic characters by means of which the taxa could be identified. They used the most conspicuous and clear-cut characters, without special regards to those considered taxonomically the most important. Both plants that were identified conclusively on the field and those not identified but suspected of belonging to the Family Moraceae were conveyed to the Botany laboratory of Benue State University, Makurdi, for identification and authentication. Mr. J.I. Waya of the Department of Biological Sciences, Benue State University, Makurdi, helped with the conclusive identification and authentication of the plant samples using taxonomic books such as FWTA, Trees of Nigeria by Keay *et al.* 1989. The plant stands identified as members of the Family Moraceae and earmarked for further studies for the purpose of this research work were tagged for easy identification and study on subsequent visits.

### 2.2 Determination of Flowering Behaviour

The plant stands identified as members of the Family Moraceae, earmarked and tagged for study were visited fortnightly during the study period for observations of their flowering behavior, which included flowering time, frequency and duration (adopted from Kamaljit *et al.*, 2003, and Anyam *et al.*, 2010). The seasons were categorized into four periods: (1) the first

wet season. (2) second wet season. (3) both-wet season, and (4) dry season. The first wet season lasts between April and June, the second wet season is between July and October, both wet seasons comprises the whole of the wet season, i.e. between April and October, and the forth period which is the dry season last from November to March.

The plant stands earmarked for study at the three sites were also observed to determine the flowering frequency. The members of Family Moraceae in flower were categorized into three levels: those that flower continually; those that were episodic; and those that flowered only annually. Continual species are those that flower more or less continually during the year, episodic species are those that flower for more than once during the year, and annual species comprise of species that flower only once during the year (adopted from Kamaljit *et al.*, 2003, and Anyam *et al.*, 2010).

Flowering duration was determined by counting the number of weeks that a species would take to flower per episode. That is, from the time the first flower buds are formed on the plant up to the time the last flower matures in a particular species.

### 2.3 Cytogenetical Studies of Members of Moraceae family.

Cytogenetical studies were carried out to determine the number and characterization of chromosome complements that are arrested at mitotic stage through a series of treatments. These studies were carried out to reveal the chromosomes of related species of plants that vary in several ways, which are collectively summed as karyotype. The details have been found extremely useful at all levels of the taxonomic hierarchy as well as resolving controversies on taxonomy and evolution of different taxa

Stem cuttings obtained from the study sites, of plant species belonging to the family Moraceae were used for propagation in polythene bags filled with

saw dust. The polythene bags were properly labeled with the names of plant species that they contained for reference purposes. The plants were watered twice a day (morning and evening) for three weeks. Fresh roots appeared within the said period.

The larger roots having firm, shiny tips with slight yellowish tinge which would give many cells suitable for study were selected. Roots having dull, brown or flaccid tips were avoided as they do not produce good cells. The root-tips were collected by removing the terminal (5-10mm) of the roots with fine pointed forceps. The root tips were collected between 08.00am and 11.00am, which is the time for highest active cell division according to Haskell (1986).

The harvested root-tips were transferred with forceps directly into test tubes nearly filled with Carnoy's fixative and stoppered to prevent evaporation or hydration. The test tubes were labelled with the names of the plant species. Root tips were fixed for a minimum of 3 hours, although, they could remain in the fixative up to 48 hours at room temperature. The root tips not used within 48 hours were prevented from deteriorating by preserving them for future use in a solution of 70% alcohol in tightly stoppered tubes and kept in the refrigerator at about 10°C. The fixed root tips immediately were then rinsed a couple of times in the ethyl alcohol to remove the acetic acid from the fixative. Acetic acid, if not removed, could reduce the stainability of the chromosomes. The acetic acid acts as neutralizer of stains.

Hydrolysis involves the maceration of the tissues in hot hydrochloric acid to soften the tissues or to hydrolyse deoxyribonucleic acid (DNA) of the chromosomes in the root-tips. The links between the cellulose walls of plant cells are broken down by the treatment with hydrochloric acid. This ensures that the stain can penetrate the cells and allows the

tissue to be squashed to spread out a layer one-celled thick under the cover slip on a glass slide.

Hydrolysis is often done prior to orcein staining. The fixative was decanted from the test tube containing the fixed root tips and was replaced with 10% HCL. The test tubes were then transferred to an oven preset at 60°C for 30 minutes after which hydrolysed root tips were transferred into specimen tubes and washed several times to remove the hydrochloric acid which otherwise would interfere with the staining process.

Formo-lacto-propiono-orcein stain is very useful in chromosomal studies as it is partly selective in its action and so enables chromosomes to be stained differentially from the other cell contents. The hydrolysed and washed root-tips were transferred into test tubes containing Formo-lacto-propiono-orcein stain and left for 30 minutes for staining. Root-tips left for longer periods in the stain became unusable owing to over-staining of the cytoplasm

The stained root tip was placed on a slide and viewed under a dissecting microscope. The end of the root- tip behind the root cap which is the region of active mitotic activity was isolated from the rest of the root tip using a sharp dissecting knife. Using mounting needles, the root tip was broken into small bits and flooded with one drop of Formo-lacto-propiono-orcein stain. It was then covered with a cover slip and placed on a paper towel which was then folded over the cover slip and pressed as hard as possible with the thumb. This forced the cells to separate from each other giving a single layer of cells making it possible to observe the cell inclusion together with the chromosomes at different stages of mitotic division.

The number of chromosomes per cell for each species of the Family Moraceae

studied was estimated from the mitotic stages studied. Efforts were made to as much as possible cover all the area covered by the root tip so as to observe as much as possible all the mitotic activity per root tip. A Carl-Zeiss Jena NU microscope equipped with an Olympus Camedia C 2000 Z camera was used for the study. This enabled observations to be made using the 100x oil immersion objectives for chromosomal counts and other chromosome characteristics.

#### 2.4 Phytochemical Studies of Moraceae

Fresh samples of some organs: leaves, stem bark, and roots of the plant species selected for the study were collected from each plant in June, 2013. These were taken to the Chemistry laboratory Benue State University, Makurdi, for phytochemical analyses. The samples were air dried at room temperature and each separately pulverized into powder by pounding using wooden mortar and pestle. After pounding, each sample was sieved using fine mesh. This gave a very fine powder of each sample which were used in the extraction of the phytochemicals using different solvents.

Phytochemical screening for major constituents was undertaken using standard qualitative methods adopted from Fadeyi *et al.* (1989), Odebiyi and Sofowora (1990), and Harborne, (1992), Abulude *et al.* (2001& 2004), and Abulude (2007). Tannins, saponins, phlobatannins, terpenoids, flavanoids, glycoside, anthraquinones, carotenoids, reducing sugars, alkaloids, and sterols tests were conducted using both the alcohol and the aqueous extracts.

#### 2.5 Cluster Analysis

Cluster analysis was carried out using Euclidean test (Computer software: GenStat Release 7.22 DE) to classify the

identified plants into a number of different groups such that similar plants were placed in the same group on the basis of the set of measured variables. In this study, the set of measured variables for the eight species of Moraceae family identified in the study area were phytochemical constituents, chromosomal numbers of the somatic cells, and phenological characters which included flowering time, frequency, and duration

### 3.0 RESULTS AND DISCUSSION

#### 3.1 Identification and Grouping of Moraceae Species

Table 1 illustrates the different species of the Family Moraceae found in the study areas, with their common and local names. A total of eight (8) species belonging to two tribes of the Family Moraceae were recorded in the study areas. The two tribes were Ficeae and Artocarpaceae. Of the eight (8) plant species of the Moraceae recorded, seven (7) belonged to the Tribe Ficeae. These were: *Ficus exasperata*, *Ficus ingens*, *Ficus platyphylla*, *Ficus polita*, *Ficus sur*, *Ficus thonningii*, *Ficus trichopoda*, and only one species belonged to the Tribe Artocarpaceae. This was *Artocarpus heterophyllus*. Out of the eight species found in the study areas, four of them were found in the wild including *Ficus exasperata*, *Ficus ingens*, *Ficus sur*, and *Ficus trichopoda*, while the other four were found in the homesteads including *Ficus platyphylla*, *Ficus polita*, *Ficus thonningii* and *Artocarpus heterophyllus*. Some of these species were reported before by several workers. In Plateau State, Nigeria (Offiah, *et al.*, 2011) reported *Ficus exasperata*, *Ficus trichopoda* and *Ficus platyphylla*. Also, in southern Nigeria reported two species, *Ficus exasperata* and *Ficus platyphylla* (Kadiri, *et al.*, 2008) In an Ethnobotanical Survey of Herbal Markets and Medicinal Plants in Lagos State recorded *Ficus sur*, *Ficus thonningii*, *Ficus trichopoda* (Kadiri,

*et al.*, 2008) Our result are in agreement with those found by (Aworinde, *et al.*, 2013) in Odeda area Southwestern Nigeria who reported plants grown and maintained in home gardens to include *Ficus platyphylla*, *Ficus polita*, *Ficus thonningii* and *Artocarpus heterophyllus*. In Taraba State of Nigeria, (Chapman and Chapman, 2001) reported several other species including *Ficus syncamorus*, *Ficus benjamina* and *Ficus rosa*. (Mbuya, 1994) identified useful trees and shrubs for Tanzania which include *Ficus sur*, *Ficus thonningii*, *Ficus trichopoda* *Ficus exasperata* and *Ficus platyphylla*.

### 3.2 Flowering pattern

Table 2 shows highest percentage of episodic flowering species followed by annual flowering species which is supported by the findings of Newstrom *et al.*,(1994b), who also found higher episodic flowering species than annual flowering species. There could be a disparity due to year to year variation in flowering phenology among tropical species in different regions of the world. Phenological phase changes such as the initiation of shoot growth or flowering of these tropical trees at specific times of the year are often thought to be triggered, i.e., to be controlled positively and directly, by the perception of an appropriate environmental cue (Ashton *et al.*, 1988, and Augspurger, 1982). According to this reasoning, any abrupt increase in the flowering frequency of a species is an indication that there is induction of flowering by some environmental cue. In the tropical forest the most likely environmental changes controlling the periodicity of tree growth and flowering are the first heavy rainfalls after a period

of severe drought or cessation of rains. In strongly desiccated, leafless trees, heavy rains cause rehydration and the opening of resting flower buds (Borchert,1994c). Synchronized flowering on new shoots at the start of the rainy season thus indicates rain induced shoot growth accompanied by the formation of lateral flowers. Flower formation on such shoots appears to be the indirect, endogenously controlled consequence of flushing and not a response to a specific environmental flowering cue (Borchert, 1994c). Table 3 shows that plant species of Moraceae in the study area flower in different months of the year, vis-à-vis in different seasons. These findings are in line with those of Opler *et al.* (1980), Akesa (2010), and Anyam *et al.*(2010), and Newstrom *et al.*(1994, a and b) who also found that woody plants flower in different months of the year and Frankie *et al.* (1974) who reported that woody plants flower in different seasons. The highest flowering activity occurred in the dry season followed by the flowering in the first wet season, and the lowest flowering activity occurred in the both-wet season of the year. This is in line with Singh and Kushwaha (2006),who also found that woody plants flower in the dry season extending into the first wet season. They concluded that this flowering periodicity has evolved as an adaptation to an annual leafless period and the time required for the fruit to develop. The direct relationship between leafless period and time lag between onset of vegetative and reproductive phases reflects the partitioning of resource use for supporting these phases.

**Table 1:** Species of the Family Moraceae found in the study areas, with their common and local names.

<b>Tribe</b>	<b>Species</b>	<b>Common name</b>	<b>Local names</b>
<b>Ficeae</b>	<i>Ficus exasperata</i> Vahl	Sandpaper fig/Toothbrush fig	Hitur (Tiv), Uli/Ikpi (Idoma), Adundu okulokulo (Etulo), Uhuo/Uho (Igede).
	<i>Ficus ingens</i> (Miq.) Miq	Homestead fig	Hon-tur (Tiv), Adundu (Etulo), Kawuri (Hausa).
	<i>Ficus platyphylla</i> Del	Gutta-percha tree	Ikondo-tor (Tiv), Ogo (Idoma), Ugu ese/Afe mni (Etulo), Gamji (Hausa).
	<i>Ficus polita</i> Vahl	Homestead fig	Kondam/Mua (Tiv), Oda (Idoma), Mku-ozu/Afaya (Etulo), Durumi (Hausa).
	<i>Ficus sur</i> Forssk	Cape fig/ Broom cluster fig/Bush fig/wild fig	Tur (Tiv), Okoklodu (Idoma), Andundu (Etulo), Obwo (Igede), Dullu/Fabrinbaure (Hausa).
	<i>Ficus trichopoda</i> Baker	Stilt-root fig	Po (Tiv), Ugu (Etulo), Bauren kiyashi (Hausa).
	<i>Ficus thonningii</i> Blume	Dome-crowned fig	Akinde (Tiv), Oda (Idoma), Mku-ozu (Etulo), Uvo (Igede), Shiriya/Guluba/Chediya (Hausa).
<b>Artocarpae</b>	<i>Artocarpus Heterophyllus</i> Lam	Jackfruit	Ahi-uke (Tiv).

**Table 2: Seasonal patterns in flowering species of Moraceae family in Benue State**

Species	Seasons			
	First wet	Second wet	Both wet	Dry
<i>Ficus exasperate</i>	1	0	0	1
<i>Ficus ingens</i>	0	0	0	1
<i>Ficus platyphylla</i>	1	1	1	1
<i>Ficus polita</i>	0	0	0	1
<i>Ficus sur</i>	1	0	0	1
<i>Ficus thonningii</i>	1	1	1	1
<i>Ficus trichopoda</i>	0	1	0	1
<i>Artocarpus heterophyllus</i>	0	0	0	1
<b>Total</b>	<b>4</b>	<b>3</b>	<b>2</b>	<b>8</b>

**Table 2: Flowering Levels of plant species of Moraceae family in Benue State**

Species	Category of Flowering		
	Annual	Episodic	Continual
<i>Ficus exasperata</i>	0	1	0
<i>Ficus ingens</i>	1	0	0
<i>Ficus platyphylla</i>	0	1	0
<i>Ficus polita</i>	1	0	0



<i>Ficus sur</i>	0	1	0
<i>Ficus thonningii</i>	0	1	0
<i>Ficus trichopoda</i>	0	1	0
<i>Artocarpus heterophyllus</i>	1	0	0
<b>Total</b>	<b>3</b>	<b>5</b>	<b>0</b>
<b>% Total</b>	<b>37.5</b>	<b>62.5</b>	<b>0.0</b>

### 3.3 Phytochemical composition of members of the Family Moraceae

Table 4 illustrates the phytochemical composition of members of the Family Moraceae in Benue State, Nigeria. Both tannins and sterols were contained in all the 8 plant species of Moraceae, whereas saponins and phlobatannins were contained in only 7 of them. None of the plant species contained either alkaloids or carotenoids. Using only aqueous extract, the highest number of plant species (7) contain tannins in their leaves followed by six species containing tannins in their stem bark. When only alcohol extract was used, all the eight plant species under study showed presence of sterol in both leaves, stem bark, and roots followed by seven species containing saponins in both leaves, stem bark, and roots. Tannins, however, were contained in seven species only in the stem bark and roots (6 species in the leaves). All the eight plant species under study showed presence of neither alkaloids nor carotenoids using both aqueous extract and alcohol extract. There was no association between phytochemical composition by species and the type of extract used, at  $p = 0.05$ , when chi – square ( $X^2$ ) analysis was conducted. Also, there was no association between phytochemical composition by species and plant parts.

### 3.4 Chromosomal number of plant species belonging to Moraceae Family

Table 5 illustrates the Chromosomal number of plant species belonging to Moraceae Family in Benue state. Number of chromosomes varied among plant species of the Family Moraceae. Both *Ficus thonningii* and *Artocarpus heterophyllus* had the highest chromosomal number of  $2n = 28$  (i.e. basic number of 14). This was followed by *Ficus exasperata*, *Ficus ingens*, *Ficus platyphylla*, *Ficus polita*, *Ficus trichopoda*, with chromosomal number of  $2n = 26$  (i.e. basic number of 13). *Ficus sur* had the lowest chromosomal number of  $2n = 18$  (i.e. basic number of 9). Several cytogenetic studies established that the species of family Moraceae have a base number of  $x = 13$  or  $14$  (Condit, 1928 & 1933; Bawa, 1973). Compared to the base number, the chromosome counts in the present study indicated that these identified species were not polyploids, they were all diploids. The findings of the present study are also in line with Pimienta (1995), whose review indicated diploidy as a common phenomenon within the family Moraceae. In this study, the basic number of  $x = 9$  was obtained in *Ficus sur*. This finding is contrary to the findings of Condit (1928 & 1933) and Bawa (1973) who found the basic number  $x$  of 13 and 14. Several authors have reported variations in chromosomal numbers in plants of the same kind that are vegetatively propagated

**Table 4: Plant species of Moraceae containing different phytochemicals (using both Aqueous and Alcohol extract for both roots, stem-bark, and leaves).**

Plant species	Phytochemical											
	Sap	Tan	Fla	C.gl	Alk	Car	C.an	F.an	Phl	R.su	Ste	Ter
<i>Ficus exasperata</i>	+	+	+	+	-	-	-	-	+	-	+	-
<i>Ficus ingens</i>	+	+	-	-	-	-	+	+	+	-	+	-
<i>Ficus platyphylla</i>	+	+	-	-	-	-	-	-	+	-	+	-
<i>Ficus polita</i>	+	+	-	-	-	-	-	-	+	-	+	+
<i>Ficus sur</i>	+	+	-	-	-	-	+	+	+	-	+	-
<i>Ficus thonningii</i>	+	+	-	-	-	-	-	-	+	-	+	-
<i>Ficus trichopoda</i>	+	+	-	-	-	-	-	-	+	-	+	+
<i>Artocarpus heterophyllus</i>	-	+	+	-	-	-	-	-	-	+	+	+
<b>Total</b>	<b>7</b>	<b>8</b>	<b>2</b>	<b>1</b>	<b>0</b>	<b>0</b>	<b>2</b>	<b>2</b>	<b>7</b>	<b>1</b>	<b>8</b>	<b>3</b>

**Key**

+ = present

- = absent

Sap = Saponnins

Tan = Tannins

Fla = Flavonoids

C.gl = Cardiac glycosides

Alk = Alkaloids

Car = Carotenoids

C.an = Combined anthocyanins

F.an = Free anthocyanins

Phl = Phlobatannins

R.su = Reducing sugar

Ste = Sterols

Ter = Terpenoids

**Table 5: Chromosomal number of plant species belonging to Moraceae Family in Benue state.**

S/No	Plant Species	Chromosomal number
1	<i>Ficus exasperata</i>	2n =26
2	<i>Ficus ingens</i>	2n =26
3	<i>Ficus platyphylla</i>	2n =26
4	<i>Ficus polita</i>	2n =26
5	<i>Ficus sur</i>	2n =18
6	<i>Ficus thonningii</i>	2n =28
7	<i>Ficus trichopoda</i>	2n =26
8	<i>Artocarpus heterophyllus</i>	2n =28

**Cluster analysis on the basis of the set of measured variables**

Fig. 1 is the result of Cluster analysis using Euclidean test using a set of measured variables including both phytochemical constituents, chromosomal numbers, and phenological characters (Flowering time, Flowering frequency, and Flowering duration ) placed *Ficus sur* completely separate from the other *Ficus* species but

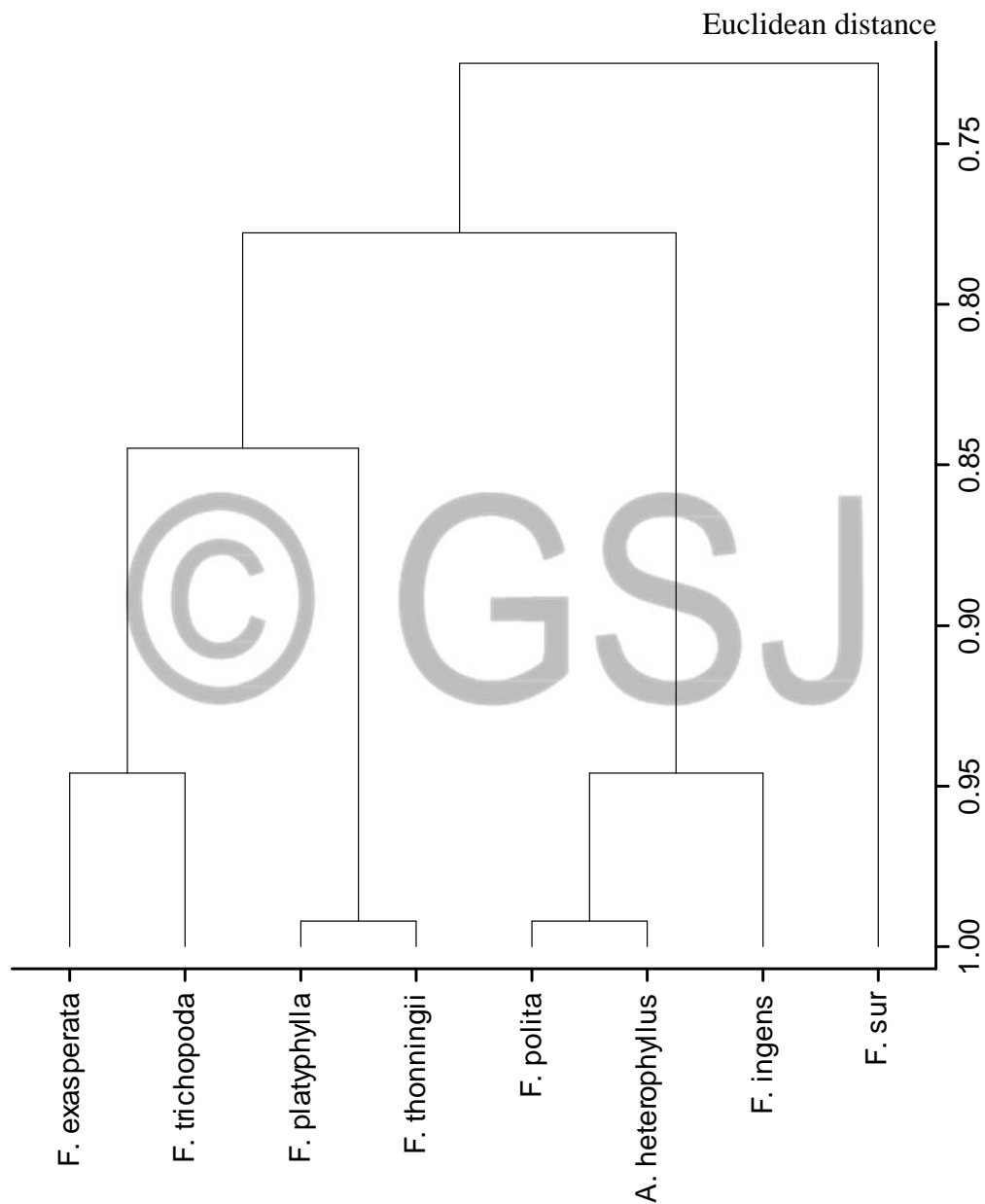
included *Artocarpus heterophyllus* in the same clade with the *Ficus* species indicating that *Artocarpus heterophyllus* is included in the same tribe (Ficeae) with the *Ficus* species and *Ficus sur* excluded from the tribe Ficeae. This finding was remarkably different from the findings of previous studies including Berg (2001), Berg (2005a), and Datwyler and Weiblen (2004). This sharp distinction in the relationship with the previously established ones, prompted a stepwise

elimination of the measured variables (characters) to ascertain the ones that are true taxonomic characters of the species under study.

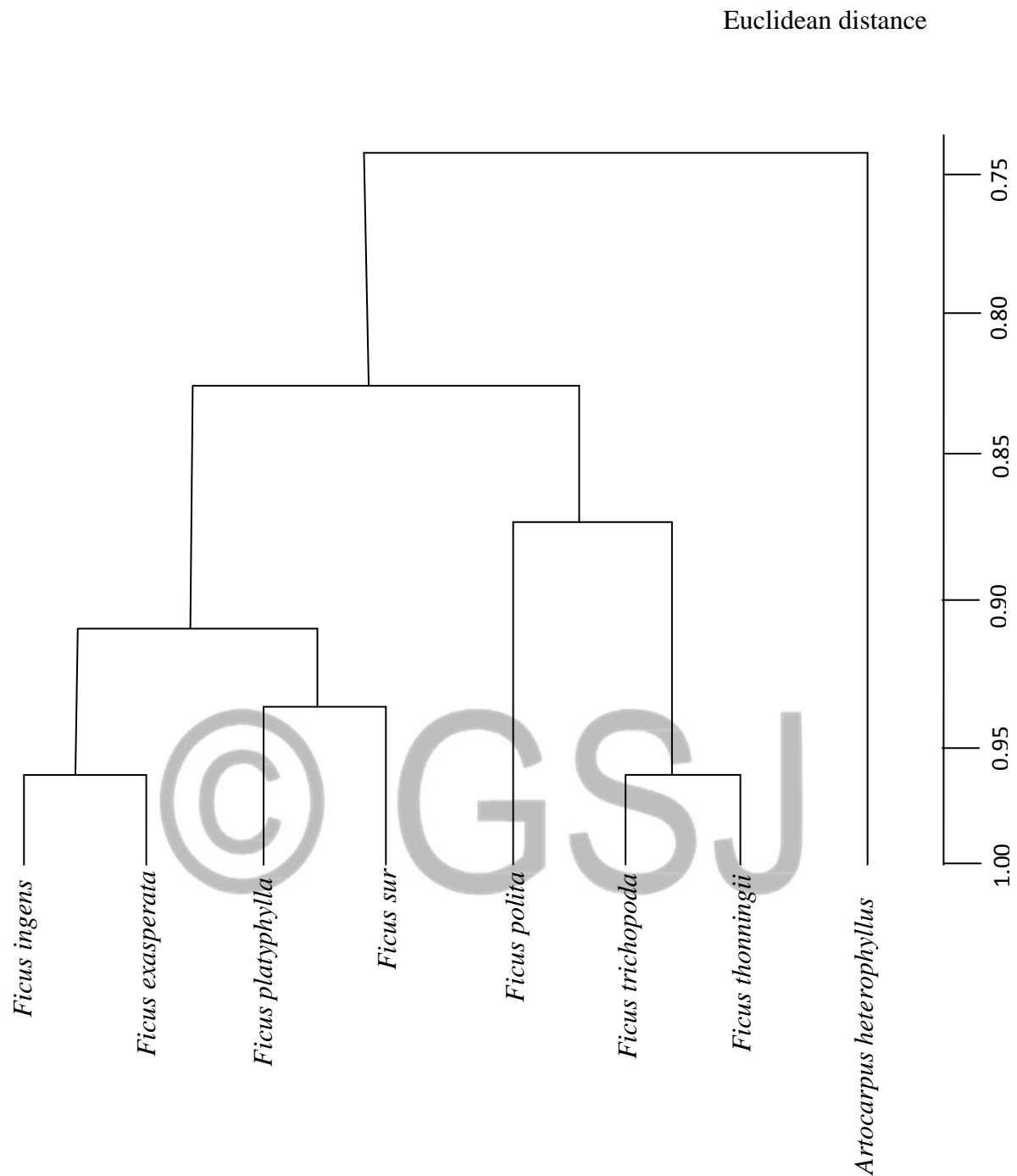
In fig. 2, the phenological characters were eliminated from the cluster analysis leaving only the phytochemical constituents and the chromosomal numbers for the analysis. This cluster analysis depicted the true picture of the taxonomic relationship among the species of Moraceae under study as established by Berg (2001), Berg (2005a), and Datwyler and Weiblen (2004). The cluster analysis revealed that *Ficus ingens* and *Ficus exasperata* are more similar to each other than they are to *Ficus platyphylla* and *Ficus sur*, i.e. their connections (clades) are the closest links to the bottom (Euclidean distance = 96% and 93% respectively). In a same vein, *Ficus ingens*, *Ficus exasperata*, *Ficus platyphylla* and *Ficus sur* in the next upper clade (Euclidean distance =89%) are more similar to each other than they are to *Ficus polita*, *Ficus trichopoda* and *Ficus thonningii* in the next upper clade (Euclidean distance =82.5%). *Artocarpus heterophyllus* is completely separate from all the other seven species and is said to be simplicifolious (one-leaved). Its placement indicates that the distribution of the set of measured variables is substantially different from the distribution in the remaining species.

The phytochemical constituents and the chromosomal numbers in this study are considered to be more of the true taxonomic characters of these plants while

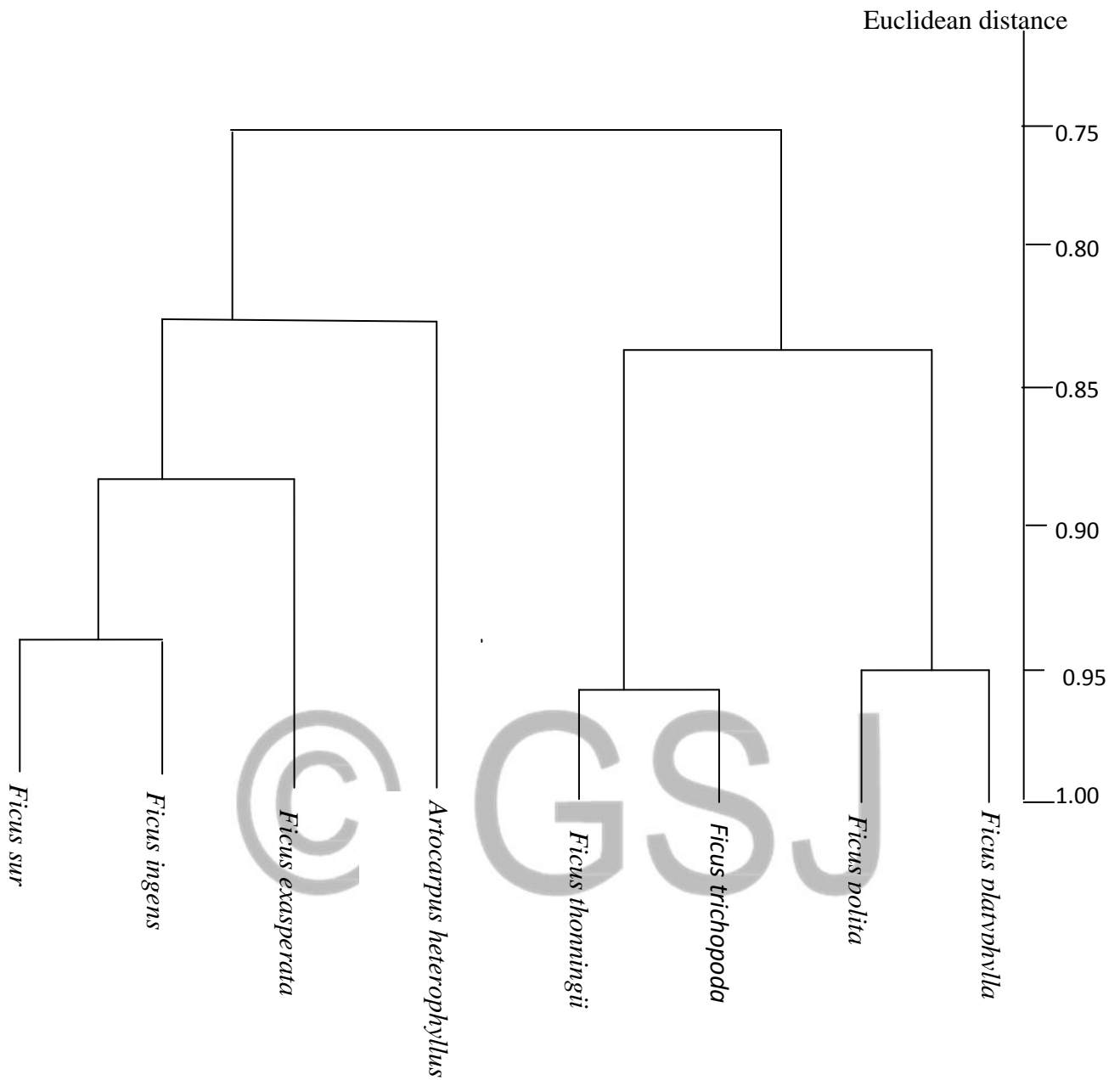
phenological characters are not true taxonomic characters to be used in the classification of these plants. Phenological behaviour of plants could vary from time to time and between taxa depending on some environmental cues and other conditions dependent on the taxa themselves. The cluster analysis involving only the phytochemical constituents (Fig.3) of the plants did not reveal much distinction between the eight species under study. There was no much distinction between the *Ficus* species themselves and between the *Ficus* species and the *Artocarpus heterophyllus* which has been shown to belong to another tribe. This indicates that the distribution of phytochemical constituents may vary irregularly between the *Ficus* species and they on their own can not necessarily be used to discriminate between the species. It is usual to accept phenomenon in taxonomic systematic that as many characters as possible should be used in classification of biological material. Thus, in this study, a combination of phytochemical and chromosomal attributes gave a better reflection of the relationship between the 8 species of Moraceae in line with the established classification scheme for the family. Cluster analysis as such is not an automatic task, but an iterative process of knowledge discovery or interactive multi-objective optimization that involves trial and failure. It is often necessary to modify data preprocessing and model parameters until the result achieves the desired properties



**Figure 1: Taxonomic relationship between the different species of Moraceae on the basis of phytochemical constituents, chromosomal numbers and phenological characters as the set of measured variable.**



**Figure 2: Taxonomic relationship between the different species of Moraceae on the basis of phytochemical constituents and chromosomal numbers as the set of measured variable.**



**Figure 3: Taxonomic relationship between the different species of Moraceae on the basis of only phytochemical constituents as the set of measured variable.**

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