# Anatomy of the leaves and young stem of *Mitragyna inermis* (O. Kuntze)

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The anatomy and morphology of the leaves and young stem of *Mitragyna inermis* (O. Kuntze) is described and illustrated.

THE extract of leaves or that of the stem of *Mitragyna inermis* O. Kuntze (M. Africana Korth) is commonly used in herbal medicine in West Africa for a variety of ailments.

Although the constituents of the plant have been studied (Badger, Cook & Onley, 1950), no complete anatomical investigation appears to have been published, the morphology and anatomy of the plant are therefore described in this communication.

*Mitragyna inermis* is indigenous to the swampy savannah, and in Ghana is found growing both to the North and South of the Volta Region. The materials used in this investigation were leaves and young stems (twigs) collected in April and December, 1962, in the Volta Region from the Sogakope District. The identity of this species was confirmed by the Forestry Department, Kumasi, Ghana.

# Macroscopy

The leaves have an opposite and decussate phyllotaxis and measure  $2 \cdot 5$ -12 cm long, and  $1 \cdot 5$ -8 cm wide. They are ovate, simple, petiolate and stipulate. The colour varies from reddish brown in the young to brownish green in the older leaves. Generally, the leaf apex is acuminate and rarely acute, with base rounded and symmetrical. The margin is entire. The upper surface is glabrous, while, on the lower surface trichomes are found mainly on the midrib and lateral veins. The midrib is prominent with 6-8 lateral veins leaving it at an angle of 40-50° and anastomosing near the margin. Venation is reticulately pinnate. Texture is thin and papery with odour slight and taste somewhat bitter.

The *petiole* is grooved and measures 6 mm-3 cm long, and 0.75-1.5 mm wide. The lower surface is rough due to the presence of short warty trichomes.

The stipules are in pairs. They measure  $5 \text{ mm}-2.5 \text{ cm} \log \text{and } 2-6 \text{ mm}$  wide. They are oblong-lanceolate and reddish brown in colour (Fig. 1B). and are deciduous, having a thin and papery texture. They are odourless and the taste is slightly bitter. The lower or outer surface is pubescent while the upper and inner surface is glabrous except near the lower half of the stipule where dark brown elongated secretory glands are found (Fig. 1.C e.s.g.). These are arranged alternately in rows of 3-5. Each elongated secretory gland has a broad and flattened base with a tapering apex (Fig. 2.G). Between these glandular structures are long hair-like trichomes (Fig. 1.C. tr.) In the young stipule the secretion of

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the secretory glands is copious, milky white and sticky and coats the adjacent young leaflets.



FIG. 1. Leaf  $\times$  1. B. Stipule (outer)  $\times 1\frac{1}{2}$ . C. Stipule (inner)  $\times 1\frac{1}{2}$ . D. Young leafy stem with fruit  $\times \frac{1}{4}$ . E. T/S midrib of leaf. F.T/S stipule. G.T/S petiole all  $\times 20$ . ca.ox., calcium oxalate; ep., epidermis; e.s.g., elongated secretory gland; f., pericyclic fibre; g., groove; ph., phloem; tr., trichome; v., vein; xy., xylem.

The young stem (twig) is rounded to semi-cylindrical with the outer surface reddish brown in colour. Slight longitudinal striations are present. A smooth transverse surface shows a narrow bark up to 15 mm wide with a buff coloured radiate xylem. The greyish central pith is up to 2 mm in diameter. Branch scars are in pairs.



FIG. 2. A. Leaf, lower epidermis. B. Leaf, upper epidermis. C. Stipule, outer epidermis. D. Stipule inner epidermis. E.T/S leaf. F.T./S stipule through vein all  $\times$  160. G.T./S elongated secretory gland. H. Long hair-like trichome and warty trichome,  $\times$  160. c.ox., calcium oxalate; c.c., central cells; e., epidermis; pal., palisade; o.c., outer cells; sto., stomata; up.e., upper epidermis; v.b., vascular bundle.

In the description that follows, the symbols R, T, and L signify measurements taken in radial, tangential, and longitudinal planes respectively. Although a wide variety of material was used, these values cannot be regarded as absolute.

## Microscopy

## LAMINA (Figs 1-3)

The transverse section through the *midrib* shows that the palisade cells do not extend over the meristele. The xylem is lignified and forms a half cylinder surrounded externally by phloem tissue. (Fig. 1.E., ph; Fig. 3.A., ex.ph.), and thick walled pericyclic fibres (Fig. 1.E., f.). The central core is made up internal phloem, parenchyma and moderately thin walled Phloem is made up of sieve cells which are restricted to groups. fibres. Generally, the cells of the parenchyma have thin pitted walls, but, occasionally some have reticulate thickening (Fig. 3.A., r.p.). The xylem is made up of vessels, tracheids, and parenchyma all of which are strongly lignified. Vessels may have alternately arranged bordered pits, spiral or annular thickening. Isolated vessel elements measure R and T, 12-40  $\mu$ , L, 100  $\mu$ -1.5 mm. The parenchyma around the xylem vessels is composed of rectangular to longitudinally elongated cells with pitted walls. These measure R and T, 9–20  $\mu$ ; L, 9–75  $\mu$ .

Within the epidermis is a collenchymatous zone 5–15 rows in radial depth. These cells have pitted walls up to 7  $\mu$  thick, and measure R and T, 15–35  $\mu$ ; L, 35–230  $\mu$ . Large intercellular spaces are often present between these cells. The collenchyma and parenchyma may contain large cluster crystals of calcium oxalate or reddish brown material not easily removed with chloral hydrate solution. Pericyclic fibres are thick walled and non-lignified, and measure from 250  $\mu$ -4.5 mm long and 7–24  $\mu$  wide. Ocassionally, some may be branched.

The upper epidermis of the lamina (Fig. 2B) consists of a single layer of polygonal tabular cells covered with a thin cuticle. The anticlinal walls are usually straight, and stomata and trichomes are absent. The mesophyll is clearly differentiated into a broad palisade layer made up of 3-5 rows of thin-walled cells, which occupies up to three quarters of the width of the mesophyll (Fig. 2E). The upper palisade cells are much more elongated than the lowermost ones. The spongy mesophyll is from 1-3 cells wide; large calcium oxalate crystals in idioblast cells are present; these are conspicuous and mainly found near the vascular bundles and sometimes between palisade cells; they are similar in size and shape to those of the midrib.

On the *lower epidermis* (Fig. 2A), stomata are found in abundance, and these are of the paracytic or rubiaceous type. The trichomes are found mainly along the veins and are of two distinct types. There are the thick walled hair-like curved trichomes each with a broad basal foot (Fig. 2.H). These may be unicellular, but are often divided into compartments by very thin septa; trichomes of 2–14 compartments have been found; they measure from 100–1020  $\mu$  long and 12–50  $\mu$  wide at the base. They are found in greater abundance than the short unicellular thick-walled conical, warty appressed trichomes (Fig. 2.H). These ranged from 10–165  $\mu$  long and 10–25  $\mu$  wide at their base. Both kinds of trichomes contained reddish brown material. The palisade ratio is 7.5-10-14; stomatal index, 11.75-15.1-17.6, and vein islet number 16-20-26.



FIG. 3. A.T/S through midrib. B.L/S through midrib both  $\times$  160. cal.ox., calcium oxalate; col., collenchyma; f., pericyclic fibre; in.ph., internal phloem; i.e. lower epidermis; l.p., lignified parenchyma; ex.ph., external phloem; ph.f., phloem fibre; r.p., reticulate parenchyma; s.c., secretory cell., xy., xylem.

The *petiole* in transverse section resembles the midrib except that there are fewer pericyclic fibres around the vascular bundle. In addition to

the main bundle, there are two accessory bundles surrounded by sheath parenchyma (Fig. 1.G v.b.). The epidermal trichomes are of the thick walled warty type as described for the lamina.

The epidermal cells of both leaf and petiole contain a dense reddish brown substance not easily removed by chloral hydrate solution.

#### STIPULE

The inner or upper epidermis (Fig. 2.D) has no stomata, and is covered by a thin cuticle. Long hair-like trichomes are found in abundance mainly towards the base of the stipule and between the elongated secretory glands (Fig. 1C, tr.). The outer or lower epidermal cells are polygonal in surface view, except over the veins where they are elongated in the direction of the main veins; both stomata (paracytic type) and trichomes are present (Fig. 2C, sto.; tr.).

Below the epidermis is fairly thick-walled pitted parenchyma, composed of rectangular to longitudinally elongated cells (Fig. 2F). Both epidermal and parenchymatous cells are filled with a dense reddish brown substance. Large calcium oxalate crystals are found in the cells along the veins. The vascular bundle, like that of the midrib of the leaf, consists of vessels, tracheids, fibres, and phloem cells. Both fibres and vessels are lignified. In macerated material examined, vessel-elements up to  $825 \mu$  long and  $4-10 \mu$  wide have been measured. Isolated fibres measured up to  $1200 \mu$ long having blunt to contorted ends.

The elongated secretory glands (Fig. 2G) have a broad base and tapering apex. They measure 500–1150  $\mu$  in length and 130–400  $\mu$  in width at their basal end. They consist of a multicellular core of thin walled isodiametric parenchyma cells (Fig. 2G, c.c.) covered with a palisade-like layer of cells. The parenchyma contains in certain cells small solitary cluster crystals of calcium oxalate which measure from 6–18 $\mu$  in diameter. Starch is absent from leaf, petiole and stipule.

## YOUNG STEM (TWIG) (Fig. 4)

The outer protective layer of the stem is made up of cork, the number of rows depending on the age of the stem. From specimens examined, the range was from 4–18 rows of radially arranged cells. The sections examined had cork cells of two types, that is cork cells tangentially and radially elongated cells (Fig. 4. ck and c'k).

All cork cells are thin walled suberised and measured R and T, 25–75  $\mu$ ; L, 35–165  $\mu$  long. The *cortex* is made up of collenchyma and parenchyma. Most cortical cells contained reddish brown secretion while some have large cluster crystals of calcium oxalate. The *pericycle* is made up of thick unlignified fibres each with a small lumen. Isolated pericycle fibres measured R and T, 7·30  $\mu$ ; L, 300  $\mu$ -8·1 mm long. The *secondary phloem* is made up of parenchyma, fibres, sieve cells, and is traversed by uniseriate medullary rays. Medullary ray cells have contents which are similar to those of the secretion cells of the cortex. No starch was found to be present. The cambial zone is well marked and consists of 2–3 rows of thin walled cells.

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The vessels of the secondary xylem have mostly bordered pits while some reticulately thickened vessels and a few spiral vessels occur in the



FIG. 4. A. T/S young stem (twig)  $\times$  17. B.T/S young stem bark. C. T/S section of xylem, both  $\times$  160. c., cambium; ca.ox., calcium oxalate; col., collenchyma; ck & c'k., cork cells; f., pericyclic fibres; ph., phloem; ph.f., phloem fibres; m.r., medullary rays; s.c., secretory cells; xy., xylem; xy.p., xylem parenchyma; v., vessel.

primary xylem. Bordered pitted vessels have tapering projections sometimes up to 325  $\mu$  long. Isolated vessel-elements measure R and T, 15– 75  $\mu$ ; L, 115  $\mu$ -1 05 mm long.

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The *medullary rays* are heterogeneous, consisting of a central core of horizontally elongated cells, bordered on either side by erect vertically elongated cells. Cells of both types are lignified and pitted. Xylem fibres are thick walled with tapering to contorted ends. Isolated fibres measure R and T, 9–30  $\mu$ ; L, 280  $\mu$ -3·06 mm. Both xylem parenchyma and medullary ray cells contain brown contents. Calcium oxalate crystals are absent.

The *pith* is made up of thin pitted walled parenchyma cells many of which contain reddish brown contents.

#### POWERED LEAF AND STEM

The features which are common to both leaf and stem powders are mainly the warty trichomes, unlignified pericyclic fibres, lignified fibres, bordered pitted vessels, cells containing reddish brown secretion or calcium oxalate. The main distinguishing features between leaf and stem powders are the long hair-like trichomes and fragements of the elongated secretory glands both of which are only found in leaf.

## Discussion

The structure of the leaf, stipule and stem of M. inermis is typically that of the family Rubiaceae. The macroscopical and microscopical characters of this species can, however, readily be distinguished from that of M. stipulosa, and M. ciliata (Shellard & Shadan, 1963). The principal distinguishing feature is the presence of both pericyclic, and phloem fibres in M. inermis. Fibres are, however, reported absent from leaf and petiole of M. stipulosa and M. ciliata.

In *M. inermis* there are two distinct types of trichomes, the long hair-like type and the conical warty appressed trichomes. Finally, the calcium oxalate cluster crystals of *M. inermis* are much larger than those of *M. stipulosa* and *M. ciliata*.

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## References

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