The morphology and anatomy of *Synsepalum dulcificum* has been described, principally because of enquiries from pharmaceutical industries considering this species as a potential replacement of the sweetening agent cyclamate. With the aid of a stereoscan and the light microscope, the pollen grains have been studied for the first time. The grains have either three, four or five colpi. The exine has two well-defined layers. The scanning electron microscope has revealed a very fine striate-granular pattern on the sexine. The importance of leaf venation in the classification of the Sapotaceae has been emphasized in a detailed description of *Synsepalum dulcificum*. The need for further studies has been noted.

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**INTRODUCTION**

The recent alarm raised because of the deleterious effects of cyclamates used as a sweetening agent in a number of soft drinks and baby foods has caused food technologists to look elsewhere for sweetening substitutes. Of special interest has been *Synsepalum dulcificum*, the pulp of whose fruits Ghanaians and other West Africans use to obscure the sour taste of various food substances. Children often express great delight in being able to impress their
unschooled friends and elders because they are able to consume sour fruits such as lemon, lime and grapefruit without expressing any distaste. I can remember very vividly my own participation in this kind of vaunting while in elementary school.

Daniell (1852) was the first to describe scientifically the unusual quality of this fruit, which he called the "miraculous berry" and whose chemical constituents many food chemists have since studied. Recently, Kurihara & Beidler (1968) studied the amino acid composition of its proteins and showed that, of the total residue, the amino acids are distributed in the following percentages: lysine, 7.9; histidine, 1.8; (ammonia), 17.4; arginine, 4.7; aspartic acid, 11.3; threonine, 6.1; serine, 6.1; glutamic acid, 9.2; proline, 6.0; glycine, 9.8; alanine, 6.3; half cystine, 2.3; valine, 8.0; methionine, 1.0; isoleucine, 4.7; tryosine, 3.6 and phenylalanine, 5.0. They suggested further that the active principal of *S. dulcisicum* is a basic glycoprotein with a probable molecular weight of 44,000. Unfortunately, their study also showed that the glycoprotein is rather labile and hence difficult to stabilize. If the purified protein could be stabilized, it would enable this natural product to be used as a sweetening agent.

The present study was prompted by the many enquiries I have received from governmental agencies and pharmaceutical companies that are working on the biology and chemistry of this species. It is intended as a contribution to the understanding of the morphology of this potentially important economic plant.

**MATERIALS AND METHODS**

Materials for this study were kindly supplied by Mr A. A. Enti of the Ghana Herbarium, Department of Botany, University of Ghana, Legon. Standard microtechnical procedures were followed in the preparation of the specimens. The woods, twigs and roots, preserved in formalin/acetic acid/alcohol (FAA), were washed in running water. Transverse, radial and tangential sections were cut on a sliding microtome, stained in Heidenhain's haematoxylin and counterstained with safranin. This was followed by dehydration and mounting in Canada balsam on slides. Epidermal preparations and transverse sections of leaves were prepared in the usual way. The leaf preparations were stained in safranin, dehydrated in an alcohol series and mounted permanently in Canada balsam on slides.

The diagnostic characters for the description of wood were selected from Tippo's (1941) list. In general the anatomical terms used follow those approved by the Committee on Nomenclature of the International Association of Wood Anatomists (1957).

For light microscopic studies, anthers of flower buds just opening were acetolysed and the pollen mounted in glycerine jelly. The photomicrographs were taken with an M20 Wild microscope.

For study under the scanning electron microscope, the pollen grains were placed on a coverslip that had been glued to a metal stub. The grains were coated with approximately 150 Å of gold evaporated from a tungsten filament in a vacuum coating unit. A stereoscan, manufactured by the Cambridge
Scientific Instrument Company, was used in the examination and photography. Terminology for the descriptions of pollen follows that of Erdtman (1966).

**GENERAL MORPHOLOGY**

The Sapotaceae is a large family comprising about 40 genera with some 800 species of evergreen trees, shrubs and rarely climbers, that grow mostly in the tropics and the subtropics. The leaves are simple, entire, generally alternate, with or without stipules. The flowers are bisexual, actinomorphic, small, solitary, cymose, and often in axillary clusters. The calyx is usually 4-8-lobed, but sometimes the sepals are free. The corolla is 4-8-lobed, the lobes 1-2-seriate, imbricate, and sometimes with external petaloid appendages. The stamens are 8-15, inserted on the corolla in 2 or 3 series. The fertile ones are generally equal in height to the corolla lobes but the staminodes, often comprising 1 or 2 of the outer series, are reduced. The anthers are 2-locular and dehisce longitudinally. The ovary is superior, of 4 or 5 united carpels. The placentation is axile with 1 ovule in each locule. The ovule is anatropous. The fruit is a hard berry with a thin testa of bony or leathery texture and with a large rather broad hilum. The embryo is large; the endosperm is scanty, sometimes fleshy.

This family is generally recognized because of its latex-producing properties. A number of genera within it are of considerable economic importance. For example, chicle, a latex substance obtained from the inner bark of the stem of *Manilkara zapota* (L.) P. van Royen (*Achras zapota* L.), is used together with sugar, adulterants and flavours in the manufacture of chewing gum. In addition to the latex, the pear-like fruit (“Sapodilla” or “Naseberry”) of this plant is eaten in many tropical areas. The star-apple, *Chrysophyllum cainito* L. is the star-shaped apple-like fruit with an acidulous pulp that is also eaten in the tropics. “Guttapercha,” a derivative of *Palauquium gutta* Burck, another member of this family, is famous for its use in the production of submarine cables, golf balls, telephone receivers and numerous adhesives.

**TAXONOMY**

*Synsepalum dulcificum* was first described as *Bumelia dulcificum* by Schumacher (1827). Later, de Candolle (1844) transferred it to the genus *Sideroxylon*, placing it in a separate section which he named *Synsepalum* because of the fused sepals, but noting that the species perhaps belonged to a distinct genus within the Sapotaceae. In 1891, Baillon (1891) proposed such a new genus and gave to it de Candolle’s sectional name *Synsepalum*.

Heine (1963) recognized three species in the *Flora of West Tropical Africa*, namely *S. glycydorum*, *S. dulcificum* and *S. stipulatum*, and regarded differences in the leaves as systematically significant. He described those of *S. glycydorum* as having elliptic-oblanceolate leaves that are abaxially densely silky pubescent. The leaves are up to 15 cm long and about 5 cm broad with 10-12 pairs of lateral veins. The leaves of *S. dulcificum* and *S. stipulatum*, on the other hand, are glabrescent beneath. The major difference between these two species is in the venation of the leaves—in the former there are 8-12 pairs of veins and the latter about 18 pairs.

The importance of venation in the classification of the Sapotaceae was
recognized by Bachni (1938), Lam (1939) and other taxonomists before them. Briefly four types of venation patterns have been recognized within the family: (1) Transverse tertiary veins. These are more or less perpendicular to the secondary veins and are exemplified in species of Palaquium and Madhuca. (2) Longitudinal tertiary veins. These are generally parallel to the secondary veins. Examples are easily found in species of Payena, Planchonella, Chrysophyllum and Pouteria. (3) Tertiary veins which are slightly larger than the minute but conspicuous areolate reticulations. These occur in species of Synsepalum and Nesoluma. (4) All veins parallel and crowded. Species of Micropholis and Manilkara exhibit this pattern.

It should be pointed out that various transitional forms exist among the four types. Further comparative studies are needed to show the extent of differences in venation between the Old and New World species.

LEAF VENATION

The following is a description of the leaf architecture of Synsepalum dulcificum (Fig. 1B; Plate 1A,B). The terminology used is based on the key devised by Dr Leo Hickey, Smithsonian Institution, in his forthcoming publication, "The paleobotany and stratigraphy of the Golden Valley Formation."

Leaves entire, symmetrical, obovate, length from 4.3 to 7.5 cm; width 3.1 to 3.8 cm; length/width ratio 2 : 1. Apex obtuse but occasionally slightly acuminate; base cuneate. Texture subcoriaceous, adaxial surface glabrous, abaxial surface pubescent; glands absent. Petiole very short, 4-5 mm long, flattened on adaxial side but rounded on abaxial side.

Venation pinnate, eucamptodromous, basal secondaries brochidodromous. Midvein stout, much thicker than the secondaries; course straight. Secondary veins 12-13 on either side of midvein; angle of divergence moderate, approximately 45°-55°; variation in angle of divergence nearly uniform. Relative thickness of secondaries moderate; course uniformly curved. Inter-secondary veins simple. Tertiary veins weakly percurrent.

Higher order venation: both quaternary and quinary veins relatively randomly oriented; the highest vein order is seven; the marginal ultimate venation looped.

Veinlets none. Areolation imperfectly polygonal.

TRICHOMES

Among the prominent features of the Sapotaceae are the characteristic unicellular two-armed trichomes observed on many parts of the plant. The abaxial leaf epidermis of Synsepalum dulcificum is heavily covered with very thick-walled two-armed hairs. It is interesting to note that some of the long hairs appear chambered in such a way that they resemble bicellular or multicellular hairs. This condition has not been reported in the anatomical literature on the family (cf. Solereder, 1908; Metcalfe & Chalk, 1950). The calyx is covered with long hairs (Fig. 1D).
MORPHOLOGY AND ANATOMY OF *SYNSEPALUM DULCIFICUM*

**Figure 1.** *Synsepalum dulcificum*: A, stamen (x10); B, habit (x1); C, seed (x3); D, flower (x5); E, cross-section of fruit (x3); F, dissected flower (x5); G, fruit (x5).

**FLORAL MORPHOLOGY**

*Calyx lobes* (sepals). These are 4 or 5 in number, mostly 5 in the specimen under examination, length 3-4 mm. The margin of the calyx is ribbed. The sepals are thick, especially at the lower end of the calyx.

*Corolla.* The 4 or 5 petals form a short tube by their basal coalescence. The corolla is about the same height as the calyx. There are five petaloid appendages forming an inner circle and coalescing at the base with the corolla (Fig. 1F). The petaloid appendages have the same whitish colour and the same height as the corolla. They differ from the petals in being much narrower and in tapering to the tip.
Androecium. The 5 stamens in the specimen under examination are epipetalous. Each stamen is situated almost opposite a petal. The anthers are attached to delicate filaments and dehisce extrorsely by means of longitudinal slits (Fig. 1A).

Gynoecium. The style is simple, erect with a very inconspicuous stigma (Fig. 1D). The ovary is superior. The outer wall of the ovary is heavily covered with hairs.

Fruit. The fruit is a one-seeded berry (Fig. 1E,G), which is small, purplish-black when ripe, and edible.

Seed. These have a hard, shiny and oily testa (Fig. 1C,E). The embryo is rather large and fills the cavity, while the radicle is slightly visible. The cotyledons are thick and fleshy.

Pollen morphology

Pollen grains 4-colporate, occasionally 3- or 5-colporate; prolate (polar axis, 26.5-27.5 μm; equatorial axis, 14.5-17.5 μm). Exine of two well-defined layers. Sexine pattern (as seen with the scanning electron microscope) very finely striat-granular with distinct pitting. Colpi margins slightly thickened. Ora narrow, oblong, latitudinally elongate (Plates 1C,D and 2A-E).

Vegetative Anatomy

Leaf

Lamina: dorsiventral. Leaf cuticle: generally thin but with finely granulated marks on abaxial surface; primary vein thickened and prominent above and below. Hairs: mainly of long, thick-walled, unicellular, 2-armed trichomes. Epidermis: consisting of variously shaped cells with sinuous walls; cell walls pitted in places. Cells on adaxial and abaxial surfaces of main veins conspicuously rectangular with non-sinuous walls; in transverse view, epidermis on both surfaces rather shallow with thin walls; outer walls slightly thicker than the remaining walls. Stomata: almost always confined to abaxial surface; guard cells without any specially modified cells (anomocytic stomata). Mesophyll: composed of one layer of poorly differentiated palisade cells; palisade layer clearly visible near midvein; spongy tissue with abundant intercellular spaces. Sclerenchymatous fibres present in mesophyll. Vascular bundles: including that of midvein embedded in mesophyll. Laticiferous sacs present in mesophyll and arranged in a horizontal row in middle of spongy tissue. Crystals: present, solitary and cuboidal forms, heaviest concentration around vascular bundle of midvein; crystal-sand also present in mesophyll.

Petiole

Outline: slightly crescentiform with two projections on adaxial surface; mid-portion of adaxial surface prominent. Epidermis: 1-layered, cells variable in shape, mostly rounded on abaxial surface and cuboidal on adaxial surface; cells thin-walled. Cuticle: uniformly thickened; slightly thicker on adaxial surface. Hairs: present. Stomata: very few, not so conspicuous as on lamina.
Cortex: cells mostly 10-14-layered, separated from epidermis by a distinct 1-layered hypodermis; middle cortical cells larger and somewhat thick-walled. Vascular bundle: abaxial strand arc-shaped, adaxial strand flattened; xylem and phloem strands transversed by 1-2 layers of parenchyma; fibres comprising of 3-5 cells surrounding the vascular strands; central ground tissue composed of fairly uniform parenchymatous cells with few distinctly large cells. Crystals: present, similar to those of lamina.

Stem

Juvenile stem (diameter 5-7 mm)

Cork: 6-12-layered; cells of innermost layer filled with tanniniferous material. Cortex: many-layered, about one-fifth of stem diameter; isolated strands of fibres and laticiferous sacs (1) present (Plate 3A). Phloem: composed of sieve-tubes and companion cells; tangential bands of 2-3 layers of fibres present; phloem traversed by uniseriate rays. Xylem: composed of continuous cylinder with narrow uniseriate rays, small radial rows of vessels, apotracheal parenchyma and thick-walled fibres. Pith: composed of thick-walled cells, conspicuously pitted; many cells filled with tanniniferous material and starch grains. Crystals: few present in cortex and pith.

Mature wood

Macroscopic features. Vessels small, visible only with a lens. Lumina mostly open. Ray cells smaller than vessels. Tangential lines of apotracheal parenchyma visible with a lens.

Microscopic features. Growth rings distinct (Plate 3B). Vessels solitary and in radial multiples grouped in oblique patterns, medium-sized (range 70-100μm, average 80μm). Perforation plates slightly oblique to oblique, simple (Plate 3C,D). Intervascular pitting alternate, rounded or slightly elongated; pit apertures narrow and confined within border. Tyloses commonly present in mature wood, commonly thick-walled with sclerotic tendencies. Parenchyma apotracheal, in tangential narrow bands but often with some scattered cells, bands usually 1 cell wide, occasionally 2 cells; some cells filled with tannin-like material. Fibres thick-walled with few simple pits on radial walls and small lumina, gelatinous. Rays heterogeneous, exclusively uniseriate (Plate 3D). Individual rays 4-32 cells high. Ray cells in tangential longitudinal sections procumbent, except the end cells, which are vertical; many cells filled with silica; cell walls conspicuously pitted.

Root

Cork consisting of five layers in juvenile plants examined; arising superficially from innermost layer of primary cortex; outer layers lignified. Cortex: narrow, 4-6 layers, cells with distinct nuclei and other cell inclusions. Laticiferous sacs: few observed in cortex. Phloem: in continuous bands, not traversed by uniseriate rays as in twig. Xylem: as in twig; vessels with simple perforation plates, narrow compared with those in wood, 40 μm in diameter. Most vessels near the outer portion of xylem surrounded by fibres. Parenchyma: apotracheal, as in twig. Wood fibres: forming broad bands of
abundant (small) cells with thick walls and rather narrow lumina. Rays: uniformly uniseriate; cells often filled with tannin-like substances. Some parenchymatous cells also filled with tannin-like substances.

DISCUSSION

Our knowledge of the morphology and anatomy of economically important plants, especially those of the tropics, is alarmingly meagre and often non-existent. One would have expected that, since 1852 when Daniell demonstrated the scientific significance of *Synsepalum dulcificum*, attempts would have been made to acquire comprehensive information on all aspects of this plant. Holle’s (1892) thesis on the anatomy of the Sapotaceae may have included the genus *Synsepalum*, but this is not available to me. Engler’s (1904) treatment of the African Sapotaceae included an illustration of a transverse section of part of the leaf of *Synsepalum dulcificum* and a comment that sclerenchyma fibres with thick walls are common in the mesophyll.

The pollen grains of *S. dulcificum* are similar to those in other members of the Sapotaceae according to Erdtman (1966). Under the light microscope the walls of the grains appear to be very finely granulate; however, the numerous pits can clearly be discerned. The systematic importance of the sculpturing of *S. dulcificum* pollen will be known only when a comparative study of all species within the genus is made.

The anatomical characters of the vegetative organs include all the major distinguishing features of the family. The unicellular two-armed hairs which occur on the leaves, and the laticiferous sacs found in all the organs, are present in *Synsepalum dulcificum*.

The confusion which exists in the systematics of the family Sapotaceae becomes evident when one reviews the literature (cf. Baehni, 1938; Lam, 1939). Recently Hutchinson (1969) summed up the situation in commenting, “There are between thirty and forty genera [within the family], and many of them are very imperfectly known. Botanists travelling in tropical forest regions, therefore, should make good collections of this family. Fruits and seeds are particularly important, accompanied, of course, by leaves and, whenever possible, flowers. Owing to recent indiscriminate creation of a large number of so-called genera, the family in that respect is in considerable confusion.” In Record’s study of the American woods of the Sapotaceae (Record, 1939), he stated his conviction that the systematic anatomy of the woods would greatly assist in the solution of taxonomic problems.

Unfortunately, the xylem anatomy of most genera in the family has never been studied. The treatment in this paper represents the first inclusive (though not very exhaustive) work on a species of the genus *Synsepalum*.

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E. S. AYENSAU
MORPHOLOGY AND ANATOMY OF SYNSEPALUM DULCIFICUM

REFERENCES


EXPLANATION OF PLATES

Synsepalum dulcificum

PLATE 1

A. Whole leaf (x2).

B. Enlarged part of leaf showing variation details (x5).

C,D. Stereoscan photographs of pollen showing four colpi and wall pitting respectively (C x2000, D x2200).

PLATE 2

A. Reticulation on pollen wall (x11,500).

B-E. Light microscope photographs of pollen in various orientations (x1000).

PLATE 3

A. T.S. of stem showing laticiferous sacs (1) in cortex (x80).

B. T.S. of stem showing growth rings.

C. Radial section of stem showing end plates of vessels with simple perforations and crystals in ray cells.

D. Tangential section of stem showing uniseriate rays.