

PHARMACOGNOSTIC ANALYSIS OF *BOUGAINVILLEA* *GLABRA*

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ABSTRACT

*Plants are a wellspring of huge number of drugs trading off to various gatherings like antispasmodics, emetics, anti-cancer, anti-microbial and so forth The WHO assessed that 80% of the number of inhabitants in non-industrial nations actually depends on traditional medicine, generally plant drugs for their essential medical service's needs. The variety Bougainvillea was named after the world explorer, Louis de Bougainville, who found it in Brazil in eighteenth century and carried it to Europe where it got both far and wide and famous, because of its flexibility, extravagance and reasonableness to flourish in corrupting natural conditions. Bougainvillea is a far and wide gathering all through the world. It has a place with the family Nyctaginaceae and, as per "The Plant List", contains roughly 18 species are industrially misused. Bougainvillea species have been appeared to have flavonoids, alkaloids, saponines, cardiac glycosides, and beta-cyanins. Momordin IIc and two quercetin glycosides were confined from *B. spectabilis*. Also, n-octacos-9-enoic corrosive 8 was disconnected from the foundations of *B. spectabilis*. Bougainvillea glabra is a decorative flowering plant from the variety of bougainvillea; family Nyctaginaceae and a local to Brazil. The examination for medicinal plants found different normal plants having discernable medicinal properties, among which one is Bougainvillea glabra. A few*

species of Bougainvillea have arisen as wellsprings of traditional medicine in human wellbeing. Past phytochemical examination of methanolic concentrate of Bougainvillea glabra leaves (MEBG) has demonstrated the presence of flavonoids, steroids, glycosides and terpenoid kinds of mixtures. Since these builds are of pharmacological interest, combined with the utilization of this plant in traditional medicine, incited us to check Bougainvillea glabra leaves for conceivable pain relieving anti-provocative and anti-pyretic exercises. This Research study will investigate the Pharmacognostic Analysis of Bougainvillea Glabra.

I. INTRODUCTION

Assurance of the purity, consistency and potency of medicinal plants and herbal products has become an important topic since the promotion of herbal medication has happened. The identification of the plants and seasonal change (which has an effect on the time of collection), ecotypic, genotypic, and chemotypic variations, drying and storage conditions, and the prevalence of xenobiotics are all variables that can trigger major discrepancies in the herbal raw material. The word "standardization" applies to the method of verifying a drug's origin as well as assessing its efficacy and purity. In order to certify the quality management of herbal products, standardization is a key step. Standardization of herbal raw drugs requires passport details for raw plant drugs, botanical verification, microscopic and molecular analysis, chemical structure recognition using different chromatographic techniques, and entire plant biological activity. Bougainvillea glabra is used to treat helminthiasis, diabetes, respiratory disease, cough, cold, and bronchitis ulcers, and diarrhoea in several parts of the world, according to ethnomedical details. The pharmacological functions of Bougainvillea glabra include analgesic, antipyretic and anti-inflammatory activities. "The plants have antibacterial action against Escherichia coli, gram positive bacteria, Bacillus subtilis and gram negative bacteria. An aqueous extract of Bougainvillea glabra leaves has anti-diabetic and anti-lipidemic properties. Anti-inflammatory steroidal compounds are present in Bougainvillea glabra. Bougainvillea glabra's methanol extract was shown to have important thrombolytic and antihyperlipidemic behaviours. Hydroalcoholic extract from Bougainvillea glabra leaves was found to be potent against certain pathogenic microorganisms.

The aim of this research was to standardize the whole plant of Bougainvillea glabra by testing different parameters. In addition, preliminary phytochemical screening and thin layer chromatography tests were used to classify various phytoconstituents.



Figure: Flowers and Leaves of Bougainvillea glabra

II. MORPHOLOGICAL CHARACTERS OF BOUGAINVILLEA GLABRA

Botanical Name – *Bougainvillea glabra*

Local Name – *Bougainvillea glabra* commonly name as the paper flower.

Family – Nyctaginaceae

Genus- *Bougainvillea*

Species -*B. spectabilis*

Kingdom - Plantae

Order - Caryophyllales

This genus has 18 species:

B. berberidifolia, *B. buttiana*, *B. campanulata*, *B. glabra*, *B. herzogiana*, *B. infesta*, *B. lehmanniana*, *B. lehmannii*, *B. malmeana*, *B. modesta*, *B. pachyphylla*, *B. peruviana*, *B. pomacea*, *B.praecox*, *B. spectabilis*, *B. spinosa*, *B. stipitata*, and *B. trollii*, with three that are horticulturally important which includes *B. spectabilis*, ***Bougainvillea glabra*** and *B. peruviana*.

Nomenclature

Bougainvillea glabra is a native plant of South America that spread throughout the tropical and warm climates. The vernacular name of *B. spectabilis* known as paper flower (English); baganbilas (Bengali); maobaojin, ye zihua (Chinese); bougainvillaea (French), booganbel (Hindi); buganvillea (Italian); bungakertas (Indonesian); felila (Japanese); bouganvila (Konkani); buginvila (Malay); cherei (Manipuri); veranera (Spanish); bogambilya (tagalog); kagithalapuvvu (Telugu); fuang fah (Thai); and bong giay (Vietnamese)

Useful Parts of Plant

The Mandsaur traditional practitioner used *Bougainvillea glabra* for a number of diseases such as diarrhea, minimizing acidity of the stomach, cough and sore throat, drying up of blood flow, dried flora and leucorrhoea and hepatitis decoction of stem. Leaves are the key ingredient used. The leaves are known to be antimicrobial, anti-inflammatory, and anthelmintic. Pinitol, betacyanine, flavonoid, tannins and alkaloids are the recorded active ingredients in plants.

Phytochemical constituents

Alkaloids, flavonoids, tannins, saponins and proteins are the components of the leaf.

The existence of the alkaloid, flavonoid, furanoid, glycoside, phenol, phlobotannin, quinonine, saponin, steroid and tannin from the stumps, flowers and the leaves of *Bougainvillea* was discovered by a phytochemical study. Bougainvinonespeltogynoids, basic oils such as methyl salicylate, terpinolene, alpha-(E)-ionone, pinitol, β -sitosterin, quercetin, and quercetin-3-O-rutinoside are other active components. In comparison, leaf extract phytochemicals shows that tannins (27.64%), saponins (14.08%), glycosides (11.49%), flavonoids (10.05%), alkaloids (4.10%), phytas (49.27%) and oilseed oxalates (27.65%).

Habit and Botanical Description and Identification Features

The trunk is an eternal woody grape, with multifreeze and wide clumping trunks that reach up to 2-4 m. It climbs by sending out slender, angled thorn-arching canes. The color of the stems turns from mid-green to dark green, brown during development. Bark and Corky are pale. The leaf lengths 5-10 cm and width 2-6 cm with rounded ovate forms. The leaves are green deep, textured leathery with underlying hairy. The leaf axils comprise a cluster of three flowers. They are slender cream in color, with hairy pipes and spectacular surroundings. The pink, very large and egg-shaped bracts, colourful, rusty-red, magenta, and purple. They are often coloured. The fruit is less than 1-2 cm long with an expanded five lobe acene. It is very rare and has a dry hard fruit shell. It is not showy. It is thorny woody weeds that are about 1-12 meters long and scrap over plants other than crochet thorns. They are always lush, where

precipitation takes place all year round or poop while the season is dry. The leaves of the ovate have an alternating length of 4-13 cm. The plant's actual flower is tiny and usually white, but three to six bracts with the vibrant colours of the plant are rose, magenta, purple, red orange, white or pink, each of which is surrounded by three clusters of flora [1].

Bougainvillea is a shrubby and always-green tropical and subtropical wood. The habit of the circular plant with a height that reaches up to 20 foot, typically numerous or clumping stalks. It climbs by sending out slim, steeply curved arched canes. Easy leaves and the ondulate leaf set are alternating. The blade has a circumference of 2–4 cm, and has several common shapes: globular, elliptic, obviated, ovate or cordate. While certain cultivars have variegated leaves, they are mid-too deep gray. The true, perfect flowers are small, tubular and with colourful bracts. Bougainvillea's bright colours are not the unsightly, mostly white or yellow bulbs, rather the bracts around each bulb [2].

Traditional Use

The main part used is leaves. The reported constituents in leaf are alkaloids, flavonoids, tannins, saponins and proteins. The leaves of *Bougainvillea glabra* choicy are reported to have insecticidal activity, anti-inflammatory, antidiarrheal activity, anti-hyperglycaemic activity, anti-ulcer, and anti-microbial activity.

Cultivation and collection

Bougainvillea can tolerate hot dry locations, with temperatures over 100°F. It does well in locations with a minimum of 65°F at night and 75–95°F during the day. *B. glabra* can tolerate slightly cooler conditions (58– 64°F) than *B. spectabilis* (64–68°F). Bougainvillea does best with at least 25 inches of rainfall per year. Bougainvillea grows well in rich, well drained, acidic (pH 5.5–6.0) soil. It does not thrive in soil that is constantly wet. Proper soil pH is essential because it affects the availability of mineral elements. A soil pH above 6.0 increases the possibility of micronutrient deficiencies, particularly iron. Bougainvillea is drought tolerant, salt tolerant, and wind resistant the bark will rub off at ground level when stems whip in high-speed winds. The plant is slow to recover from this, compared to other shrubs.

Environment

Bougainvillea will flower sooner and more profusely with high light intensities moderate temperatures and longer nights. Short day lengths enhance flowering: 8– 11-hour day lengths with high light intensity and temperatures above 58–64°F. Heavy shade inhibits flowering. Drought stress can stimulate flowering even under long day lengths.

Growth regulators

After the first pinch and another 24 hours after the second pinch. Dikegulac sodium at 2 ounces/gallon can be used in lieu of the second pinch-plus-BA application to improve branching commercial formulation of dikegulac sodium (18.5% active ingredient) has been used at 1 ounce/gallon. Sprays should be applied to unpinched shoots when they reach 3 inches or to pruned plants three days after pruning. Avoid treating plants that are under stress. To retard growth Cycocel (chlormequat) has been used on potted bougainvillea as a soil drench at 0.01– 0.02 ounce per pot when the axillary buds swell following the first pinch. (Paclobutrazol) are also effective it is recommended that you test plant growth regulators on just a few plants before extensive use.

Propagation & Cuttings

For propagation usage may be made of softwood endpoints maturing green wood and matured intermediate stem parts of wood. Stem cuts can have a thickness of 1/8 inch or more and should be three to five nodes or more. Leaves can be left on the cuttings at rooting but extract leaves from areas of the stem below the rooting medium surface. Using a well-drained root media like a peat perlite mixture from 1:1 (per volume). Other rooting media like man-made sand and peat or coir perform well. Insert 1–2 inches of cuttings thoroughly in the medium and water. Cuttings may be specifically rooted in jiffy-7 or shallow jip pans. In larger pots of 5-6 inches in diameter, multiple cuttings may be rooted together.” Even blocks may be used for fum propagation. Depending on the community and the application of the rooting hormone, “the period for rooting takes between 4-12 weeks. To prevent harm to the broken roots, transplant young plants with care. When stitching cuttings and transplants again, a wide-spectrum fungicide drench helps to avoid root rot [3].

Grafting

Some cultivars that have little or no chlorophyll in their leaves are difficult to grow from cuttings and need to be grafted onto a vigorous rootstock to be propagated. Grafting is useful with delicate cultivars that have fragile root systems. It is also used when it is desired to have multiple cultivars on one plant^[31]

Extraction procedure

The *Bougainvillea glabra* leaves were dried in shade and crush in the grinder. The dried powder was obtained. The dried powdered material was initially defatted with petroleum ether (60-80°C) in a Soxhlet apparatus for 72 hours according to successive solvent

extraction. The petroleum ether extract was dried and collected. The mark was dried and successively extracted with hydro-alcohol (50:50) each for 72 hours. The extract was filtered, and the solvent was removed by distillation under reduced pressure and percentage yield of the extracts were determined.

III. PHYTOCHEMICAL SCREENING

The phytochemical screening was carried out by standard procedures [4]. Different extracts of *Bougainvillea glabra* were subjected to preliminary phytochemical screening for the detection of various phytochemical constituents such as carbohydrates, proteins, amino acids, steroids, tannins, flavonoids, alkaloids, and glycosides.

Several tests are as follows:

A. Test for Alkaloids

Sample (chloroform, ethyl acetate and methanol extract) was evaporated, to the residue dilute hydrochloric acid was added. It was shaken well and filtered. With the extract, subsequent tests were carried out.

Dragendorff's test

A small quantity of the sample was treated with the few drops of Dragendorff's reagent, the appearance of reddish-brown precipitate indicated the presence of alkaloids.

Mayer's test

Sample (2-3 ml) was treated with few drops of Mayer's reagent. The appearance of white precipitate indicated the presence of alkaloids.

Hager's test

Sample (2-3 ml) was treated with Hager's reagent. The appearance of yellow precipitate indicated the presence of alkaloids.

Wagner's test

Sample (2-3 ml) was mixed with few drops of Wagner's reagent. Appearance of reddish-brown precipitate indicated the presence of alkaloids.

B. Test for Proteins

Biuret test (General test)

A test sample (3 ml) was mixed with 4% NaOH and few drops of 1% CuSO_4 solution were added. Violet or pink color not appeared.

Millon's test

A test sample (3 ml) was mixed with 5 ml of million's reagent. The White precipitate is formed. On warming precipitate turn's brick red or the precipitate dissolves giving red coloured solution.

Xanthoprotein test for proteins containing tyrosine or tryptophan.

Test solution (3 ml) was mixed with 1 ml of conc sulphuric acid. White precipitate is formed. It was boiled then precipitate turned yellow. Ammonium hydroxide was added, finally precipitate turned orange.

C. Tests for Amino Acids

Ninhydrin test (General test)

Test sample 3 ml and 3 drops of 5% ninhydrin solution were heated in boiling water for 10 min. The purple colour does not appear.

D. Test for Steroids

Sample 2 ml was mixed with 2 ml of conc Sulphuric acid, it was well shaken then chloroform layer appeared red and acid layer has shown greenish yellow fluorescence.

Liebermann-burchard reaction

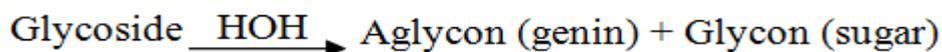
Sample (2 ml) was mixed with chloroform. 1-2 ml of acetic anhydride was added, and 2 drops conc. Sulphuric acid was added from the sides of the tube. First red then blue and finally green colour appeared.

Liebermann's reaction

Sample (3 ml) was mixed with 3 ml of acetic anhydride. It was first heated and then cooled, later few drops of conc. Sulphuric acid were added, and blue colour appeared.

E. Test for Glycosides

Free content of the sugar extract was determined. The sample was hydrolyzed with mineral acid (dilute hydrochloric or dilute sulphuric acid). Again, the total sugar content of the hydrolyzed extract was determined. Increase in the sugar content indicated the presence of glycoside in the extract.



Tests for Cardiac Glycosides

Keller Killiani test

The sample was dissolved in 2 ml chloroform. H₂SO₄ was added to form a layer and the colour at interphase is recorded. Brown ring at interphase is characteristic of deoxy sugars in cardenolides.

In 2 ml extract, GAA and 1 drop of 5% of FeCl₃ and conc H₂SO₄ were added. Reddish brown colour will be seen at the intersection of two liquid layers, and top layer appears bluish green, confirming the presence of glycosides.

Test for saponin glycosides

Foam test: The drug sample or dry powder was shaken vigorously with water. The persistent foam was observed.

Hemolytic test: drug sample or dry powder was added to one drop of blood placed on glass slide. Hemolytic zone was observed.

F. Tests for Flavonoids

Shimoda test: Sample extract was treated with 5 ml of 95% ethanol; few drops of conc. Hydrochloric acid and 0.5g of magnesium turnings were also added. The pink colour was observed. Addition of increasing amount of sodium hydroxide to the residue shown yellow colouration, which decolorizes after addition of acid indicates the presence of flavonoids.

G. Tests for Tannins

The sample was treated with 10% lead acetate solution; the appearance of white precipitate indicated the presence of tannins. When the extract was treated with aqueous bromine solution, the appearance of white precipitate indicated the presence of tannins.

H. Tests for Sterols

The sample was treated with 5% potassium hydroxide solution appearance of pink colour indicates the presence of sterols.

IV. ANATOMICAL FEATURES OF BOUGAINVILLEA

a) Stem

Transverse and longitudinal sections were made of the young stem and stained to help reveal the characteristics of the Bougainvillea anatomy. Starting at the periphery of the stem and progressing inward, numerous multicellular trichomes arise from a single layer of epidermal cells. Just inside the epidermis, there are successive layers of newly arising cork cambium, parenchyma cells, phloem, tracheary elements of the xylem, and a thick band of fibres, all

surrounding the central pith. Embedded in the pith are vascular bundles which are themselves composed of xylem and phloem (Fig. 1A). A closer examination of individual vascular bundles reveals fibres, both removed from and immediately surrounding the vasculature, the latter being composed of both phloem and tracheary elements of the xylem, presumably vessel elements. Interestingly, parenchyma lying near the fibres has thickened, sclerified cell walls (Fig.1B). In very young stems (Fig.1C), angular collenchyma, having primary cell walls thickened in the corners, lies immediately below the epidermis. Longitudinal sections of stem are shown in Figure 1 D, E, F. Fibres and sclerified parenchyma are illustrated in figure 1D. Figure 1E reveals the helical and scaliform secondary cell wall thickenings of the tracheary elements of the xylem. Peripheral to the xylem, phloem sieve tube members are identified by the presence of sieve plates (Fig. 1F). Phloroglucinol staining of a transverse section of a young stem reveals the highly lignified nature of the fibres associated with the xylem (pink staining Fig. 1G). The anomalous secondary growth seen in *Bougainvillea* is illustrated in a transverse section of a more mature stem (Fig. 1H), where individual layers of vasculature have been deposited. A new vascular cambium has been initiated in the parenchyma layer toward the outside of the stem, the successive vascular cambia defining anomalous growth [5].

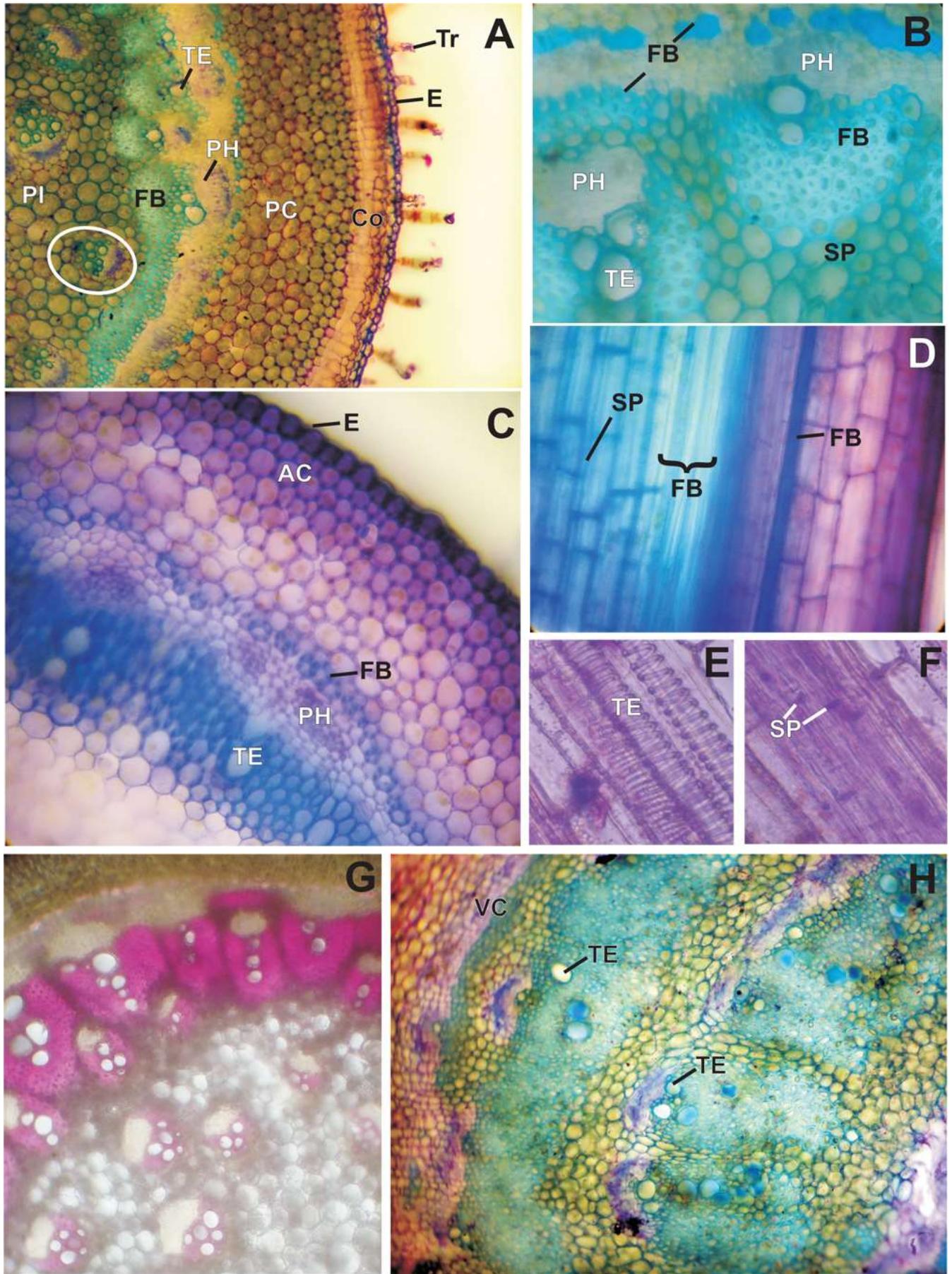


Figure 1. Anatomy of Bougainvillea stem

1A. Transverse section of young stem stained with TBO, and revealing the surface trichomes (Tr), single cell layer thick epidermis (E), and underlying forming cork layer (Co) also known as the periderm, phloem (PH), tracheary elements of the xylem (TE), a ring of fibres (FB) and the pith (PI) containing small vascular bundles (oval).

1B. Magnified view of the vascular bundles revealing a thin layer of fibres (FB towards the periphery of the stem, with a thicker layer capping the phloem (PH). Xylem tracheary elements (TE) are also surrounded by fibres and sclerified parenchyma (SP)

1C. Transverse section of a very young stem stained with TBO. A layer of angular collenchyma (AC) having irregularly thickened primary cell walls lies immediately under the epidermis (E). FB, fibres; PH, phloem; TE, tracheary elements of the xylem

1D. Longitudinal section of young stem stained with TBO illustrating fibres (FB) and sclerified parenchyma (SP).

1E and F. Longitudinal section of stem stained with acid fuschin revealing the helical and scalariform nature of the cell wall thickenings of xylem tracheary elements (TE in 1 E) and sieve plates in the sieve tube members of the phloem (SP in 1F)

1G. Transverse section of a young stem stained with phloroglucinolHCl. Pink staining reveals lignified cell walls of the xylem tracheary elements and fibres.

1H. Transverse section of older stem revealing anomalous secondary growth. Successive vascular regions are added as the stem expands laterally. Two distinct regions of mature vasculature are shown with a tracheary element (TE) of the xylem indicated in each. A new layer of vascular cambium (VC), which will give rise to new xylem and phloem, has been initiated in the parenchyma more proximal to the outside of the stem.

Macerates of the stem stained with TBO similarly revealed the helical secondary wall thickenings in some of the vessel elements (Fig. 2A). Fibres, vessel elements with simple perforation plates, and tracheids with bordered pits (Fig. 2B) and individual raphide crystals (Fig.2C) were also seen.

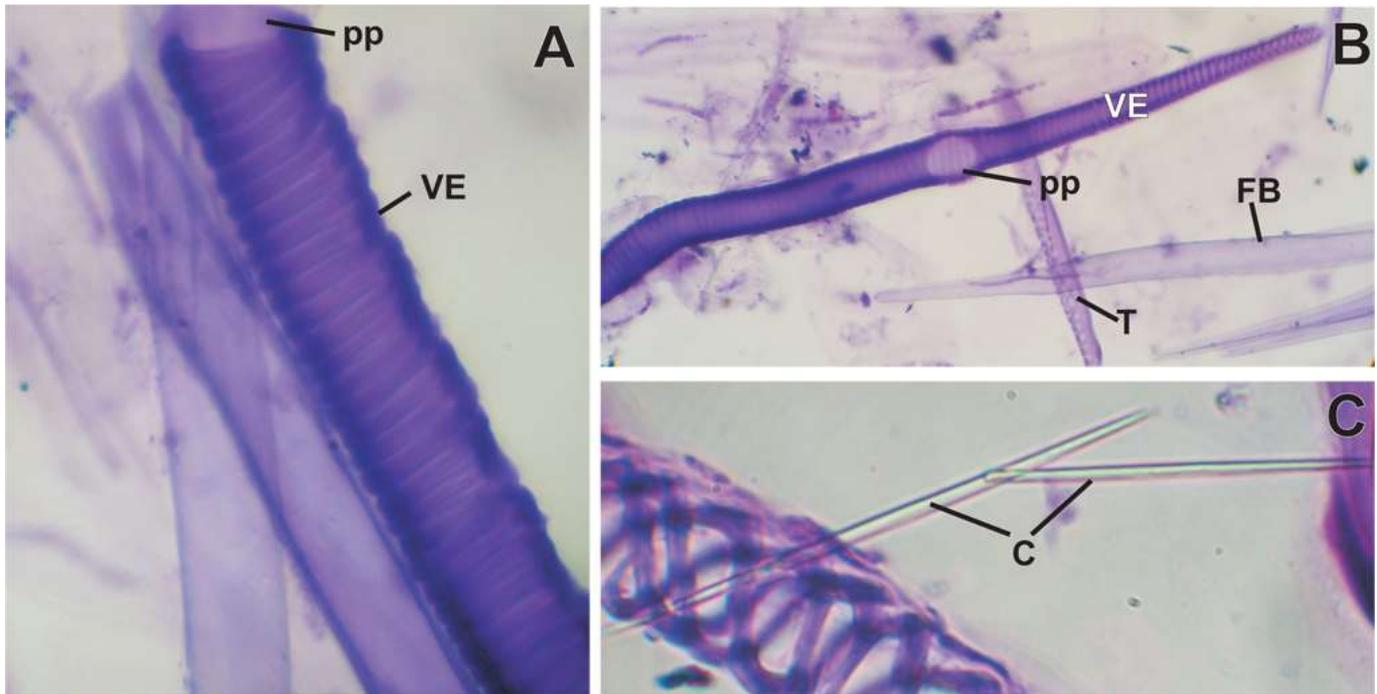


Figure 2. Stem macerates allow identification of elements of the vasculature and raphide crystal. All images stained with TBO.

2A. A vessel element (VE) with simple perforation plate (pp)

2B. Elongated vessel element (VE) with simple perforation plate (pp) lies over a tracheid (T) having bordered pits. FB, fibre

2C. Numerous spear-shaped raphide crystals (C) were found in all macerates

b) Roots

Young roots in primary growth sectioned transversely and stained with TBO, revealed the formation of a diarchvascular arrangement with xylem having two protoxylem poles with metaxylem between. The developing phloem formed two arches surrounding the xylem.” Young roots also showed an extensive number of root hairs (Fig. 3A). “As secondary growth began in the older roots (Fig. 1B), secondary tracheary elements were added outwards from the metaxylem and phloem began to surround the secondary xylem. The formation of a periderm was initiated at this point [6].

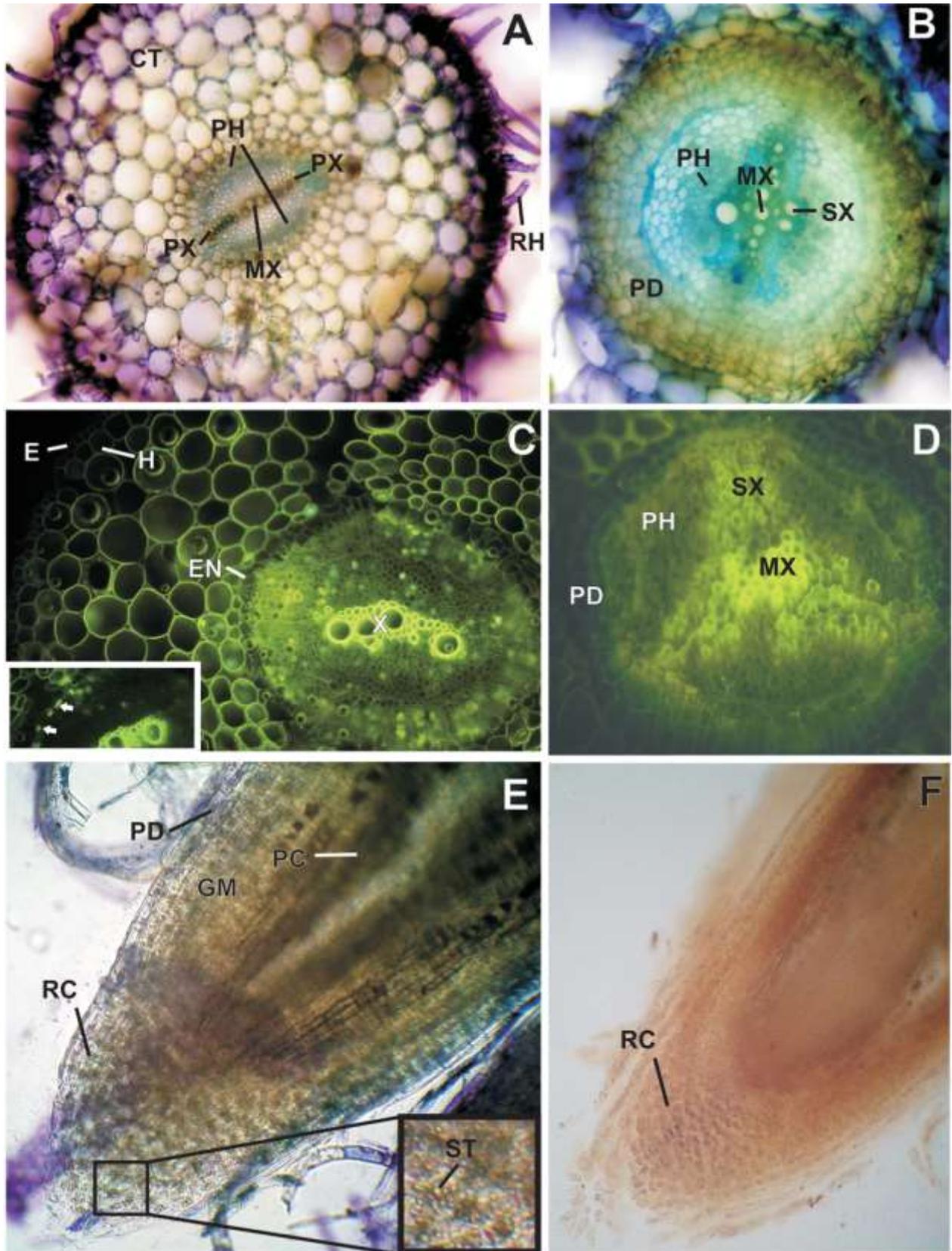


Figure 3. Young roots in primary growth sectioned transversely and stained with TBO.

Roots that were stained with berberine and viewed under UV light confirmed the same results as the TBO, showing the diarch arrangement, cortical parenchyma, hypodermis and epidermis (Fig. 3C). The berberine stain viewed under blue light revealed the diarch arrangement with Casparian bands formed in the surrounding endodermis (Inset Fig. 3C).” It should be noted that a triarch arrangement was also identified in older regions of the root and when stained with berberine and viewed under blue light, tissue having a blue fluorescence, owing to the presence of suberin, was revealed (Fig. 3D).

Longitudinal sections of the root tip stained with TBO revealed root cap cells, protoderm, procambium and ground meristem (Fig. 3E). “A closer view of these root cap cells showed that statoliths are present (Inset Fig. 3E). Longitudinal sections stained with I2KI revealed an open organization of the root system and confirmed that statoliths contain starch (purple/black staining in root cap) (Fig. 3F).

3A. Transverse section of a very young root stained with TBO, revealing the diarch nature of the vasculature. The xylem consists of two protoxylem (PX) poles with metaxylem (MX) between. Archs of phloem (PH) lie outside the xylem. Cortex (CT) surrounds the vasculature, which in turn is surrounded by an epidermal layer having numerous root hairs (RH)

3B. Transverse section of older root stained with TBO. Secondary growth has been initiated as indicated by the addition of secondary xylem adjacent to the metaxylem, phloem (PH) is more developed, and the periderm (PD) is starting to form with the lateral expansion of the root.

3C. Transverse section of young root stained with berberinehemisulfate and viewed under UV light shows the epidermis (E), hypodermis (H), xylem (X) and the region of the endodermis (EN). Viewing under blue light confirms the region of the endodermis by revealing the Casparian bands associated with the cells of this layer (inset, arrows).

3D. Transverse section of old root stained with berberinehemisulfate and viewed under blue light illustrates the expansion of the secondary xylem (SX) around the region of metaxylem (MX). The blue tinge reveals the initiation of a periderm (PD), similar to that seen in 3 B, with suberin in these cells being responsible for the blue fluorescence.

3F. longitudinal section of root tip stained with TBO and illustrating the locations of the procambium (PC), ground meristem (GM) and protoderm (PD). The apical meristem of the root is protected by a root cap (RC) which has statoliths (inset, ST).

3H. Longitudinal section of root stained with I2KI revealing an open organization of the root apex and demonstrating that the statoliths are most likely starch (purple-black deposits in root cap).

c) Spines

Spines of the Bougainvillea plant are very solid structures, which appear green when young (Fig. 4A) and woody (Fig. 4B) when mature. Spines sectioned transversely and stained with TBO revealed a very thick periderm, followed by a ring of fibres, overlaying a ring of sclerified parenchyma. Within this ring, a parenchymatous pith contains some vascular bundles (Fig. 4C), albeit very small (Fig. 4D). Longitudinal sections reflect what is seen in the transverse section, showing fibres and extensive sclerified parenchyma. Note that the sclerified parenchyma have very thickened cell walls (Fig. 4E). A closer view of this longitudinal section reveals some tracheary elements composed of vessel elements and tracheids seen among the fibres, albeit small and difficult to find (Fig.4F).

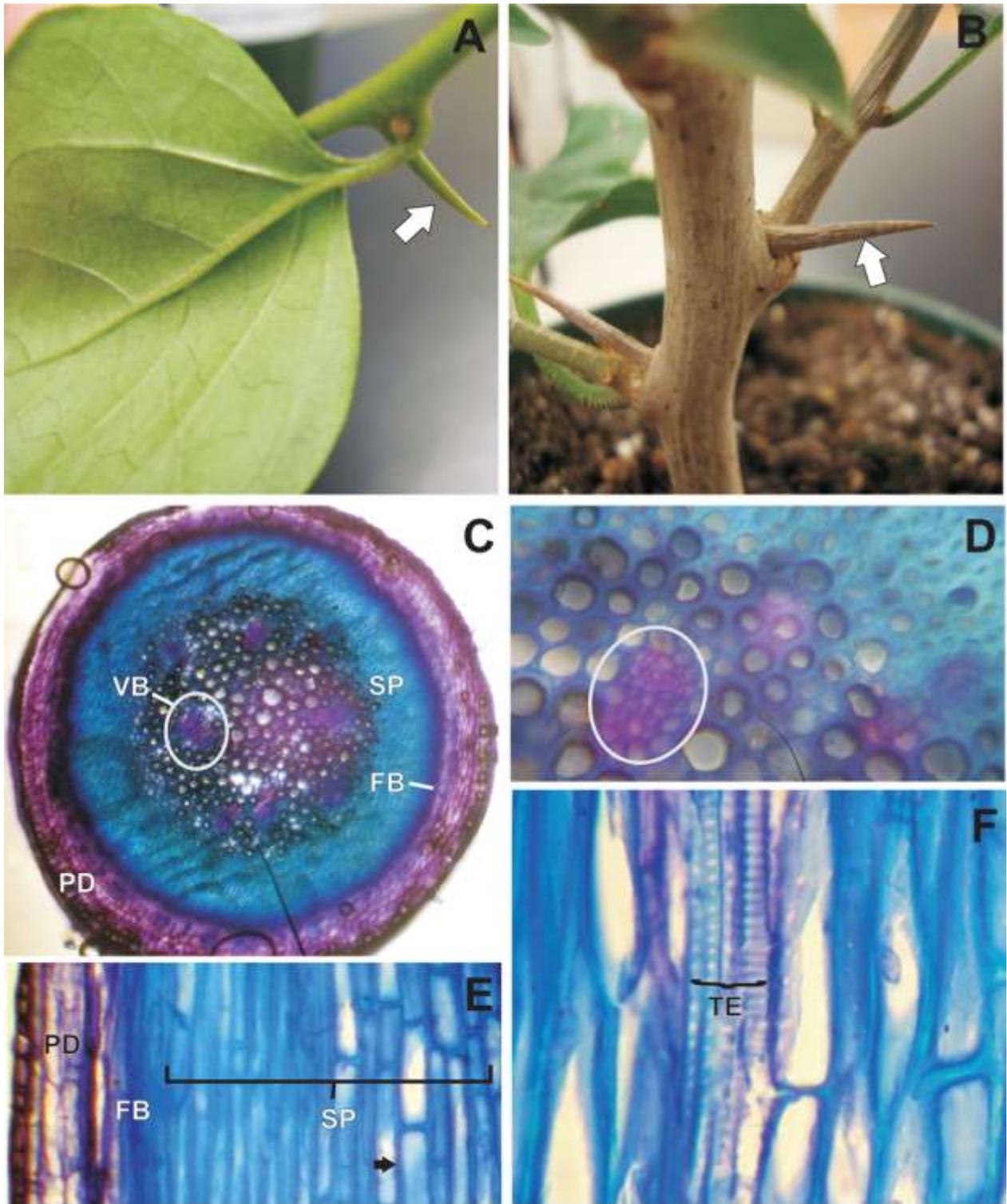


Figure 4. Anatomy of Bougainvillea protective spines

4A and B. Spines arise as modified leaves (arrow, 4 A) and become woody with age (4 B).

4C and D. Transverse section of spine stained with TBO revealing a well-developed periderm (PD) surrounding a thick sclerenchymatous layer composed of fibres (FB) and sclerified

parenchyma (SP). Small vascular bundles (VB) are present in a central pith-like region (4 C). The vascular bundles contain relatively few cells (oval, 4 D) [7].

4E. Longitudinal section of spine stained with TBO and focusing on the sclerenchymatous layer reveals long fibres (FB) beside a region of short rectangular sclerified parenchyma cells (SC) having thickened cell walls.

4F. longitudinal section of spine stained with TBO and focusing on the small vascular bundles. Xylem tracheary elements individually have bordered pits (presumably tracheids), and helical wall thickenings (tentatively vessel elements) [8].

d) Leaves

Leaves of the Bougainvillea plant display an alternate, simple arrangement along the stem with an undulate leaf margin and ovate shape (Fig. 5A). Leaves and bracts that were sectioned transversely and stained with TBO revealed a variety of structures including the midvein (xylem and phloem), midrib parenchyma, palisade and spongy mesophyll, covering trichomes and raphide crystal bundles (Fig. 5B). A closer view of the transverse leaf viewed using brightfield and cross polarizing microscopy confirmed that these structures contained crystals (Inset Fig. 5B). Intact leaves viewed under the dissecting microscope revealed the venation pattern of the leaf, showing the midvein, lateral veins, areoles and trichomes (Fig. 5C). A closer view of a cleared leaf stained with TBO revealed numerous multicellular trichomes on the adaxial surface, although trichomes have been found on both surfaces (Fig.5D). Epidermal peels revealed that stomata complexes are generally found on the abaxial side of the leaf (Fig. 5E) while nail polish impressions further demonstrated this finding (Fig. 5F). Nail polish impressions also revealed that stomata were absent on the adaxial surface and that there were differences in the shapes of the pavement cells found on the abaxial versus the adaxial side of the leaf (Fig. 5G).

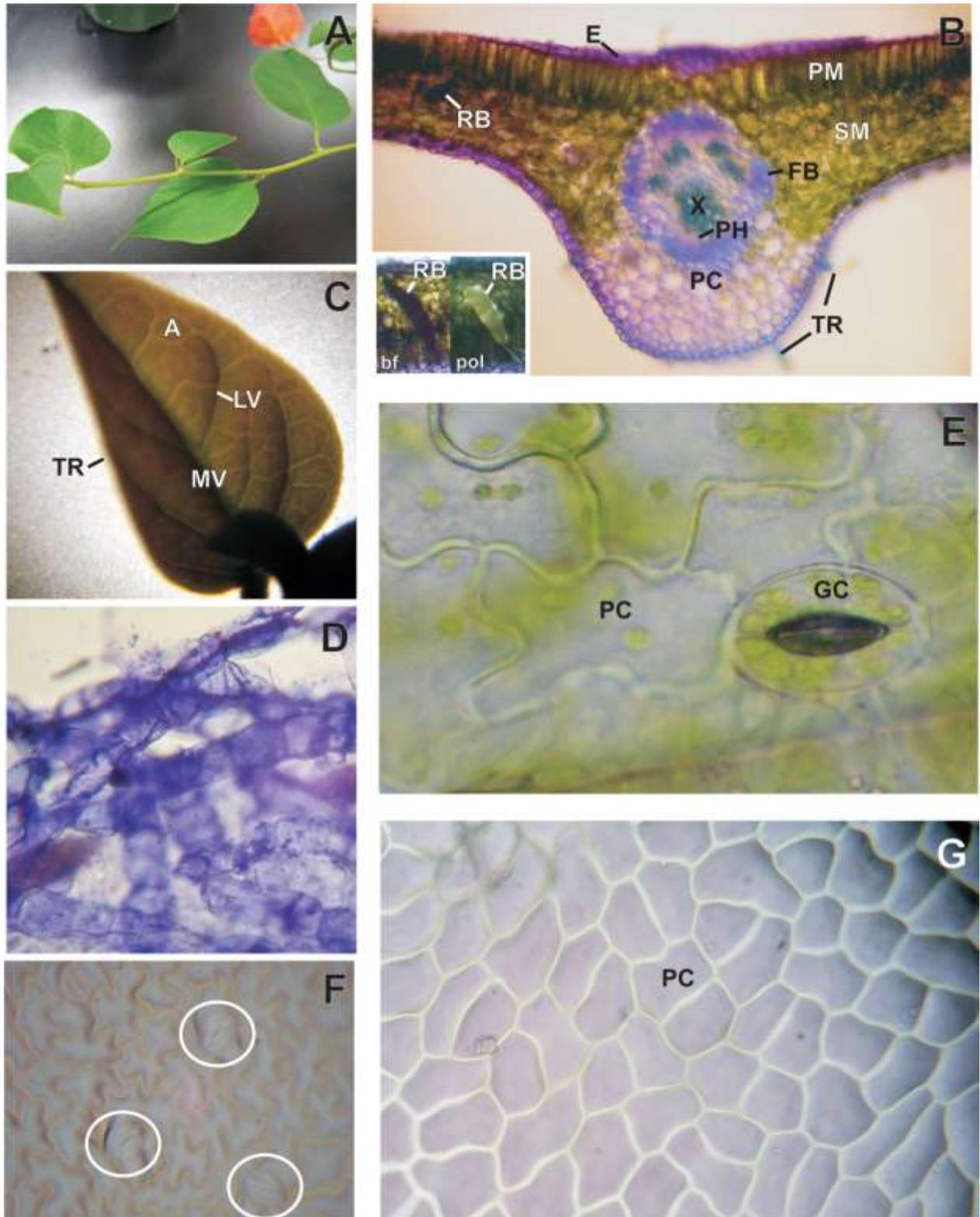


Figure 5. Morphology and anatomy of Bougainvillea leaves

5A. Leaves of Bougainvillea are arranged in a simple alternate arrangement on the stem. Leaves have ovate shape with undulate margin.

5B. Transverse section of leaf through the midvein and stained with TBO reveals a fairly typical arrangement of photosynthetic mesophyll, with a single layer of palisade mesophyll (PM) nearest the adaxial (upper) surface and spongy mesophyll (SM) below. The epidermis (E) is a single cell layer with multicellular trichomes (TR, difficult to distinguish in this section). The vascular bundle of the midvein has regions of xylem (X) and phloem (PH) surrounded by scattered regions of fibers (FB). There is an extensive layer of parenchyma tissue (PC) between the lower epidermis and the vascular bundle, which may aid in support. Bundles of raphide crystals (RB) are scattered throughout the mesophyll, the inset showing that the dense structures seen in brightfield microscopy (bf) contain crystal structures as observed under cross polarized light (pol) [9].

5C. Intact leaf viewed under dissecting microscope shows net-venation pattern of the Bougainvillea leaf, with mid-vein (MV), lateral veins (LV) and areoles (A). Trichomes are numerous on the abaxial surface.

5D. Close up view of cleared leaf stained with TBO shows the very numerous multicellular trichomes of the leaf, the only structures seen in this image.

5E. Close up of epidermal peel of the abaxial surface of the leaf illustrating the guard cells (GC) of the stomatal complex and pavement cells (PC) of the epidermis. The stomatal complex lacked obvious subsidiary cells.

5F and G Nail polish impressions of the abaxial and adaxial leaf surfaces illustrate that stomatal complexes exist only on the abaxial surface (ovals, 5F). Pavement cells of the abaxial surface have the shape of jigsaw puzzle pieces, whereas those on the adaxial surface (PC) are more regular in shape.

e) Flower

Longitudinal sections of the flower left unstained revealed the internal anatomy including the perianth, filaments, anthers, and pollen grains (Fig. 6A). On the external surface of the perianth, pink purple trichomes were evident. A macro photo shows the tubular shape of the flower, surrounded by the bright pink bract (Inset Fig 6A). Cross polarization of the flower also revealed these pink purple trichomes as well as pollen grains and numerous crystal bundles seen throughout the entire organ (Fig. 6B).

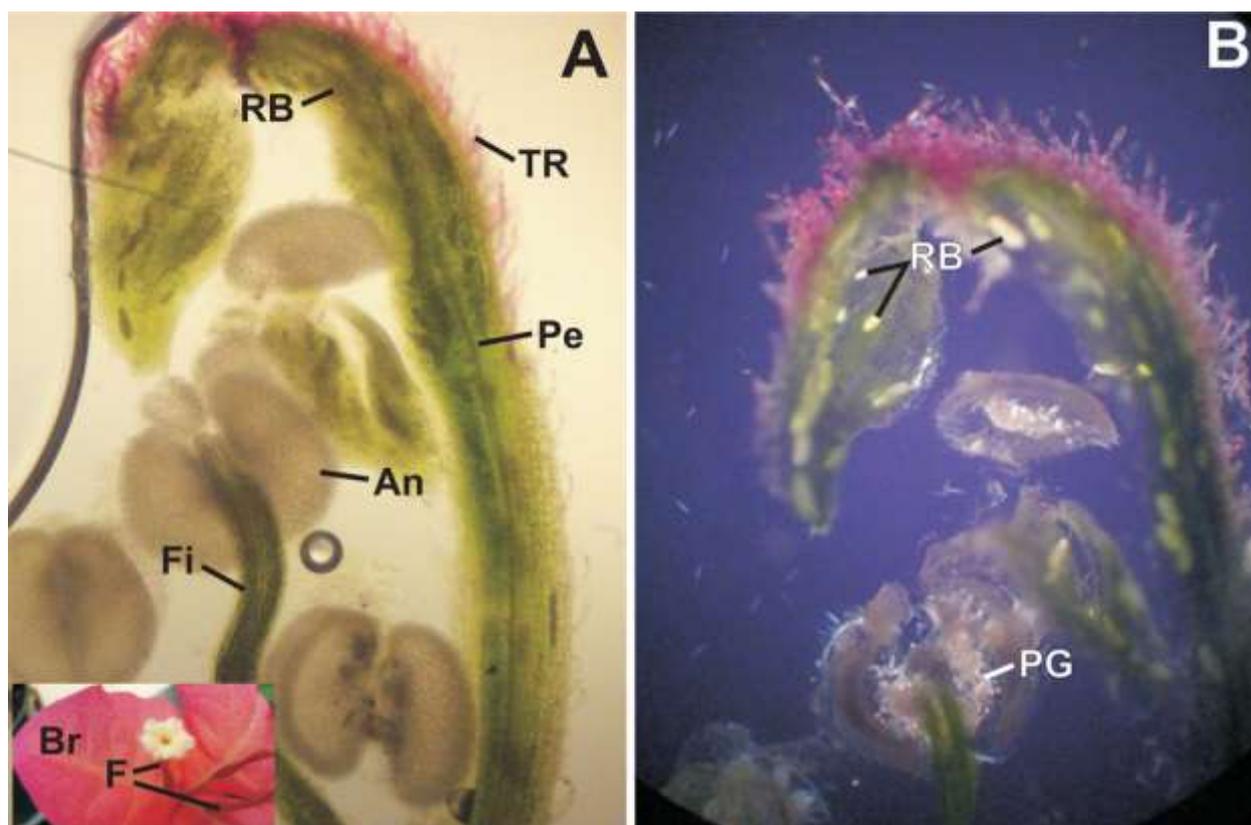


Figure 6. Morphology and anatomy of *Bougainvillea* flowers

6A and B. *Bougainvillea* flowers are perfect flowers (F) that reside in groups of three above colourful leaf-like bracts (Br, inset 6A). The fused perianth is slender and tubular in nature (inset 6A, PE 6A) and has numerous trichomes (TR) on the outer surface (6A). Stamens with anthers (AN) and filaments (Fi) were obvious within the sectioned flower.” As with the leaves, bundles of raphide crystals are present in the perianth, presumably as a protective measure against herbivores. “6A, bright field unstained, 6B cross polarizing microscopy. Pollen grains (PG), having thick almost crystalline cell walls are revealed using polarized light (6B).

V. STANDARDIZATION

a) Loss on drying/Moisture content

Place about 3.0 grams of *Bougainvillea glabra* plant powder, in an accurately weighed moisture disc. For estimation of loss on drying, it was dried at 100 °C for 4 h in an oven, then it was cooled in a desiccator for 30 min, and it is weighed without delay. The loss of weight was calculated as the content in mg per g of air-dried material.

b) Determination of total ash

Bougainvillea glabra plant powder (5 gm) was placed in a previously ignited (300 °C for 45 min) and tarred crucible accurately weighed. The dried material was spread in a uniform layer in the container and the substance was ignited by gradually increasing the temperature to 400 °C for 4 h in a muffle furnace (Nabertherm) until it was white, indicating the absence of carbon. Then it was cooled in a desiccator and weighed. Total ash content was measured in mg per gram of air-dried material.

c) Determination of water-soluble ash

The jet containing the complete ash, wrapped in a watch glass, and softly cooked for fifteen (15) ml was applied to the container for 15 minutes. Insoluble content on an ash less filter paper was obtained. It was then washed with hot water and ignited in a muffled furnace for 10 minutes at 400 °C. The residue should refresh for 20 minutes and then weight without pause in an effective desiccator. The residue's weight has been decreased to mg by the gross ash weight. Content for water-soluble ash per g of air-dried material has been measured as mg.

d) Determination of acid-insoluble ash

In a crucible containing the complete ash fifteen ml (15) of hydrochloric acid (60g/l) was applied. It had a watch glass sealed and then softly boiled for fifteen minutes. A tiny volume of hot water was applied to the watch glass and the solvent was added to the drain. The insoluble matter had been collected on a less filtered casserole (Whatmann-41) and then cleaned with hot water until neutral. In a muffle oven (Nabertherm), a steady rise in heat at 450°C for 2 hours was brought through to the initial crush, then the filter paper comprising the insoluble substance was moved into the crushed material [10]. The residue should refresh for 20 minutes and then weight without pause in an effective desiccator. The acid-insoluble ash content of the air-dried material has been measured as mg per g.

e) Determination of pH range

The pH of different formulations in 2% w/v (2g: 100 ml) and 20% w/v (20g: 100 ml) of water-soluble portions of whole plant powder of *Bougainvillea glabra* were determined using standard simple glass electrode pH meter.

f) Determination of hot water and ethanol-extractable matter

Placed separately in a precisely weaved, glass stopped conical flask, roughly 2.0 g of whole plant powder in the *Bougainvillea glabra*. 50 mL of purified water was applied to the container for the calculation of hot-water extractability and measured for overall weight including bottles. The ingredients were well shook and stood for 45 minutes. A flask reflux

condenser was attached and softly heated for one hour. The flask was adjusted with purified water for its initial overall weight and well shook and screened easily with a dry filter.

Then 15 ml of the filtrate was moved onto a precisely weighted, tarred, flat-bottomed dish (petri disc). Finally, it was dried in the oven for 4 hours at 100°C and weighed without pause in a desiccator. The usage of ethanol instead of distilled water to regulate extractable content in ethanol has been supervised using a similar method. The extractable substance was measured as mg per gm of air-dried material.

Three times were calculated various physicochemical constants and the average values were reported. For certain parameters the plant extract displayed various values. With respect to drying failure, 8.1 ± 0.24 and a sum of ash value of 6.26 ± 0.05 are reported for *Bougainvillea glabra*." The findings were 1.35 ± 0.03 and 1.10 ± 0.07 respectively for insoluble acid ash and sulphated ash. The pH of a raw drug reveals when the medication is consumed in the bowel or stomach. The pH of the various formulations of whole plant powder *Bougainvillea glabra* was observed to be 4.12 ± 0.03 and 2.92 ± 0.05 respectively, in 2 percent w/2/v (2g: 100 ml) and in 20 percent w/v (20g: 100 ml). The extractive values were 23.45 ± 1.13 and 12.32 ± 0.41 for both water and ethanol. It was observed that water-soluble ash was 1.34 ± 0.04 . "The findings from the determination of extractive value reveal that the extractive value of Water Soluble was 23.45 ± 1.13 greater than the extractive value of ethanol, 12.32 ± 0.41 . Table 1 revealed the analysis.

g) Determination of solvent extractive values

The table 2 shows the extractive values of *Bougainvillea glabra*.

The air-dried powder of *Bougainvillea glabra* was extracted with different solvent systems like chloroform, ethyl acetate and methanol. The percentage yields of different extracts were found to be 3.8 % 4.2 % and 4.8 % for chloroform, ethyl acetate and methanol, respectively.

Table 1: Physicochemical parameters of *Bougainvillea glabra*

Parameters	Values
Loss on drying	8.15 ± 0.24 W/W
Total ash value	6.26 ± 0.05 W/W
Water soluble ash	1.34 ± 0.04 W/W
Acid insoluble ash	1.35 ± 0.03 W/W
pH of 2% w/v formulation solution	4.12 ± 0.03

pH of 20% w/v formulation solution	2.92±0.05
Water soluble (hot) extractive value	23.45±1.13 W/W
Ethanol soluble (hot) extractive value	12.32±0.41 W/W

VI. DISCUSSION

The appearance of many onto genetically linked yet separate cambias characterized anomalous growth. Per year parenchymatic cells in the stem are split into a new phloem and Xylus tissue, which are often situated at the periphery and xyls at the center of the stem. As development progresses, due to the behaviour of any individual transition, the vasculature appears arbitrarily distributed. There appears to be a collateral structure of a fibre, sclerenchmya and probable angular collenchyma across these wraps. the organisation of the vascular bundles. These forms of cells have additional defense and support the developing stem. The plant in Bougainvillea is also distinguished by its ripening wood stem and its peridermic layer. Sections of the mature periderm were hard to get. The periderm is usually present in older stones of Bougainvillea and offers additional protection for the stalk as it rises. The phallium suberated will help greatly the Bougainvillea plant, which develops in the hot, dry climate, as it leads to water loss. Quite young shoots had not a mature periderm but a main cell wall epidermal coat. Within this epidermis is the angular collenchyma (characteristic of the stalks) that protects the easily elongating stalk and then chloroplastic parenchyma that reflects the photosynthesis of the young stem. The thin layer of fibers located beyond the vascular bundles covers the outermost phloem layer. A second fiber band of sclerified parenchyma is observed going farther inwards. This dense band of fiber encloses the innermost vascular bundles. When secondary development advances, some of the parenchymatic cells inside and outside the band become meristematic and help develop new vascular cambia and a fiber sheet, granting them additional strength and flexibility as the tree develops.

The patterns of diarch and triarch contained inside the root indicate further that Bougainvillea is a dicotyledonous herb. The two agreements, and the diarch arrangement in the young Bougainvillean origins, were taken by Esau & Cheadles and by Stevenson & Popham. Due to the rising diameter of the old racine and the inclusion of secondarily and floem the transition to triarch arrangement occurred. In the older root parts, a substantial portion of the cortax, including the endodermis and pericylus, was crushed and slackened and the xylis and phloem were left to the surroundings by a freshly developed peridermic sheet to avoid water loss to the atmosphere. As Esau and Cheadle say, a large part of the ploem associated with the usual

secondary xylem rise is crushed during the root expansion, so only the new phloem is enabled.

The Casparian bands exposed portions painted with Berberin and viewed under blue light. Despite Bougainvillea's dry climate, she had modified them so that water could remain safe at her roots. The cortex which tends to have secondary wall deposits in its cell walls is another adaptation that may have helped to maintain water. Statoliths of starch in the root cap cells revealed by TBO and I2KI, which help to lead the downward growth of root, a gravitropism phase.

The sharp structures on the stem are recognized as spines and modified leaves around leaf blossoms. These systems are also part of the defensive mechanism of this facility. Young spines are green and photosynthesis members. As these systems get older and woodder, their photosynthesis is no longer feasible.” The vascular classes are therefore slightly weaker and fail gradually. This helps the plant to move its reserves of electricity to other systems. Finally, a mature spine will comprise mostly of sclerified parenchymas, fibers and a dense periderm, which tends to establish a good defensive and antipestal structure.

VII. CONCLUSION

The present study was taken to thoroughly standardize the weed in compliance with criteria and standard lab protocols of the World Health Organisation (WHO). “The qualitative and quantitative parameters of the Bougainvillea glabra were evaluated for extraction, standardization and phytochemical screening. In order to examine their dominance, protections and standardization for their healthy usage, the whole plant in Bougainvillea glabra was extensively researched on its organoleptic character, physical-chemical characteristics, and main active constituents. Each parameter values have been found to be within the limits prescribed by the WHO. The plant extracts of Bougainvillea Glabra have been contained in chloroform, ethyl acetate and methanol extracts, as shown in phytochemical tests, and TLC. The findings of the research also suggest that the Bougainvillea glabra leaves of the plant may be used to cure different diseases, such as cancer and cardiac diseases etc.” The knowledge produced by the present study shall provide details which will be helpful in recognizing, authenticating, and preventing adulteration of this medicinal plant.

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