

MORPHOHISTOLOGICAL STUDIES OF TWO PLANT SPECIES USED IN ETHNOMEDICINE

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Abstract

Morphohistological studies of two forest species of Annonaceae have been reported; the species covered are *Xylopi aethiopica* (Dunal) A. Rich and *X. quintassii* Engl. And Diels. Wood fibres range in length from $965 \pm 4\mu\text{m}$ in *X. aethiopica* to $853 \pm 3\mu\text{m}$ *X. quintassii*. Leaves are hypostomatic with paracytic stomata found only in the lower epidermis. The epidermal cell walls are straight. Epidermal crystals are cluster crystals, which are present in all the species. Communities bordering the Pre-Suhien Forest Reserve notably, Abrafo-Odumase, Mfuom and Ankaako use these plant species for the treatment of stomach and bronchial troubles as well as rheumatism.

Keywords: Annonaceae, ethnomedicine, Morphohistological, *Xylopi aethiopica*, *Xylopi quintassii*.

Introduction

For centuries, plant-based medicaments have been man's prime therapeutic weapons and are still in the frontline for treating a large number of diseases. Furthermore, the deeper one goes into the study of plants used in traditional medicine, the more one perceives new possibilities for additional plant derivatives. Man's survival has been dependent on his innate curiosity, his desire to examine by trial and error all aspects of his environs and conclude on which ones are harmful and which ones give him the greatest nourishment. Through this quest for survival, many plants of medicinal value have been examined and identified for use by various ethnic groups long before the advent of orthodox medicine. They use different parts of the proven to be of various ailments for survival. The practise by which ailments are cured or treated by the use of raw materials from plants is known as traditional or herbal medicine.

The World Health Organisation (WHO) estimated that up to 80% of the world's population rely on plants for their Primary Health Care. In most rural communities throughout Africa, traditional folk medicine remains the primary source of health care as most modern medical services, are inaccessible.

The different treaties on medicinal plants have mentioned different uses of parts or products of *Xylopi aethiopica* and *X. quintasii*. According to Irvine (1961) and Abbiw (1990), the roots of *X. aethiopica* is used as a cough medicine and a decoction of the bark by the Hausas as an ointment for sores, wounds as well as cuts. The bark, fruit and the seeds, as stated by Irvine (1961) and Abbiw (1990), are used to treat bronchitis, dysentery and biliousness. An oily extraction of the seeds is used as a lotion for boils and eruptions, and as a liniment for lumbago (Irvine, 1961 and Abbiw, 1990). The beaten soft inner bark of *X. quintasii* is rubbed on hands with knotty swellings (Irvine, 1961). Again, it has antimicrobial properties, therefore, it is used in Liberia as a mouthwash. Report by Burkill (1985), indicates that *X. quintasii* is used in herbal medicine to treat various ailments, such as rheumatism, stomach trouble and waist pains.

This research is aimed at providing a better identification procedure; based on diagnostic features, by which *X. aethiopica* and *X. quintasii* could be recognized, both in the field and in the laboratory, as well as, their ethno-medicinal importance to the three communities bordering the fringes of the Pra-Suhien Forest Reserve.

Materials and methods

Study Location

Samples for the studies were collected from the Pra-Suhien II Forest Reserve at Abrafo-Odumase, in the Central Region of Ghana. Questionnaire was used to assess the ethnomedicine importance of the two plant species to the three communities bordering the fringes of the reserve; namely, Abrafo-Odumase, Mfuom and Ankaako.

Morphohistological studies

Morphological and histological studies were carried out on *Xylopi aethiopica* and *X. quintasii* with the view to determine those characters, which could be diagnostic for the delimitation of the species.

Morphological studies

Morphological observations included plant height, girth size, bole shape, bark slash and leaf

Plant Height

Tree heights were assessed using the Optical Reaching Clinometer PM-5 (Borshch-Komponiets, *et al.*, 1986).

Girth Size

Plant girth, was assessed by using a measuring tape. The tape was levelly wound round the trunk and pulled tight.

Moss, lichens and loose bark were removed prior to measurement (Kohl, 1992). Measurement, were taken at a height

of approximately 1.5metres, from breast height.

Bole Shape

Features noted were the shape of the bole: whether cylindrical, fluted or tapering, and the presence or absence of buttresses (Cudjoe, 1970).

Bark Slash

A slash was made obliquely downwards with a sharp cutlass. The cut was made to penetrate all the various layers of the bark until a whitish layer of cambial tissues emerged. Slashed areas were photographed (Hawthorne, 1990).

Leaf Morphology

In addition to the above observations, leafy and fruiting branches of the plant species, was collected for examination in the laboratory. Observations were made on nature of leaf margin, leaf tip, leaf base, leaf surface, leaf arrangement and number of lateral nerve pairs on the lamina surface. Measurements were also taken to determine leaf size and length of the petiole. In all fifty leaves per three individuals of the plant species from each of the three sample locations were used for the laboratory study.

Histological Studies

Histological studies carried out include: bark-fibre dimensions and leaf epidermal studies.

Bark-fibre Dimensions

Bark samples were harvested from four different trees for each of the two species under investigation. Each sample was macerated following the method of Franklin (1946). The macerated materials were thoroughly washed with distilled water. Samples of the washed materials were stained with safranin O and mounted in dextrene polystyrene Xylene (DPX). The cover glass placed on the material of each slide was sealed with cutex. Five slides were prepared from each bark sample, which gave a total of twenty slides for each of the two species. Each slide was examined with a light microscope at a magnification of X40, X100 and X400. Slides, which were found to be satisfactory, were photographed. Fibre length and width were determined for 10 fibres, on each of the 20 slides prepared for each species, using a pre-calibrated ocular micrometer. The mean fibre length and width were calculated for each of the four species.

Leaf epidermal studies

Foliar materials of epidermal studies were collected fresh from plants growing in their natural environment in the forest. Samples of the material for slide preparations were taken from identical regions of each fresh leaf, generally from mid-way between the leaf base and apex of lamina including the mid-rib.

The epidermal peels were obtained using a sharp pointed forceps. In materials where peeling proved difficult, a thin layer of nail varnish was spread over the leaf surface and allowed to dry. Peels were made from both adaxial and abaxial surfaces of each sample. The epidermal strips obtained were cleared of chlorophyll in chlorahydrate (O'Brien and McCully, 1981). The strips were thoroughly washed with distilled water, stained with safranin O, and then mounted in a drop of pure glycerol on a glass slide. A cover glass was placed over the drop and sealed with cutex (O'Brien and McCully, 1981). Forty slides were prepared for each of the two species. The slides were examined with the light microscope and the epidermal features studied. The features observed include: nature of the epidermal cells; nature of trichomes (if present); nature of distribution and dimension of stomata. The stomatal dimensions considered were the length and breadth, and also the stomatal index (SI), which is given as:

$$SI = \frac{S}{S + E} \quad (\text{Metcalf and Chalk, 1979}).$$

Where: **S** = the number of stomata per field of view,
E = the corresponding number of epidermal cells.

Results

Enumeration of the taxa

The diagnostic features by which the two (2) species may be recognized are outlined here:

a) ***XYLOPIA* LINN. (F.T.A. 1: 20)**

Flowers bisexual. Sepals more or less connate, rarely nearly free, valvate in aestivation. Petals 6 in two series, broadened and hollowed at the base, valvate, 3 outer linear, linear-subulate, oblong or rarely ovate, thick, usually scarcely expanding or connivent; 3 inner shorter, similar in form and triquetrous above or ovate-lanceolate, keeled and acute. Stamens indefinite, the innermost sometimes coherent by their filaments into a sheath surrounding the ovaries or inserted upon a sheathing, deciduous, annular extension of the torus, usually narrow-linear, anther-cells frequently multiciliate, connective truncate dilated or more or less subpeltate. Torus nearly plane or slightly convex, plane or concave in the center. Carpels few or numerous, ovaries usually strigose, style elongate, exerted, forming a connivent cone; ovules usually 2 – 10. Fruit-carpels oblong or narrow linear, continuous or torulose.

KEY TO THE SPECIES

Branchlets and leaves densely pilose with spreading hairs;

Flowers shortly pedicellate.....*X. quintasii*

Branchlets and leaves thinly pilose; flowers with pedicels

Up to 1 cm. Long.....*X. aethiopica*

***XYLOPIA QUINTASII* Engl. And Diels**

(01)

Commonly known as “Negro pepper”. *X. quintasii* is also a tree up to 30 metres high and a girth of 1.7 metres. The bole is slender, somehow fluted, short buttress and straight. The bark is dark grey to dark brown. The bark slash is olive-brown, fragrant, and fibrous. Leaves are simple and alternately arranged. Mature leaves are obovate in shape. They range in size from 12 – 18 cm. In length and 5 – 6 cm. Wide. The leaf is obtuse and the apex acuminate. Lateral nerves range between 6 to 9 pairs.

***XYLOPIA AETHIOPICA* (Dunal) A. Rich**

(02)

X. aethiopica is popularly called “African or spice tree seed”. It is a tree up to 18 metres high and with a girth up to 70 cm. The bole is slender, cylindrical, straight and with short horizontal branches clustered at the top. The bark slash is white in colour, very fragrance when fresh, and thick fibrous. Leaves are simple and alternately arranged. Mature leaves are oblong to elliptic in shape. They range in size from 14 – 15 cm. in length and 5 – 6 cm wide. The leaf base is semi-obtuse and the apex acuminate. Lateral nerves range from 7 to 8 pairs. The fruits are in clusters; they are long slender, cylindrical and slightly constricted.

HISTOMORPHOLOGICAL STUDIES

Bark fibre characteristics

The characteristics features of the fibres obtained from the maceration of stem bark are summarized in Table 1. In all species, the fibres are pointed at both ends and all the walls have bordered pits (Fig. 1). The fibres are longest in *X. aethiopica* ($965 \pm 4\text{mm}$) and shortest in *X. quintasii* ($853 \pm 3\text{mm}$).

Table 1
Characteristic features of Fibres

Species	Nature of Fibres	Nature of pits	Dimensions Length (mm)	Width (mm)
<i>X. aethiopica</i>	Pointed	Distinctly bordered	965 ± 4	11.5 ± 0.2
<i>X. quintasii</i>	Pointed	Distinctly bordered	853 ± 3	12.6 ± 0.32

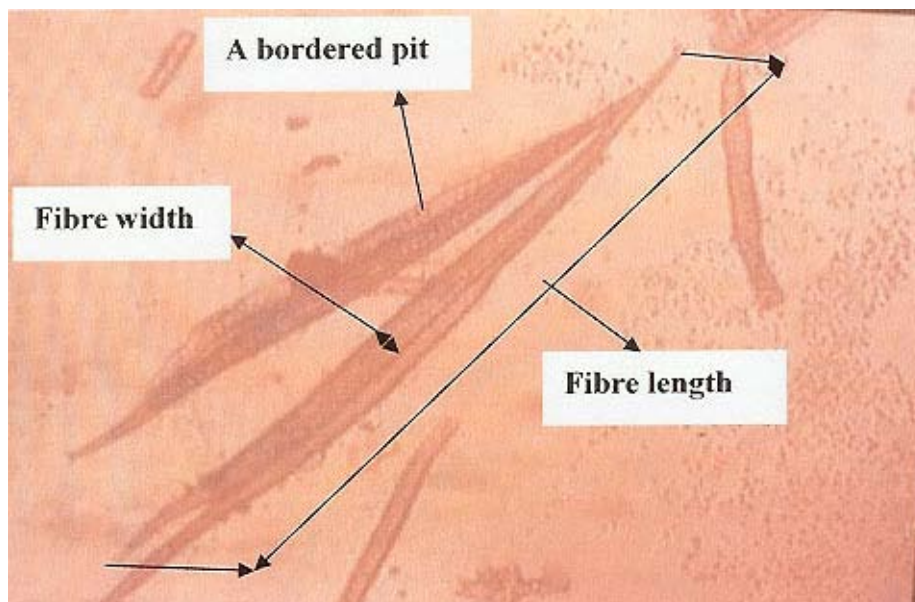


Fig. 1. Nature of fibres (X100), isolated from the root and stem of both *X. aethiopica* and *X. quintassi* obtained from the locations. Scale: 1 : 10.7mm.

Leaf Epidermal Features

The structure of the leaf epidermis of the presently investigated taxa is summarized in Table 2.

Table 2
A Summary of Leaf Epidermis Features

Species	Leaf Surface	Epidermal Cell Wall	Stomatal Distribution	Mature Stomatal Type	Stomatal Measurement		
					L(mm)	B (mm)	SI
<i>X. aethiopica</i>	L	Straight	Hypostomatic	Paracytic	23.1±0.3	11.7±0.1	0.11±0.001
	U	Straight					
<i>X. quintasii</i>	L	Straight	Hypostomatic	Paracytic	20.9±0.2	11.9±0.1	0.2±0.001
	U	Straight					

In *X. aethiopica*, the cells of both the upper and lower epidermis are straight walled. Cluster crystals are found in the lower and upper epidermis (Fig. 2a-b). The leaves are hypostomatic with stomata restricted to the lower epidermis.

Mature stomata measure 23.1±0.3mm X 11.7±0.1mm. Stomatal Index is 0.11±0.001.

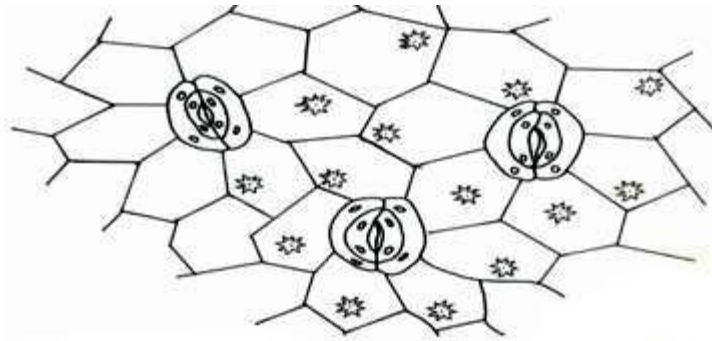


Fig. 2a: Lower leaf epidermis of *X. aethiopica* (x 400).

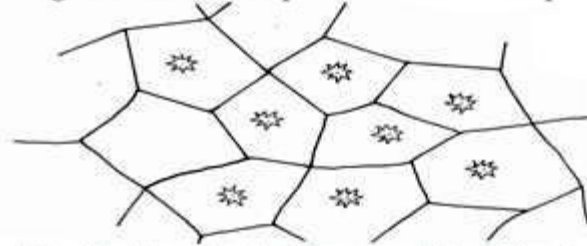


Fig. 2b: Upper leaf epidermis of *X. aethiopica* (x 400).

The cell walls of both the upper and lower epidermis in *X. quintasii* are straight. Cluster crystals are found in the lower and upper epidermis (Fig. 3a-b). The leaves are hypostomatic with stomata restricted to the lower epidermis. Mature stomata are paracytic (Fig. 3a). They measure $20.9 \pm 0.2 \mu\text{m} \times 11.9 \pm 0.1 \mu\text{m}$. The stomatal Index is 0.2 ± 0.001 .



Use of the plant species in herbal medicine and their biodynamic notes

The plant parts used and ailments used for have been summarized in Table 3.

Table 3: A summary of responses of custodians in the three towns

Town	Custodians	<i>X. aethiopica</i>		<i>X. quintasii</i>	
		Part Used	Ailment Used For	Part Used	Ailment Used For
Abrafo-Odumase	Thompson	Seeds; Roots	Rheumatism	Stem bark	Rheumatism; Stomach Troubles; Waist pains.
	Eshun	Stem bark	Bronchial Troubles; Rheumatism	Stem bark	Waist pains
Mfuom	Kodua	Stem bark	Bronchial Troubles; Rheumatism	Stem bark	Rheumatism; Stomach Troubles; Waist pains.
Ankaako	Amosaah	Seeds; Leaves	Bronchial Troubles	Seeds; Roots	Stomach Troubles; Waist pains.
	Yanfraku	Stem bark	Bronchial Troubles; Rheumatism	Stem bark	Rheumatism; Stomach Troubles; Waist pains.
	Sanfo	Stem bark; Roots	Rheumatism	Leaves	Rheumatism; Waist pains.

Discussion

Precise tree identification in the forest is significant to all forestry operations, whether for timber or Non-Timber Forest Products (NTFPs) such as herbal or traditional medicine. Plantations, whether timber or arboretum cannot be raised without seeds, which must be collected from correctly identified trees.

Histomorphological studies

Bark-fibre characteristics

The light microscope shows the bark-fibres as pointed at both ends; also, the walls have distinct pits, which are bordered. Metcalfe and Chalk (1979) have used fibre lengths to group tree species into various classes as short, medium-sized and long, based on some ranges (less than 500 to 900 as short; 900 to 2200 as medium-sized and over 3000 as long respectively). The use of the above criteria has separated *X. aethiopica* and *X. quintasii* into medium-

sized and short, respectively. The above information has been shown in Table 1.

Leaf epidermis features

Leaves, probably anatomically the most varied organs of Angiosperms (Carlquist, 1961), have reported very valuable help in taxonomy. Irvine (1961), Tomlinson (1974) and Metcalfe and Chalk (1979) have reported the use of epidermal characters such as leaf surface, epidermal cell wall pattern, nature of stomata as identifiable aid of some families and genera, and sometimes for species. In the presently investigated plant species, the epidermal cell walls are straight-walled in both epidermis. The leaf stomatal distributions are hypostomatic, i.e., restricted to the lower epidermis and mature stomatal types are paracytic. The epidermis of the taxa revealed the presence of cluster crystals on the upper and lower epidermis.

Stomatal frequency is one of the most widely used characters in pharmacognostic studies (Krishnamurthy and Sundaram, 1970). Many other workers like Ahmad (1979) have established the significance of stomatal frequency as a taxonomic tool in the family Acanthaceae. From the current study it is apparent that stomatal frequency does not vary much among the two species, namely *X. aethiopica* and *X. quintassi*. Therefore, the use of stomatal frequency as a delimiting factor is not possible for the two species.

Traditional medicinal uses

The use of the bark decoction of *X. aethiopica* to treat bronchitis and rheumatism is reported in this current work. Earlier reports by Abbiw (1990) and Bouquet and Debray (1974), reported of the use of the species to treat bronchitis, stomach-aches, dysenteric conditions, febrile pains and rheumatism.

Earlier report by Burkhill (1985), *X. quintassi* was said to be useful in traditional or herbal medicine in treating various ailments such as rheumatism, stomach trouble and waist pains. This work outlines similar uses of the species in treating the above ailments, as outlined in Table 3.

Conclusion

The variations shown by bark and leaf epidermal features may therefore prove useful in plant species identification. The ethnobotanist or forest field worker must not remain complacent with the identification of trees by gross morphology and histological characters. With the ever-increasing application of herbal medicine in the traditional health care system, more taxonomists should be trained. This would promote, the correct identification of plant species for the treatment of the right ailments, to avoid fatalities at the numerous herbal centers springing up.

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