

Morpho-Epidermal Characterization of Plant Species used as Spices in South Eastern Nigeria

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ABSTRACT

Morpho-epidermal characterization of plant species used as spices in Southeastern Nigeria was carried out to determine the unique features of the species. The four plant species are *Ocimum basilicum*, *Ocimum gratissimum*, *Piper umbellatum* and *Piper guineense*. *O. basilicum*, *O. gratissimum* are herbs while *P. umbellatum* and *Piper guineense* are vines. Macro-characters and leaf epidermal studies are essential for taxonomy of plant species but only few publications have been seen on this aspect for the taxa, hence, the studies were carried out. The studies were carried-out using standard methods. The results of the studies showed that simple leaf type is common among the four species and leaf shape was elliptic in all the species studied. The leaf size showed considerable variations among the four species. The leaf length of *O. basilicum* ranged from 0.7 -3.8cm with the mean 3.01, the leaf width ranged from 0.2 - 1.0cm with the mean 1.8. The leaf length of the *O. gratissimum* ranged from 3.7- 6.0cm with the mean of 4.53, the width ranges from 2.8 - 4.0cm with the mean of 2.53. The length of the *P. umbellatum* ranged from 10.1 - 12.1cm with the mean 8.14, the width ranged from 5.9 - 9.0cm with the mean 7.54. The length of the *P. guineense* ranged from 10.2 - 13.1cm with the mean 9.10, the width ranged from 7.1 - 9.2cm with the mean 6.23. The abaxial and adaxial surfaces of the four species showed paracytic and diacytic stomata except in *P. guineense* which was amphiparacytic. The studies have shown that the four species are more related in their quantitative macro-characters and simple leaf type is common among the four species.

Keywords: Morpho-epidermal, Characterization, Spices, Stomata, Nigeria.

INTRODUCTION

Spices are natural plants products used mainly for enhancing flavours of foods [1]; [2]. Spices serve as one of the major ingredients in food preparation and processing throughout the world [3]. The genera *Ocimum* and *Piper* are aromatic annual and perennial herbs and vines in the families of Lamiaceae and Piperaceae respectively, native to the tropical and warm temperate regions with the greatest number of species in Africa [4]. The genus *Ocimum* is characterized by great variability among its constituent species, including morphology, growth habit, the colour of flowers, leaves and stems, and chemical composition.

Piper species are either herbaceous and

vines with some growing as shrubs or small trees. A few species, commonly called 'ant Piper' like *Piper cenocladum* which lives in mutualism with ants [5]. The fruit of the *Piper* plant, called pepper corn when it is round and pea sized, as is usual, is dispersed by birds and other animals. The leaves are usually alternate, simple and entire and stipules are absent, with petioles 6.5 - 30 cm long, with sheath blade semicircular and cordate. The flowers are minute, usually bisexual and without perianth. Few publications have been recorded on these species, hence, the studies were carried out to analyze the features of four species based on macro-characters and leaf epidermal features.

MATERIALS AND METHODS

Fresh specimens of *Ocimum basilicum* L., *Ocimum gratissimum* L., *Piper umbellatum* L. and *Piper guineense* Schumach & Thonn were collected from different locations of Ebonyi State and identified in Ebonyi State University Herbarium.

Macromorphological study: Leaf length and width, were measured following the method of [6]. From each specimen, a total of 20 leaflets were randomly selected for measurement.

Foliar epidermal studies: Epidermal preparation methods also followed [7]. The standard median portions of the leaves obtained by cutting with razor blade were soaked in concentrated trioxonitrate (v) acid for about 25 to 35 minutes depending on the nature of the leaves. The appearance of air bubbles on the surfaces of the leaves indicated their readiness for separation. They were transferred into some water in the Petri dish with a pair of forceps. Both epidermises were carefully separated by teasing them apart and pulling the epidermis back on itself using camel hair brush and dissecting needle. The camel hair brush was also used to remove the adhering tissue debris. The separated surfaces were rinsed in distilled water and then transferred into 50% ethanol for about two to three minutes to harden. They were rinsed again in distilled water and stained with safranin for about five minutes and excess stains were washed off in water. They were mounted in 25% glycerol on slides with the edge of the cover slips sealed with nail varnish to prevent dehydration. The slides were labelled appropriately and examined under the light microscope while photomicrograph of each slide was taken at a magnification (x400), using Canon digital camera fixed to light microscope and connected to personal computer

RESULTS

The macro-characters and leaf epidermal features of the species studied were recorded in the tables 1, 2, 3 and photographs shown in figures 1, 2 and 3 below.

Table1: Qualitative macro-characters of the species studied.

Character/species	O. basilicum	O. gratissimum	P. umbellatum	P. guineense
Leaf arrangement	opposite	opposite	alternate	alternate
Leaf shape	elliptic	elliptic	orbicular	elliptic
Leaf surface	glabrous	pubescent	pubescent	glabrous
Leaf type	simple	simple	simple	simple

Table 2: Quantitative macro-characters of the species studied.

Species Studied	Average Length of Leaves	Average Width of leaves	Average petiole length of Leaves	Average Internode of leaves
O. basilicum	0.7(3.01±0.21)3.8	0.2(1.8± 0.38)1.0	0.2(0.55± 0.10)0.8	3.5(5.105± 0.71)5.0
O. gratissimum	3.7(4.53±0.14)6.0	2.8(2.53±0.87)4.0	0.7(1.01±0.72)2.0	7.1(6.05±0.51)8.2
P. umbellatum	10.1(8.14±0.30)12.1	5.9(7.54±0.21)9.0	5.8(4.01±1.02)8.1	11.8(10.05±0.30)13.2
P. guineense	10.2(9.10±0.31)13.1	7.1(6.23±0.12)9.2	6.7(5.01±0.52)8.8	12.1(11.05±1.01)14.0

NB. All measurements were in centimetre

Table 3: Qualitative leaf epidermal features of the species Studied.

Species	wall pattern		Cell shape		Anticlinal cell		Stomatal type		
	Trichome		Adaxial Surface	Abaxial surface	Adaxial surface	Abaxial surface	Adaxial surface	Abaxial surface	
O. basilicum		polygonal	polygonal	straight	straight	absent	diacytic	present	present
O. gratissimum		polygonal	polygonal	straight	straight	absent	paracytic,	present	present
P. umbellatum		hexagonal	pentagonal	straight	straight	absent	paracytic	present	absent
P. guineense		polygonal	polygonal	slightly curved	slightly curved	paracytic,	paracytic,	present	present

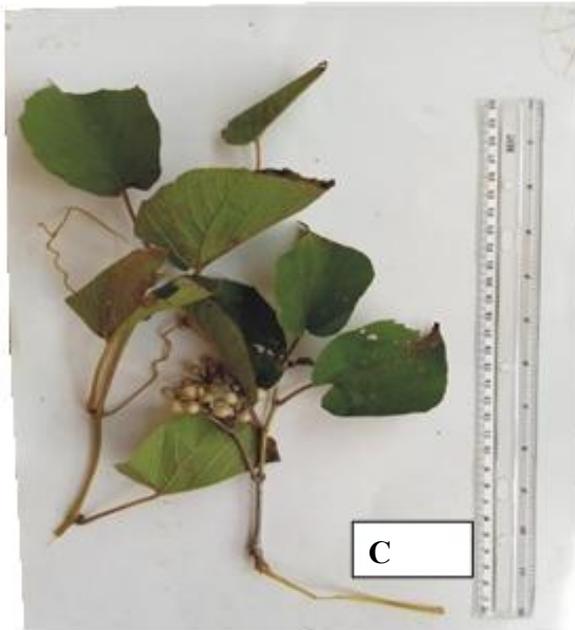


Figure 1: Photographs of species studied.

A: *O. basilicum*

B: *O. gratissimum*

C: *P. umbellatum*:

D: *P. guineense*

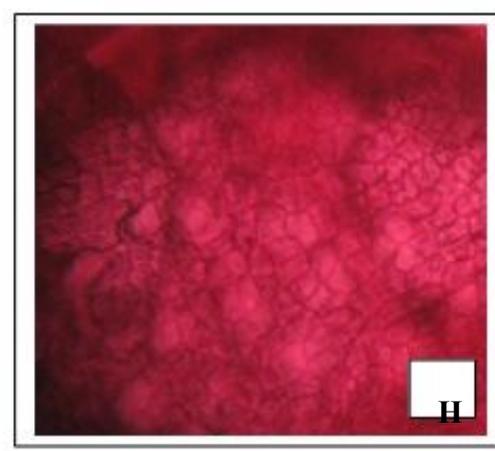
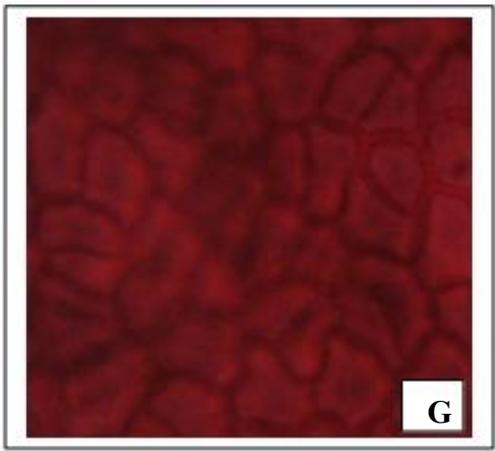
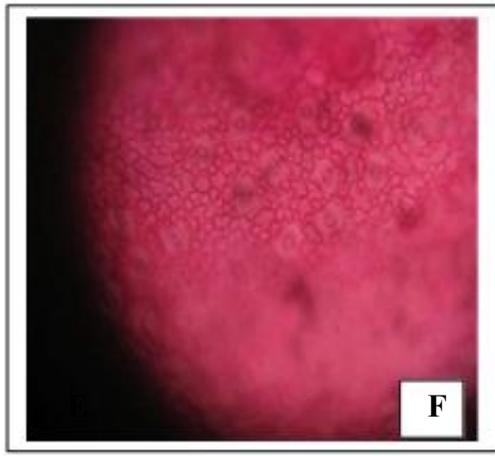
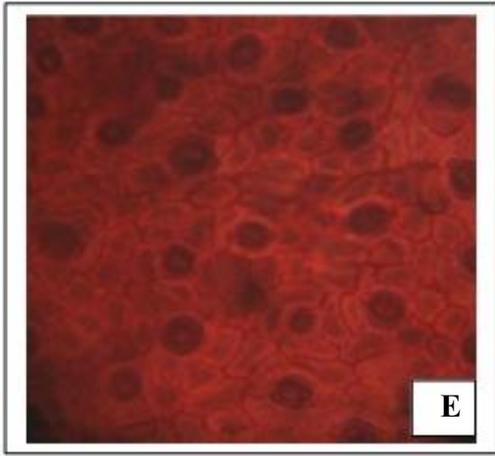


Figure 2: Photomicrographs of the foliar epidermal surfaces of the spices studied E: Abaxial epidermal surface of *O. basilicum*
F: Adaxial epidermal surface of *O. basilicum*
G: Abaxial epidermal surface of *O. gratissimum*
H: Adaxial epidermal surface of *O. gratissimum*

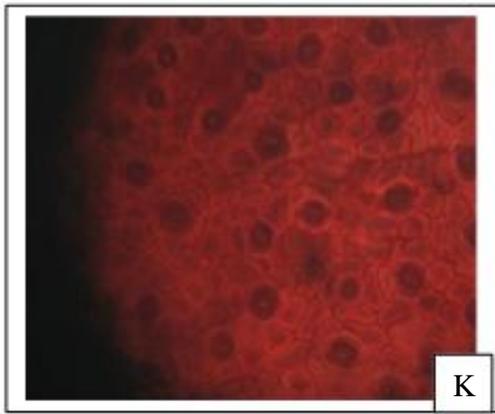
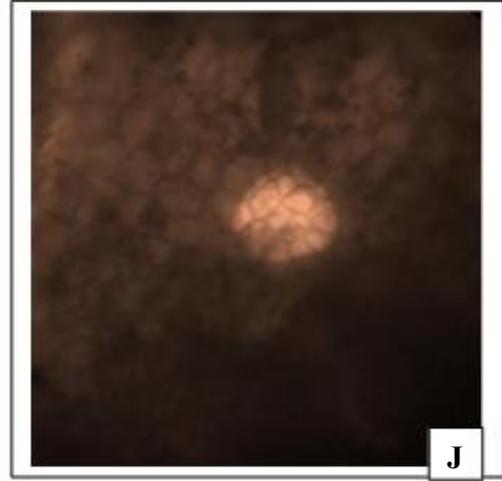
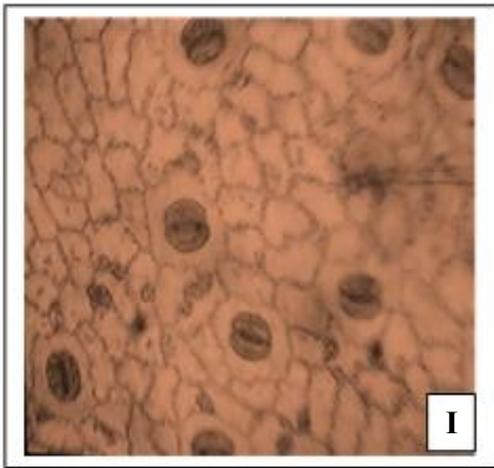


Figure 3: Photomicrographs of the foliar epidermal surfaces of the species studied I: Abaxial epidermal surface of *P. umbellatum*

J: Adaxial epidermal surface of *P. umbellatum*

K: Abaxial epidermal surface of *P. guineense*

L: Adaxial epidermal surface of *P. guineense*

DISCUSSION

The species of *Ocimum* and *Piper* studied have simple leaves in common. *P. umbellatum* has a denticulate leaf margin with fine dentition while the leaves of *O. basilicum*, *O. gratissimum* and *P. guineense* have undulate and entire leaf margins, respectively. The major differences are clear on the quantitative macro-characters as shown in table 2. It was observed that the abaxial of *P. guineense* and *P. umbellatum* have paracytic stomata unlike the adaxial surface of *P. umbellatum* which possesses no stomata but trichome bases. The cell shapes among the species studied were rounded, isodiametric, pentagonal and hexagonal. *O. basilicum*, *O. gratissimum*, *P. umbellatum* and *P. guineense* respectively. The results of the foliar epidermal features of these studies agree with the studies of [8] [9] on the comparative morpho-anatomy of *Piper* species and cultivars in India which reported similar results. The leaf length and width of the of *O. basilicum* ($0.7(3.01 \pm 0.21)$ 3.8; $0.2(1.8 \pm 0.38)$ 1.0) and *P. guineense* ($10.2(9.10 \pm 0.31)$ 13.1; $7.1(6.23 \pm 0.12)$ 9.2) were the smallest and highest respectively. The result of the epidermal cells in table 3 reveals that the adaxial surface of *O. basilicum* and *O. gratissimum* are largely polygonal, with straight anticlinal walls.

Stomata and trichomes are generally present as well as secretory cavities distributed throughout the surface. Large number of stomata was present in most of the species studied. Stomata and associated epidermal cells is an important source of taxonomic characters. The pattern and frequency of stomata on any leaf surface are under tight genetic control, but may be modified by environmental factors like CO₂ [5]. The acclimation of plants in response to environmental conditions may reflect changes on physiological and macromorphological parameters and on the production of secondary metabolites in the spices [8]. Trichomes were found on the epidermis of the species that can serve as good taxonomic tools. Presence of the trichomes may be for defense against the disturbances of the animals that feed on them. So, macro-characters, stomata, trichomes and other features studied are important for classification and identification purposes by many taxonomists. The studies have shown that the four species are more related in their quantitative macro-characters and simple leaf type is common among the four species.

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