East Point College of Pharmacy

East Point Campus, Jnana Prabha, Virgo
Nagar PostBengaluru – 560049, Karnataka

Approved
by
Pharmacy Council of India, New Delhi

Affiliated
to
Rajiv Gandhi University of Health SciencesKarnataka
Bengaluru – 560041 India

LAB MANUAL

PHARMACOGNOSY AND PHYTOCHEMISTRY-II

B. PHARM 5th SEMESTER

EAST POINT COLLEGE OF PHARMACY
East Point Campus, Jnana Prabha, Virgo Nagar Post,
Bengaluru – 560049, Karnataka

B Pharmacy

Program Outcomes (PO's)

PO 1- Pharmacy Knowledge

Possess knowledge and comprehension of the core and basic knowledge associated with theprofession of pharmacy, including biomedical sciences; pharmaceutical sciences; behavioral, social, and administrative pharmacy sciences; and manufacturing practices.

PO 2- Planning Abilities

Demonstrate effective planning abilities including time management, resource management,delegation skills and organizational skills. Develop and implement plans and organize workto meet deadlines.

PO 3- Problem analysis

Utilize the principles of scientific enquiry, thinking analytically, clearly and critically, whilesolving problems and making decisions during daily practice. Find, analyze, evaluate and apply information systematically and shall make defensible decisions

PO 4- Modern tool usage

Learn, select, and apply appropriate methods and procedures, resources, and modernpharmacy-related computing tools with an understanding of the limitations.

PO 5- Leadership skills

Understand and consider the human reaction to change, motivation issues, leadership and team-building when planning changes required for fulfillment of practice, professional and societal responsibilities. Assume participatory roles as responsible citizens or leadership roles when appropriate to facilitate improvement in health and wellbeing.

PO 6- Professional Identity

Understand, analyse and communicate the value of their professional roles in society (e.g.health care professionals, promoters of health, educators, managers, employers, employees).

PO 7- Pharmaceutical Ethics

Honor personal values and apply ethical principles in professional and social contexts. Demonstrate behaviour that recognizes cultural and personal variability in values,communication and lifestyles. Use ethical frameworks; apply ethical principles while making decisions and take responsibility for the outcomes associated with the decisions

PO 8- Communication

Communicate effectively with the pharmacy community and with society at large, such as,being able to comprehend and write effective reports, make effective presentations and documentation, and give and receive clear instructions

PO 9- The Pharmacist and society

Apply reasoning informed by the contextual knowledge to assess societal, health, safety andlegal issues and the consequent responsibilities relevant to the professional pharmacy practice.

PO 10- Environment and sustainability

Understand the impact of the professional pharmacy solutions in societal and environmentalcontexts, and demonstrate the knowledge of, and need for sustainable development.

PO 11- Life-long learning

Recognize the need for, and have the preparation and ability to engage in independent and life-long learning in the broadest context of technological change. Self-access and use feedback effectively from others to identify learning needs and to satisfy these needs on an ongoing basis.

Programme Specific Outcomes (PSO's)	
PSO 1	Acquire a thorough foundational knowledge in pharmaceutical sciences, including pharmacology, pharmaceutics, medicinal chemistry, and pharmacognosy, to excel in further academic pursuits
PSO 2	Gain expertise in the application of contemporary pharmaceutical techniques and technologies, enhancing employability across various sectors including the pharmaceutical industry, academia, and research institutions.
PSO 3	Equip with entrepreneurial skills and knowledge of pharmaceutical business management, including market analysis, product development, regulatory affairs, and financial planning, to initiate and run successful ventures in the pharmacy sector

Course Outcomes (CO's)	
Code: BP508P Pharmacognosy and Phytochemistry-II	
CO 1	To study histology, morphology, powder characteristics and extraction off various crude drugs.
CO 2	Discuss isolation and detection of active principles present in various crude drugs
CO 3	Study of various chromatographic techniques
CO 4	Distillation of volatile oils and detection of by phytoconstituents TLC

Table of Content

MORPHOLOGY, HISTOLOGY AND POWDER CHARACTERISTICS, EXTRACTION, OF TRADITIONAL DRUGS

Experiment-1

MORPHOLOGY AND MICROSCOPY OF CINCHONA

AIM: To study the morphology and microscopy of Cassia Cinnamon.

REFERENCE: M.A. Iyengar, Study of Crude drug, Pg. No: 40

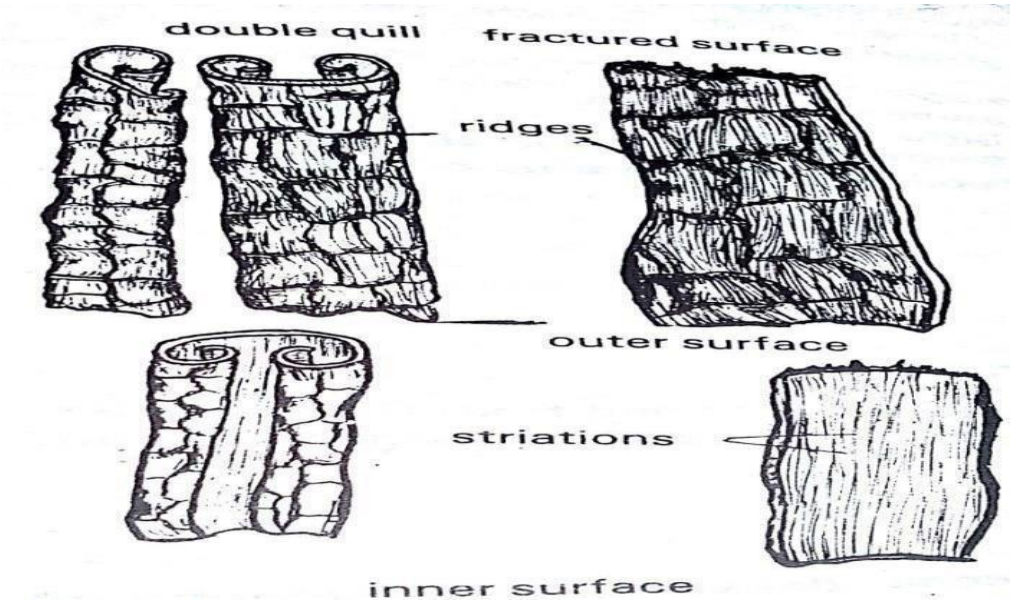
C.K. Kokate, Pharmacognosy, 45th ed., Pg. No: 3.54. M.A. Iyengar, Anatomy of crude drugs, Pg. No. 36. M.A. Iyengar, Powder microscopy of crude drugs, Pg. No. 16.

SYNONYM: Jesuit's bark, Peruvian bark

BIOLOGICAL SOURCE: It consists of dried bark of cultivated trees of ***Cinchona calisaya, Cinchona ledgeriana, Cinchona officinalis, Cinchona succirubra***. It contains not less than 6% of total alkaloids.

Family: Rubiaceae

MORPHOLOGY:



MORPHOLOGY OF CINCHONA

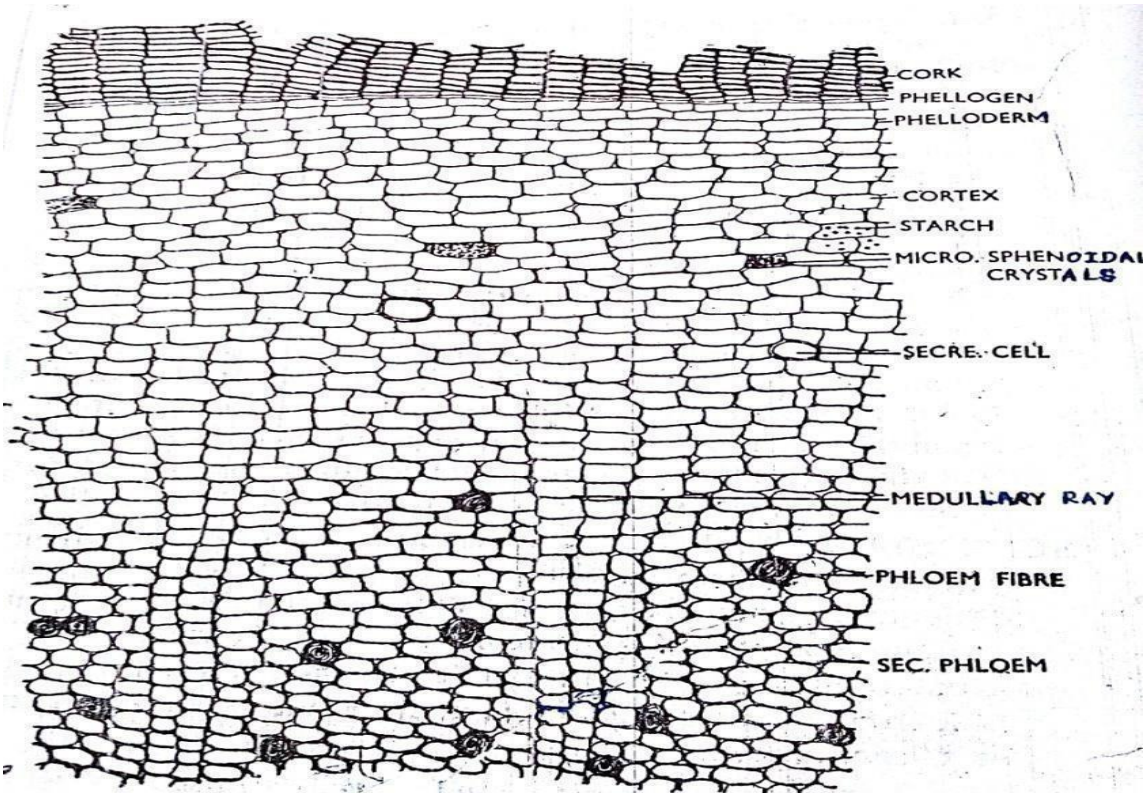
- Condition: dry
- Shape: Flat or channeled, single quill or double quill, curved pieces
- Size: Varies, 12-40 mm in diameter and 1.5-5 mm in thickness
- Outer surface: Yellowish brown or reddish brown
- Inner surface: Red to deep reddish brown
- Odour: Slight and characteristic
- Taste: Astringent and intensely bitter

Chemical Constituents:

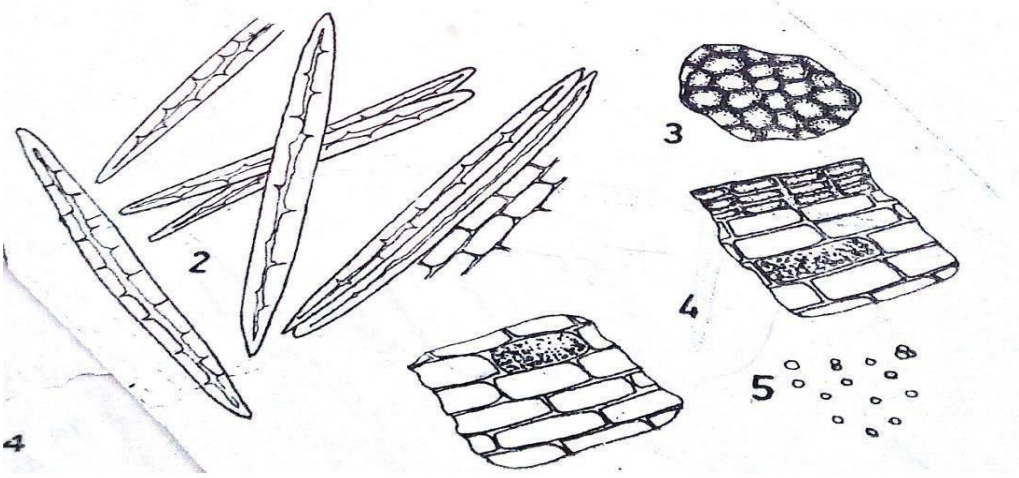
- Quinoline alkaloids (25 alkaloids)
- Major: **Quinine, Quinidine, Cinchonine, Cinchonidine**.
- Minor: Quinicine, cinchonicine, hydroquinine, hydrocinchonidine and homocinchonidine
- Phlobatannin - Cinchotannic acid, on hydrolysis gives cinchona red
- Glycoside – Quinovin
- Bitter essential oil

Uses:

- Antimalarial (Quinine)
- Cardiac depressant (Quinidine)
- Bitter stomachic, Antipyretic



TRANSVERSE SECTION OF CINCHONA



POWDER MICROSCOPY OF CINCHONA

TRANSVERSE SECTION:

Transverse section shows a periderm, a wide cortex and a large secondary phloem.

- **Periderm**
  - **Cork:** Several layers of radially arranged rows of thin walled cells with dark brown contents. The cork cells are impregnated with suberin.
  - **Phellogen:** 2-3 layers of thin walled rectangular cells without any cellular contents.
  - **Phelloderm:** 6-8 layers of thin walled rectangular cells without any cellular contents. Like cork, they are arranged at times in radial rows.
- **Cortex:**

Several layers of thin walled and tangentially elongated cells containing yellowish brown matter. Some of the cortical cells are filled with microsphenoidal crystals of calcium oxalate and the rest with minute starch grains. Besides, isolated secretion cells (latex ducts) are also found in the cortical parenchyma.

- **Secondary Phloem:**

It consists of phloem parenchyma, phloem fibres and medullary rays.

Phloem fibres: Characteristic of cinchona bark occur intermingled with phloem parenchyma and in between medullary rays. Fibres numerous, mostly isolated, at times in groups of 2 -3 rounded to oval, in various sizes, yellow in colour, thick walled, strongly lignified with a small lumen and stratifications.

- **Medullary Rays:** Traverse radially the phloem parenchyma, 1-3 cells wide, extend upto cortex, cells radially elongated and contain starch grains.

POWDER MICROSCOPY

1. **Fibres:** Phloem fibres or bast fibres, numerous either entire or in fragments, spindle shaped, yellowish, thick walled, strongly lignified, porous walls having simple pores or branched pores and measure around 500-1350µ in length and 50-130µ in width.
2. **Cork:** Typical thin walled cork cells which appear reddish brown in colour.
3. **Calcium oxalate crystals:** Microsphenoidal crystals in dark coloured parenchyma.
4. **Starch grains:** Minute, both simple and compound (2-5) and the individual grains measures 3-10µm In diameter.

Experiment-2

MORPHOLOGY AND MICROSCOPY OF CINNAMON

AIM: To study the morphology and microscopy of Cassia Cinnamon.

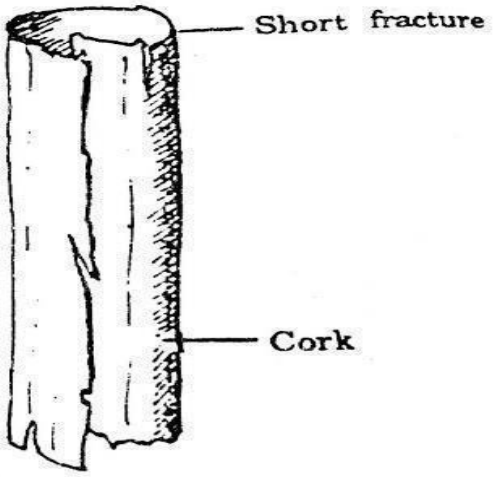
REFERENCE: C.K. Kokate, Pharmacognosy, 45th ed., Pg. No: 1.44. M.A.Iyengar, Anatomy of crude drugs, Pg. No. 34. M.A.Iyengar, Powder microscopy of crude drugs, Pg. No. 15.

SYNONYM: Dalchini

BIOLOGICAL SOURCE: Cassia consists of dried stem bark of ***Cinnamomum cassia*.** It contains not less than 1%v/w of volatile oil.

Family: Lauraceae

MORPHOLOGY:



MORPHOLOGY OF CASSIA CINNAMON

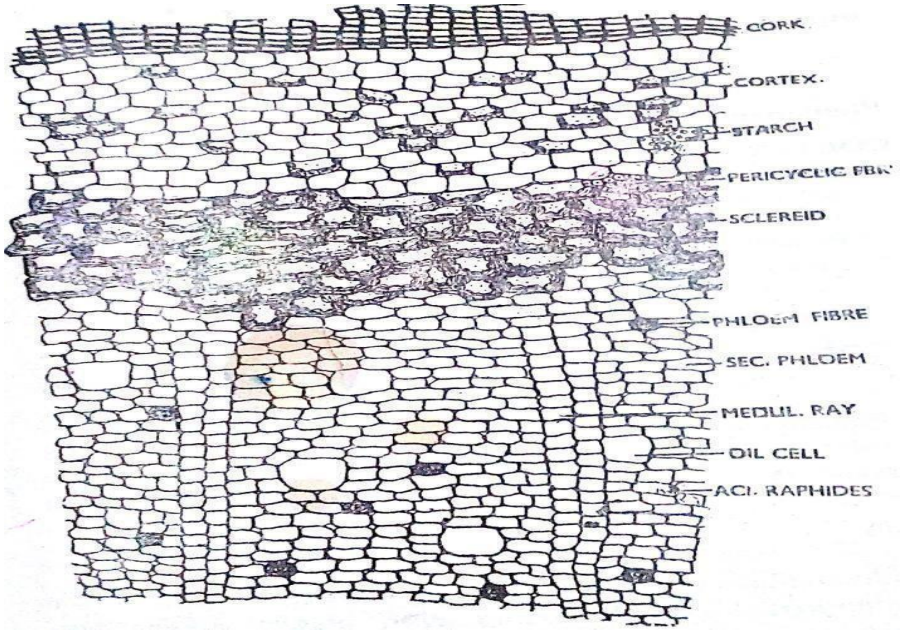
- Condition: dry
- Colour: Dull yellowish brown with grayish patches on cork (outer surface)
  Dark yellowish brown with fine striations (inner surface)
- Odour: Characteristic, Sweet and Aromatic.
- Taste: Slightly sweet, aromatic.
- Shape: Single quills, channeled.
- Size: 5- 40 mm (l), 12-18 mm (w), 1-3 mm (t)

Active Constituents:

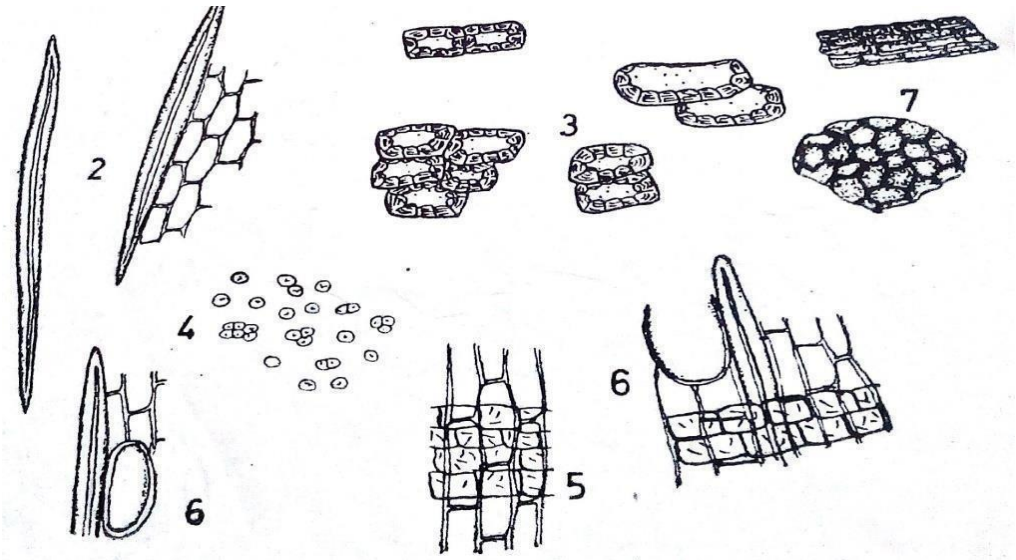
- Volatile oil (1-2%) - **Cinnamic aldehyde** (75-90%)
  Cinnamyl acetate
  Eugenol (small amount).
- Mucilage, starch, calcium oxalate and tannins.

Uses:

- Carminative
- Flavouring agent
- Aromatic
- Stimulant
- Mild astringent



MICROSCOPY OF CINNAMON



POWDER MICROSCOPY OF CINNAMON

TRANSVERSE SECTION:

Transverse section shows a periderm, cortex, a band of sclerenchyma and secondary phloem.

- **Periderm:**
  - **Cork:** The outer few layers are thin walled and the inner ones are lignified and thick walled
  - **Phellogen and Phelloderm:** They cannot be distinguished either from cork.
- **Cortex:** It consists of 10-15 layers of parenchyma in which sclereids are scattered either isolated or in groups. Each scleried is more or less rectangular and pitted with thick inner and radial walls. Some parenchymatous cells contain minute acicular raphides and abundant starch.
- **Sclerenchymatous band or stone cell layer:** A continuous well developed band of sclereids occur in between the primary cortex and secondary phloem. The sclereids are lignified and pitted and are characteristic of cassia bark. The inner and radial walls are thickened than that of the outer walls giving the appearance of the letter U. Towards the outerside of the Sclerenchymatous band, few pericyclic fibres are seen.
- **Secondary phloem:** It comprises of phloem parenchyma, phloem fibres and medullary rays.
  - **Phloem parenchyma** consists of thin walled cells containing abundant starch and few minute acicular raphides. Numerous big isolated oil cells are seen frequently in the phloem region which is a characteristic feature of cassia bark.
  - **Phloem fibres** are mostly single and isolated, rarely in groups of 2 – 3, embedded in phloem parenchyma. The fibres are circular and lignified with stratification.
  - **Medullary rays:** It divides radially several times the phloem parenchyma, 1-3 cells wide and externally extend upto the stone cell layer where they become wider. Ray cells also contain starch and acicular raphides.

POWDER MICROSCOPY:

1. **Odour:** Sweet fragrance
2. **Fibres:** Isolated bast fibres measures 250-600µ in length and 15-30µ in width.
3. **Stone cells:** Almost U shaped as one wall is thinner than the other.
4. **Starch grains:** Abundant starch grains are present measuring not less than 10µ.
5. **Calcium oxalate crystals:** Presence of small, acicular raphides in the parenchyma.
6. **Oil cells:** Big and isolated oval in shape.

Experiment-3

MORPHOLOGY AND MICROSCOPY OF SENNA

AIM: To study primary cell inclusions (Non-living)

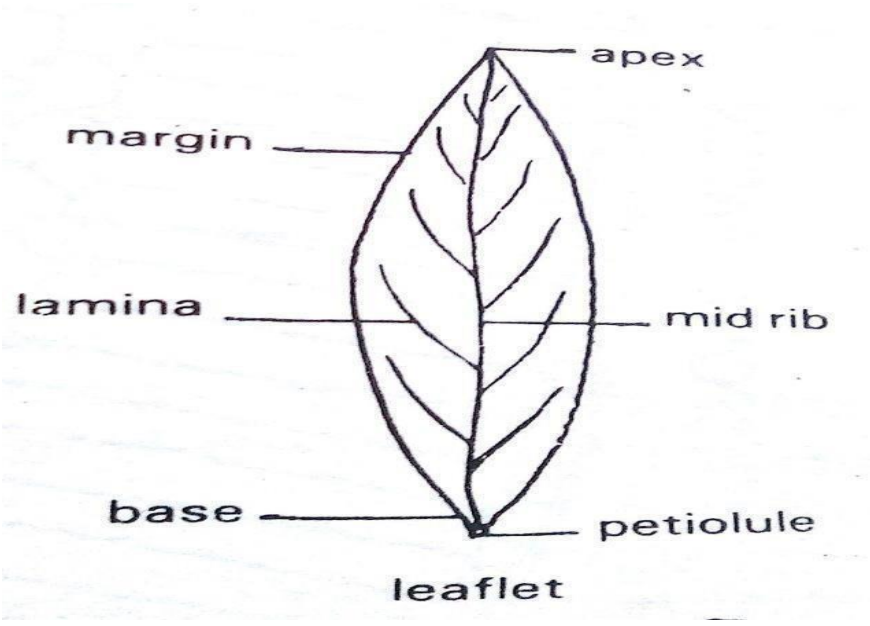
REFERENCE: C.K.Kokate, Pharmacognosy, 45th ed., Pg.No. 8.9. M.A.Iyengar, Anatomy of crude drugs, Pg. No. 49 M.A.Iyengar, Powder microscopy of crude drugs, Pg. No. 27.

SYNONYM: Tinnevely senna, Alexandrain senna, Folia senna, Cassia senna

BIOLOGICAL SOURCE: It consists of dried leaflets of ***Cassia angustifolia*** (Indian Senna) and ***Cassia acutifolia*** (Alexandrain Senna)

Family: Leguminosae

MORPHOLOGY:



MORPHOLOGY OF SENNA

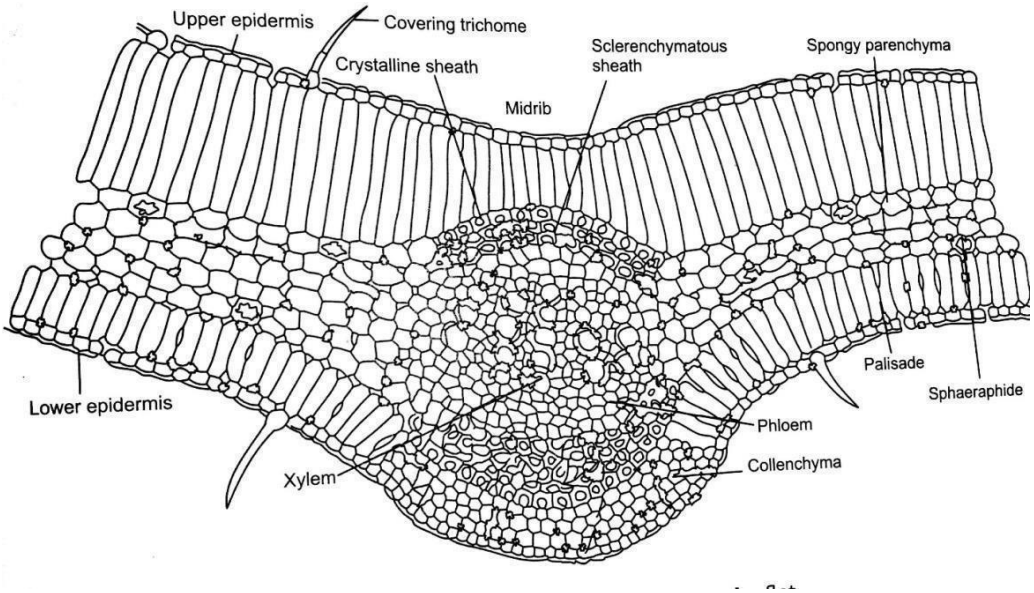
- Colour: yellowish green
- Shape: ovate to lanceolate
- Size: 25-60 mm (l), 7-8 mm (w)
- Odour: no odour
- Taste: mucilagenous and slightly bitter
- Petiole: a small petiole present
- Margin: entire
- Apex: Acute
- Base: Asymmetrical

Active constituents:

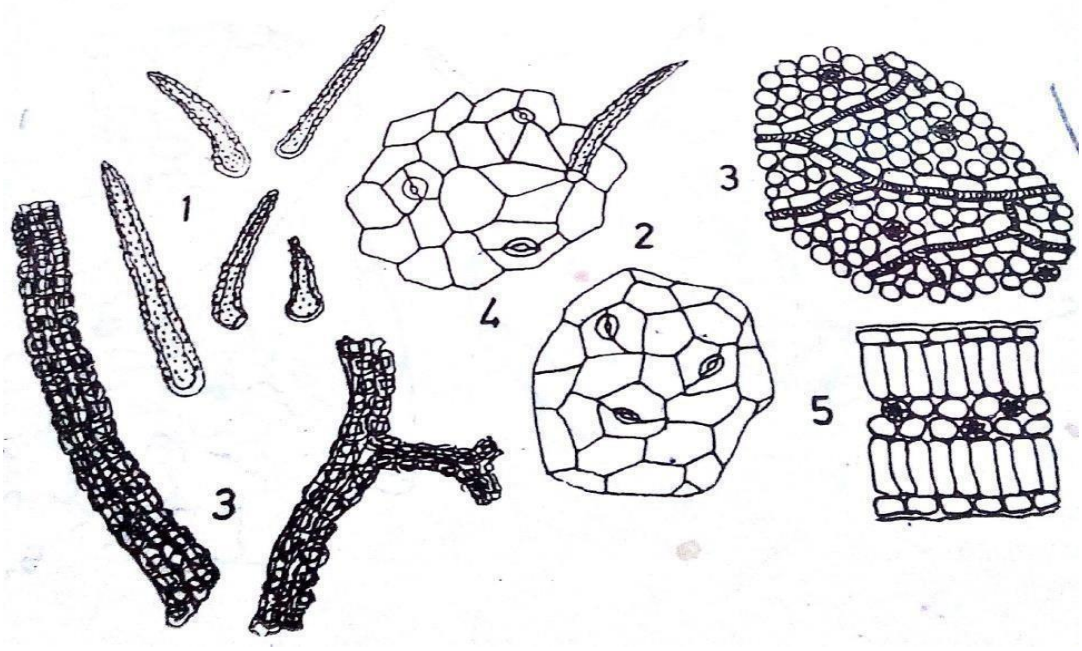
- **Glycosides:** Anthracene – dianthrone glycosides
  - **Sennoside A**
  - **Sennoside B** } Homodianthrones
  - **Sennoside C**
  - **Sennoside D** Heterodianthrones
  - Rhein, aloe emodin
- Flavonoid: Kaempferol
- Mucilage, resin, isorhamnetin, myricyl alcohol, chrysophanic acid.

Uses:

- Purgative in case of habitual constipation
- Given along with carminatives to prevent the gripping action of sennosides.



TRANSVERSE SECTION OF SENNA LEAF



POWDER MICROSCOPY OF SENNA

TRANSVERSE SECTION:

Transverse section of senna leaflet shows an isobilateral condition.

LAMINA:

Upper epidermis:

It is single layered with polygonal cells covered with a thick warty cuticle. Some epidermal cells contains mucilage. Only covering trichomes are seen and the trichomes are non glandular, short, thick, warty, unicellular, nonlignified and slightly curved at the bulbous base. Paracytic stomata are seen at regular intervals.

Mesophyll:

It is differentiated into palisade and spongy parenchyma. Being an isobilateral leaf, palisade is further differentiated into upper and lower palisade.

- **Upper palisade:** It is single layered, compact with elongated, narrow, columnar cells and continues over the midrib also.
- **Spongy parenchyma:** It is thin, narrow, loosely arranged between the upper and lower palisade. Vascular strands are seen frequently. Sphaeraphides are seen in parenchyma.
- **Lower palisade:** It is seen only in the lamina region (absent in midrib). Cells are smaller than those of upper palisade, loosely arranges and their cells are wavy.

Lower epidermis: Is very similar to upper epidermis.

MIDRIB:

It represents a flat ventral surface and convex dorsal surface. The epidermal layers are continuos in the midrib. The cells of lower epidermis are smaller with thick cuticle. The upper palisade cells which occur below the upper epidermis in the midrib region are relatively smaller. The lower palisade is not represented in the midrib, instead a patch of collenchyma is present.

Vascular bundle of collateral type is found in the central portion of midrib with xylem towards the ventral surface and phloem towards the dorsal surface. The vascular bundle is covered on both the sides by a patch of sclerenchymatous fibres and these fibres are ensheathed with a layer of parenchyma, the individual cells of which contains prismatic calcium oxalate crystals (crystal sheath).

POWDER MICROSCOPY:

1. **Trichomes:** Only covering type, short, thick, unicellular, warty and frequently curved near the base.
2. **Stomata:** Rubiaceous or paracytic type meaning thereby the two subsidiary cells are parallel to the stomal pore.
3. **Calcium oxalate:** Occuring as cluster crystals in the cells of the mesophyll and as prisms in a sheath of cells around the fibres and as well freely distributed in powder.
4. **Epidermis:** With polygonal epidermal cells in surface view.
5. **Mesophyll:** Fragments of leaf showing isobilateral arrangements.

Experiment-4

MORPHOLOGY AND MICROSCOPY OF CLOVE

AIM: To study the morphology and microscopy of Clove.

REFERENCE: M.A. Iyengar, Study of Crude drug, Pg. No: 44 .

C.K. Kokate, Pharmacognosy, 45th ed., Pg. No: 1.81.

M.A.Iyengar, Anatomy of crude drugs, Pg. No. 58.

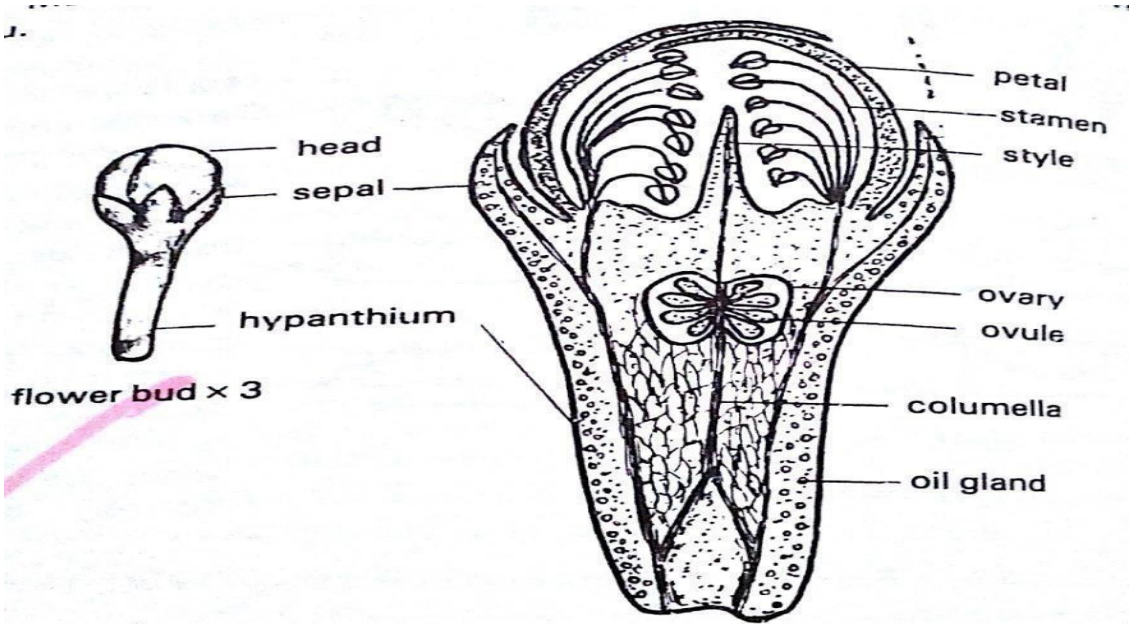
M.A.Iyengar, Powder microscopy of crude drugs, Pg. No. 32.

SYNONYM: Lavang

BIOLOGICAL SOURCE:

It is the dried flower bud of ***Eugenia caryophyllus*** (*Syzygium aromaticum*). It contains not less than 15% of volatile oil.

Family: Myrtaceae



MORPHOLOGY:

MORPHOLOGY OF CLOVE FLOWER BUD

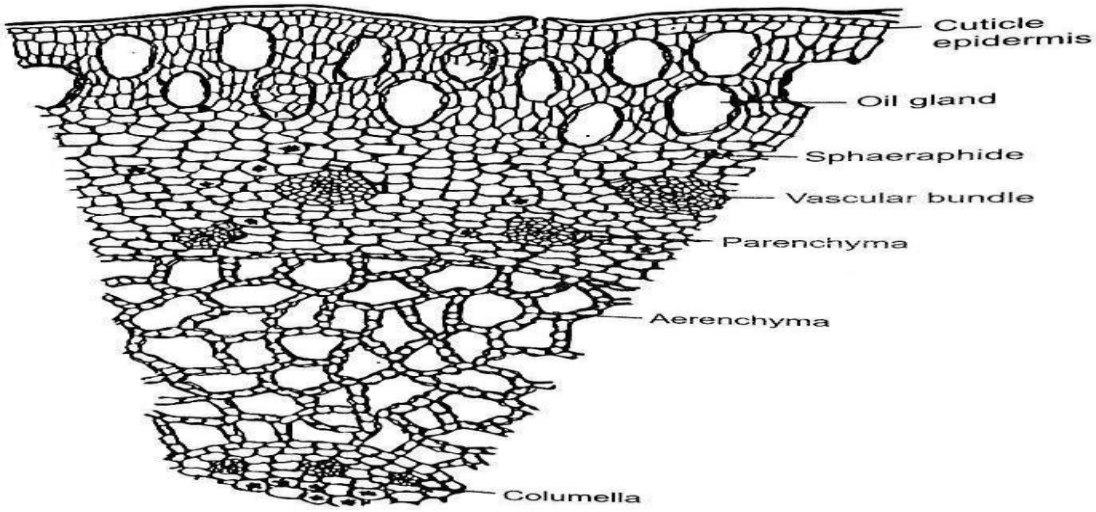
- Shape: Cylindrical hypanthium
  Hypanthium is surmounted with 4 thick acute divergent sepals
  Dome shaped corolla.
- Size: 10-17.5 mm (l), 4 mm (w), 2 mm (t)
- Colour: Crimson to dark brown
- Odour: Slightly aromatic
- Taste: Pungent, aromatic followed by numbness.
- Volatile oil is found in the glands or ducts which is found throughout the flower bud. Cloves are heavier than water.

Active Constituents:

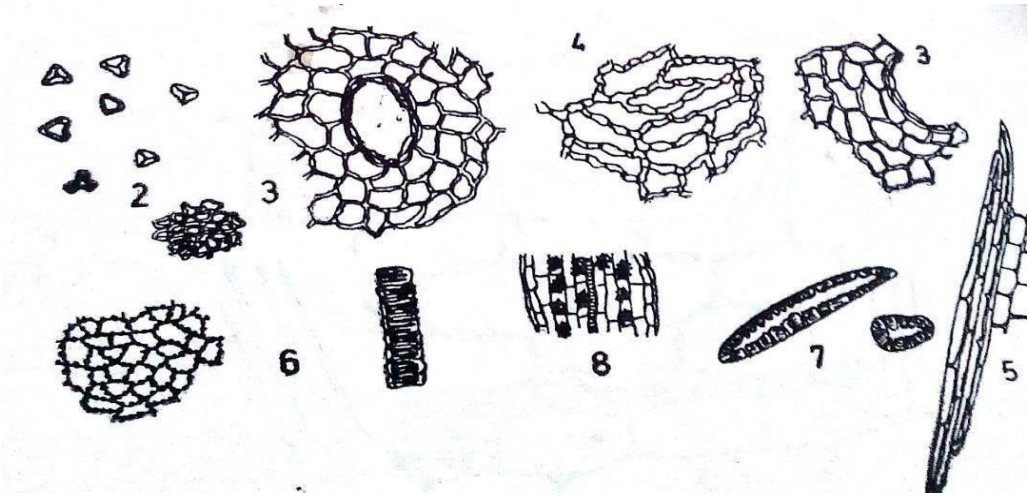
- Volatile oil (15-20%)
  Eugenol (70-90%), eugenyl acetate, caryophyllenes,
  Esters, ketones and alcohols
- Tannins (10-13%) - Gallotannic acid
- Resin, chromone and eugenin.

Uses:

- Carminative, Antiseptic
- Stimulant, Aromatic, Flavouring agent
- Dental Analgesic
- Antispasmodic



TRANSVERSE SECTION OF CLOVE



POWDER MICROSCOPY OF CLOVE

TRANSVERSE SECTION:

Transverse section through hypanthium shows epidermis, cortex and collumella.

- **Epidermis:**

It is single layered small cells with straight walls and has a very thick cuticle. Epidermal layer gets intercepted by ranunculaceous type of stomata.

- **Cortex:** 3 distinct zones can be made out

Peripheral region containing 2 -3 layers of big ellipsoidal, schizo-lysigenous oil glands embedded in the radially elongated parenchymatous cells

Middle region containing 1 or 2 rings of bicollateral vascular bundles associated with a few pericyclic fibres embedded in thick walled parenchyma

Inner region is made of loosely arranged aerenchyma

- **Columella:**

This forms the central cylinder containing thick walled parenchyma with a ring of bicollateral vascular bundles. Numerous cluster crystals are seen scattered throughout the columella and to certain extent in the middle cortical zone.

Transverse section through ovary region:

It shows identical structures as that of hypanthium but for the absence of central columella. This central region is occupied by a bilocular ovary with several ovules showing axile placentation.

POWDER MICROSCOPY:

1. **Odour:** Typical aroma of Eugenol.
2. **Pollen grains:** Small, biconvex with rounded or triangular outline and a smooth exine. Masses of unripe pollen packets are also seen.
3. **Oil glands**: Fragments of parenchyma containing entire or a portion of oil glands.
4. **Aerenchyma:** Portion of loose parenchyma.
5. **Fibres:** Sclerenchymatous fibres associated with parenchymatous cells.
6. **Anther:** Fibrous layer of anther in surface view.
7. **Sclereids:** From the stalk, oval to subrectangular, thickened walls.
8. **Calcium oxalate:** It is in the form of cluster crystals.

Experiment-5

MORPHOLOGY AND MICROSCOPY OF EPHEDRA

AIM: To study the morphology and microscopy of Ephedra.

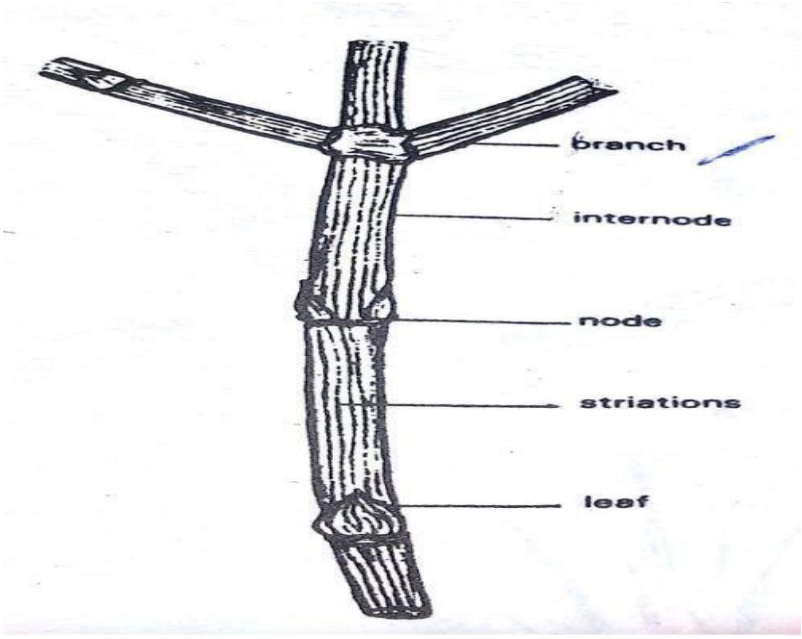
REFERENCE: Pharmacognosy by C.K. Kokate, 45th ed., P.No: 3.61.
M.A.Iyengar, Anatomy of crude drugs, Pg. No. 76.
M.A.Iyengar, Powder microscopy of crude drugs, Pg. No. 53.

SYNONYM: Ma-Huang

SOURCE: It consists of dried young stems of ***Ephedra gerardiana*** and ***Ephedra nebrodensis***. It contains not less than 1.0 % of total alkaloids, calculated as ephedrine. The other species includes ***E. sinica*** and ***E. equisetina***.

Family: Gnetaceae / Ephedraceae.

MORPHOLOGY:



MORPHOLOGY OF EPHEDRA STEM

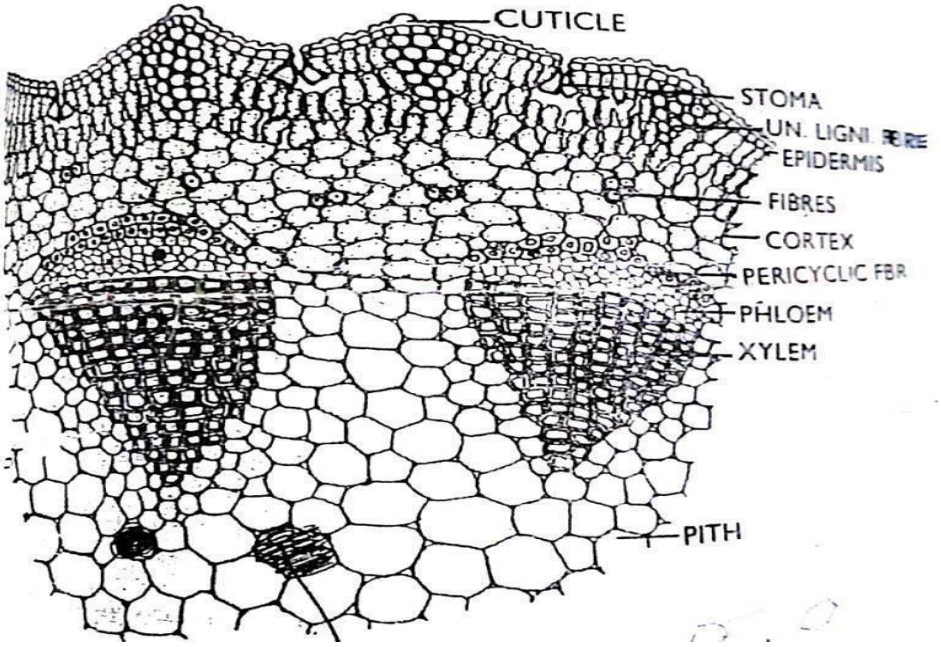
- It is a small woody shrub of 1m height.
- Shape: Cylindrical. Nodes and internodes are clearly seen.
- Colour: grey to greenish
- Odour: aromatic (fresh), no odour (dried)
- Taste: bitter and astringent
- Size: 5 mm in diameter
- Surface: Striations on surface and scaly leaves at the nodes.
- Fracture: Fibrous in cortical region whereas pith shows powdery mass

Active Constituents:

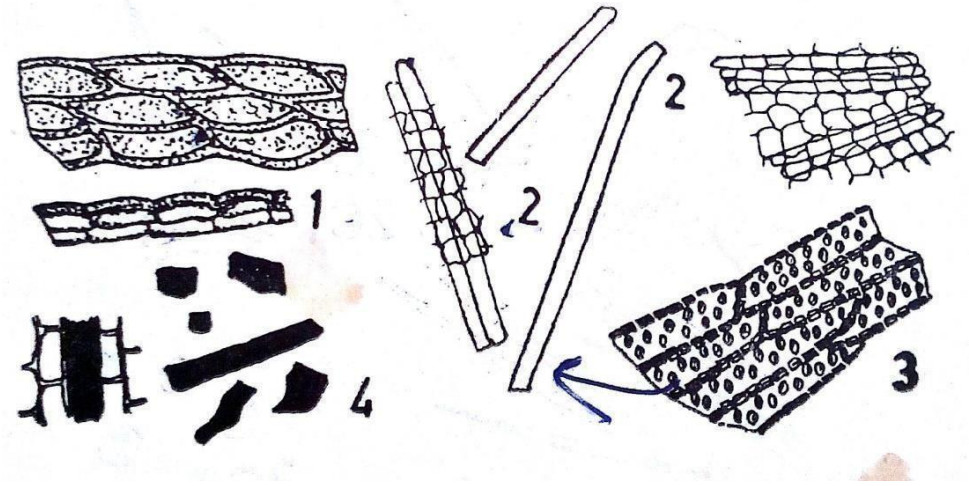
- Amino Alkaloids (0.5-3%)
    - **Ephedrine**(30-90%),
    - nor-ephedrine
    - n-methyl ephedrine
    - Pseudoephedrine
    - Ephedradines are present in roots

Uses:

- Sympathomimetic effects
- Antiasthmatic
- Bronchodilator
- Treatment of allergic conditions like hay fever



TRANSVERSE SECTION OF EPHEDRA



POWDER MICROSCOPY OF EPHEDRA

TRANSVERSE SECTION:

Transverse section of stem is more or less circular in outline. Following are the important tissues from periphery to centre.

- **Epidermis:** It is a single row of rectangular cells with a very thick and smooth cuticle. Sunken stomata are seen which are restricted to the furrows.
- **Cortex:** The outermost 2-3 layers of cortical parenchyma appearing like loosely arranged palisade cells contain chloroplasts. Groups of un-lignified fibres appearing like a bunch of grapes occur below the ridges where no palisade like cells are seen. Scattered lignified fibres either isolated or in groups of 2-4 occur in the inner layers of oval cortical parenchyma.
- **Vascular bundles:** Around 10 vascular bundles are found which is collateral, conjoint, open arranged in the form of a ring. Groups of lignified pericyclic fibres crown the phloem on the outer side. Cambium is indistinguishable. Xylem consists of vessels, tracheids, fibrotarcheids and parenchyma. IN mature stem, xylem is seen as a well-developed continuous band.
- **Pith:** It is large and made up of this walled, lignified, big polygonal parenchyma with inter cellular spaces. Some cells of the pith contain brownish matter.

POWDER MICROSCOPY:

1. Epidermis: Fragments of epidermal cells whose outer walls are ridged.
2. Fibres: Both lignified and unlignified fibres of uniform thickness, long, slender and cylindrical (like glass rod) appear either entire or in fragments.
3. Wood elements: Consisting of only tracheids with branched pits.
4. Brownish matter**:** Abundant and possess regular shape and form. They originate from pith.

Experiment-6

MORPHOLOGY AND MICROSCOPY OF FENNEL

AIM: To study the morphology and microscopy of Fennel.

REFERENCE: C.K. Kokate, Pharmacognosy, 45th ed., Pg. No: 1.37. M.A.Iyengar, Anatomy of crude drugs, Pg. No. 62. M.A.Iyengar, Powder microscopy of crude drugs, Pg. No. 42.

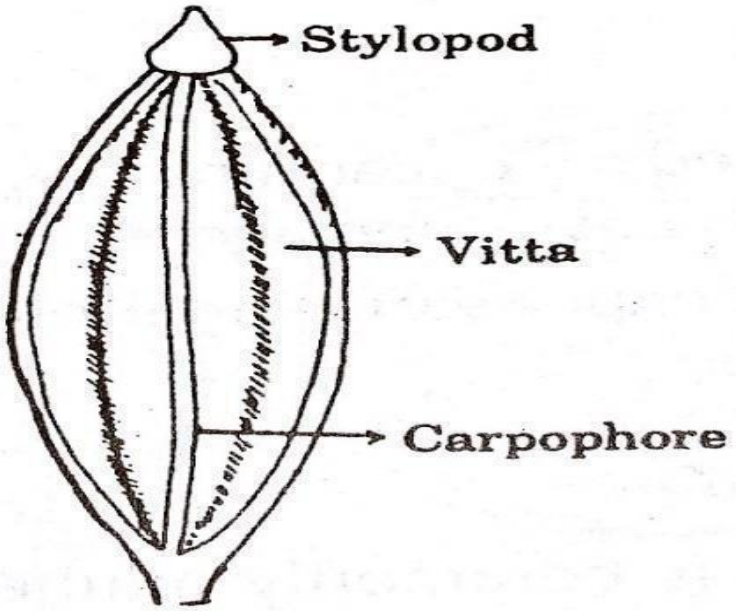
SYNONYM: Saunf, fennel fruits

BIOLOGICAL SOURCE:

It consists of dried ripe fruits of ***Foeniculum vulgare***. It should contain not less than 1.4 % v/w of volatile oil.

Family: Umbelliferae

MORPHOLOGY:



MORPHOLOGY OF FENNEL:

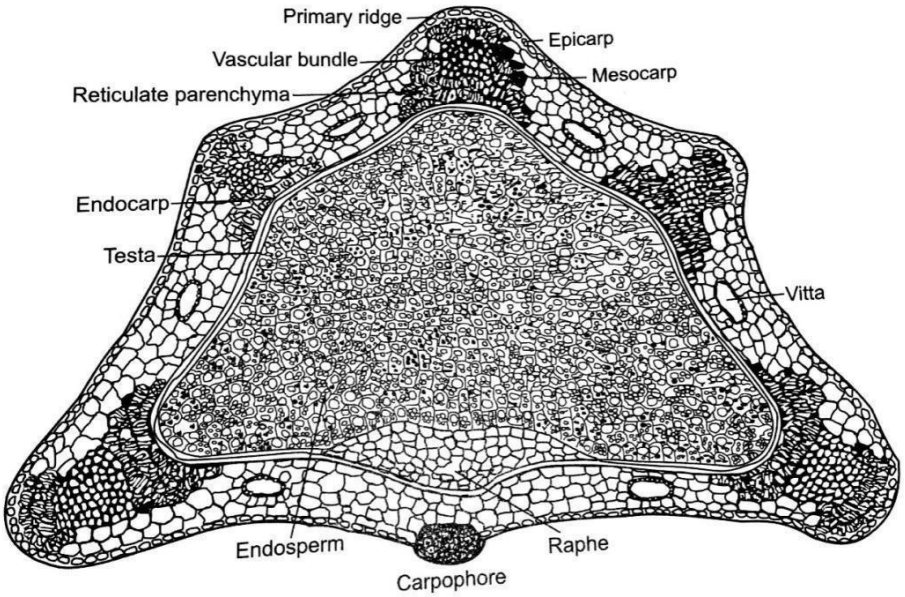
- Type: Cremocarp
- Shape: straight or slightly curved, oblong, laterally compressed tapering towards the base and apex, with a short stylopod at the apex and a thin pedicel at the base.
- Size: 5-10 mm (l), 2-4 mm (b)
- Surface: Each mericarp has two surfaces- the dorsal and the commissural surface. Dorsal surface is glabrous with 5 straight prominent primary ridges. Commisural surface is flat and shows the carpophore which holds the mericarps together.
- Colour: Greenish or yellowish brown
- Odour: Strongly aromatic
- Taste: Sweet and aromatic

Active Constituents:

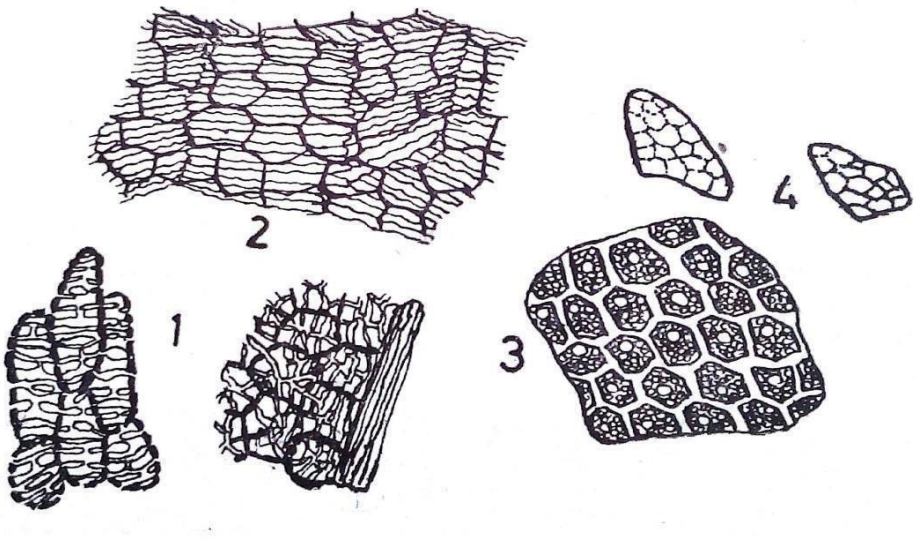
- Volatile oil (3-7%)- Anethole (50%), a phenolic ether, Fenchone (20%), a ketone
- Phellandrene, Methyl chavicol, anisic aldehyde, limonene, terpineol etc.
- Fixed oil
- Proteins (20%)

Uses:

- Carminative
- Aromatic
- Stimulant
- Flavouring agent



TRANSVERSE SECTION OF FENNEL



POWDER MICROSCOPY OF FENNEL

TRANSVERSE SECTION:

Transverse section of mericarp shows two prominent surfaces- the commissural surface and the dorsal. The commissural surface is flat with two pronounced ridges and the carpophore in the middle.

- **Pericarp:**
    - **Epicarp :** It consists of layer of polygonal, tangentially elongated cells with smooth cuticle.
    - **Mesocarp:** The bulk of mesocarp is made upof parenchyma. Bicollateral vascular bundles appear in the mesocarp below the primary ridges. Reticulate and lignified parenchyma, a characteristric feature of fennel appears in the mesocarp surrounding the vascular bundles. Besides, yellowish brown and elliptical vittae (schizogenous oil ducts), 4 on the dorsal surface between the ridges and two on the commissural surface are seen.
    - **Endocarp:** It is seen as a single layer between testa and mesocarp in the form of parquetry layer.
- **Testa:** It is single layered and yellowish in colour
- **Endosperm:** It is thick walled, polygonal colourless parenchyma containing oil globules and aleurone grains. A crescent shaped embryo is seen in the sections through apical region of mesocarp. Raphae, a ridge of vascular strand appears in the middle of commissural surface just in front of the carpophore.

POWDER MICROSCOPY

1. **Mesocarp:** Lignified and reticulate nature of parenchyma
2. **Endocarp:** Cells showing parquetry arrangement.
3. **Endosperm:** Polyhedral, thick walled cells containing aleurone grains, minute calcium oxalate crystals and oil globules.
4. **Vittae:** Many in the form of yellowish brown fragments.
5. **Odour:** Characteristic sweet aromatic.

Experiment-7

MORPHOLOGY AND MICROSCOPY OF CORIANDER

AIM: To study the morphology and microscopy of Coriander

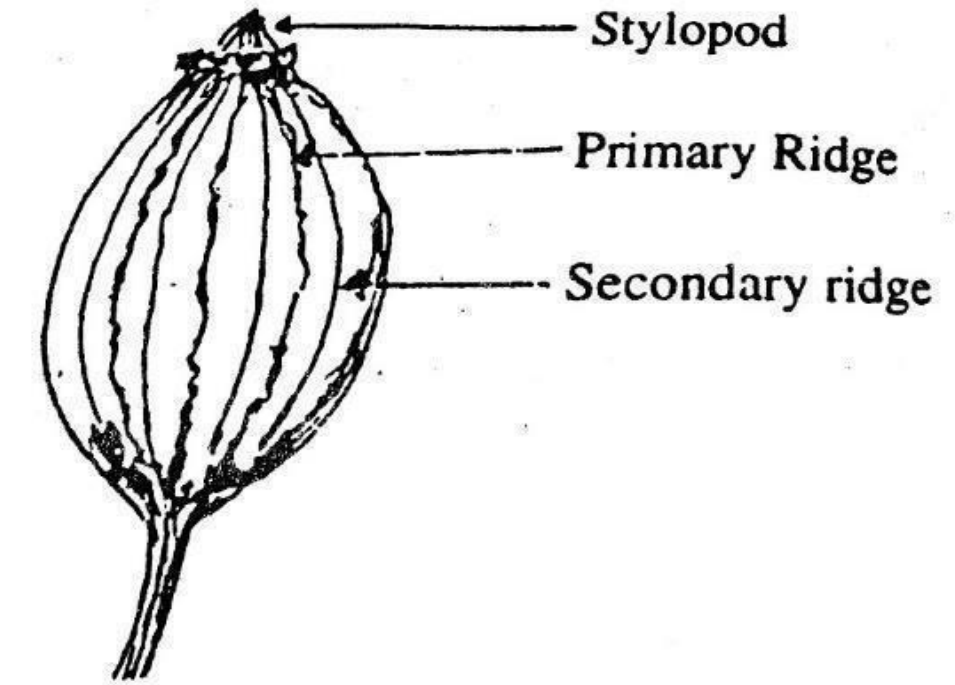
REFERENCE: C.K. Kokate, Pharmacognosy, 45th ed., Pg. No: 1.33.
M.A.Iyengar, Anatomy of crude drugs, Pg. No. 64.
M.A.Iyengar, Powder microscopy of crude drugs, Pg. No. 40.

SYNONYM: Coriander fruits, dhaniya.

SOURCE: It consists of dried ripe fruits of ***Coriandrum sativum***. It contains not less than 0.3% v/w of volatile oil.

Family: Umbelliferae

MORPHOLOGY



MORPHOLOGY OF CORIANDER FRUIT

- Shape: Sub globular cremocarpous fruit, with a pedicel in the base

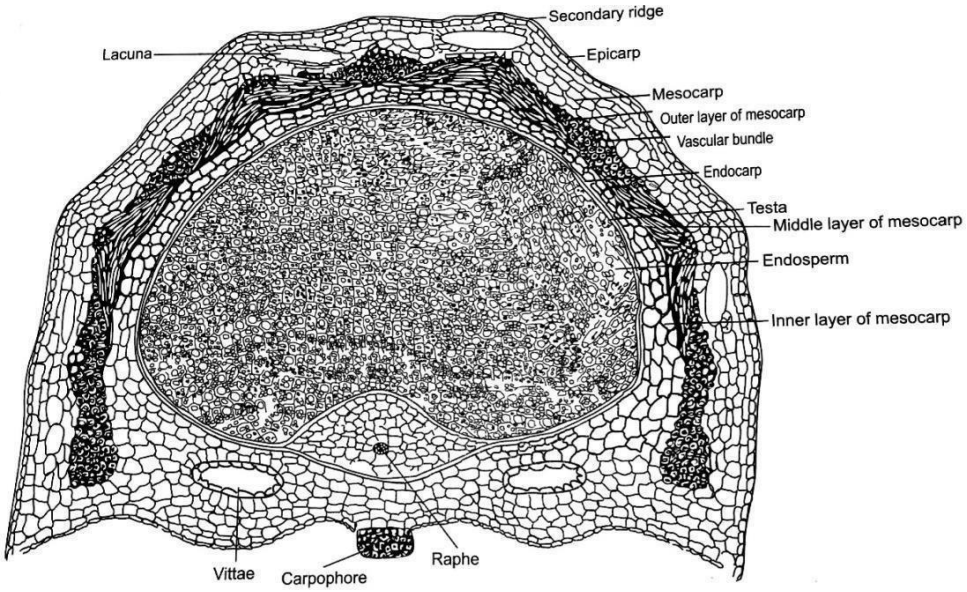
  [A **cremocarp** is made of two hemi spherical mericarps held together by a central stalk called as carpophore. It is obtained from a bicarpellary pistil. Each mericarp has two surfaces, a flat surface, commissural surface and a round surface called as dorsal surface].
- Size: 2-4 mm (d), 4-30 mm (l)
- Colour: Yellowish brown to brown
- Odour: Aromatic
- Taste: Spicy and Characteristic.
- Dorsal surface of each mericarp shows 5 primary ridges and 4 secondary ridges. Primary ridges are wavy, inconspicuous. Secondary ridges are straight and conspicuous. Primary and secondary ridges are alternate

Active Constituents:

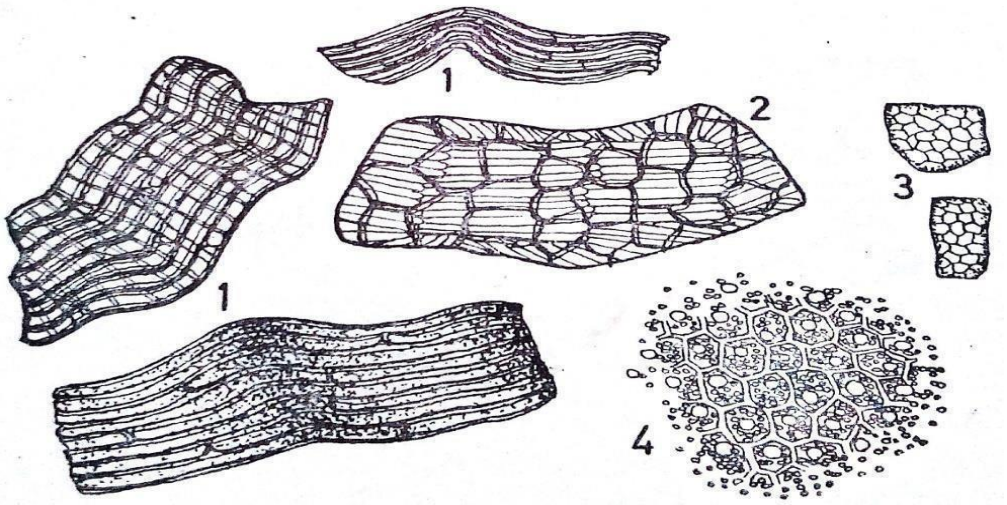
- Volatile oil (0.3-1%) – Coriandrol / D-Linalool (90%), Coriandryl acetate
- Minor - geraniol, borneol, pinene
- Fixed oil (13%)
- Proteins (20%)

Uses:

- Carminative,
- Flavouring agent,
- Stimulant,
- Aromatic



TRANSVERSE SECTION OF CORIANDER



POWDER MICROSCOPY OF CORIANDER

TRANSVERSE SECTION:

T.S of mericarp shows two prominent surfaces, the commissural and the dorsal.

- Pericarp
- Testa
- Endosperm (kidney shaped) are the important tissues of mericarp.

- **Pericarp:**
  - *Epicarp:* It consists of single row of small thick walled cells.
  - *Mesocarp:* three zones can be seen

    Outer loosely arranged tangentially elongated parenchyma
  - Middle compact sclerenchyma: outer region of this band is represented by longitudinal fibres (below primary ridges) and the inner region is made of tangentially running fibres (below secondary ridges). Vascular bundle crown the sclerenchymatous region of mesocarp below the primary ridges.

    Inner, irregular, polygonal and lignified parenchyma.
  - *Endocarp:* It is made up of typical parquetary layer.
- **Testa:** It is single layered and yellowish in colour.
- **Endosperm:** It is thick walled, polygonal, colourless parenchyma containing fixed oil and aleurone grains. An embryo is seen only if the T.S is taken through the apical region.

POWDER MICROSCOPY:

1. **Sclerenchymatous layer:** Groups of fusiform fibres of sclerenchyma running wavy and at times crossing with each other or with thin walled lignified cells of the mesocarp.
2. **Endocarp:** Fragments of parquetry arrangement of thin walled lignified cells with polygonal cells of mesocarp.
3. **Vittae:** Few brown fragments of vittae.
4. **Endosperm:** Fragments of endosperm with aleurone grains and oil globules.

ISOLATION OF PHYTOCONSTITUENTS

`

Experiment-8

ISOLATION OF CAFFEINE FROM TEA LEAVES

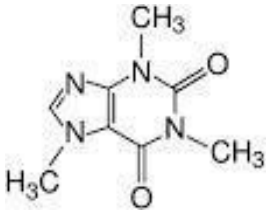
AIM: To isolate caffeine from tea leaves.

REFERENCE: C.K. Kokate, Pharmacognosy, 2015, 45th ed., Pg. No. 8.73.

MATERIALS & APPARATUS: Tea leaves**,** separating funnel, beakers, lead aceate solution, chloroform, dil. H_2SO_4, charcoal, ethanol.

PRINCIPLE:

CAFFEINE is chemically 1,3,7-Trimethyl xanthine, which is a purine base present along with other related bases like theophylline and theobromine in coffee, tea, and cocoa. Tea is obtained from the leaves of *Thea sinensis* (Theaceae). Coffee is obtained from the seeds of *Coffea arabica* (Rubiaceae). Although caffeine is largely produced synthetically it is also isolated from tea leaves or recovered from coffee seeds. Tea leaves contain 1 -4% of caffeine while coffee seeds contain 1-2% of caffeine.



Structure of Caffeine

PROPERTIES:

- Occurs as a white powder or white glistering needles matted together.
- It is odourless and has bitter taste.
- May sublime without decomposition at 178 °C.
- They may contain water molecules and hence hydrous in nature, which is soluble in chloroform and alcohols, sparingly soluble in water but increases at 80 °C.

IDENTIFICATION TEST:

Murexide test: To a few mg of caffeine sample in a porcelain dish, add few crystals of potassium chlorate and few ml of con. HCl. Heat to evaporate and expose the residue to ammonia vapours. The residue develops pink / purple colour.

PROCEDURE:

- Boil 20g of tea powder with 150ml of water for 30mins.
- Filter the mixture through muslin cloth.
- To the filtrate, add basic lead acetate (20-25ml).
- Filter and to the filtrate, add few ml of dil. H_2SO_4 (5ml) until it turns acidic to litmus.
- Filter and partition the filtrate with chloroform (20ml) thrice (20x3ml).
- Separate the chloroform layer, add activated charcoal and boil for few minutes.
- Filter and evaporate the chloroform layer on a water bath to dryness.
- Recrystallize with hot water or alcohol.

Uses: Caffeine is used as a Central Nervous System stimulant and mild diuretic.

REPORT: Caffeine was isolated from tea leaves and the percentage yield was found to be .....

Experiment-9

ISOLATION OF DIOSGENIN FROM DIOSCOREA

AIM: To extract Diosgenin from Dioscorea tubers.

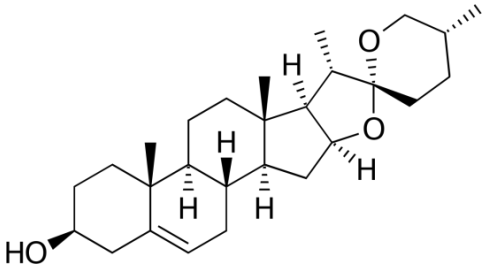
REFERENCE:

- C.K. Kokate, Text Book of Pharmacognosy, 51 ed, Nirali Prakashan, Pune. 2015: 9-48.
- Shah B, Seth AK. Isolation of phytopharmaceuticals, Text book of Pharmacognosy and Phytochemistry, 2nd ed. CBS publishers & Distributors. 2014: 451.

MATERIALS & APPARATUS: Beaker, petridish, funnel, vacuum pump, Con. H_2SO_4, acetone, dil. HNO_3.

Discussion:

Diosgenin is a steroidal saponin glycoside from the tubers of various species of Dioscorea such as Dioscorea floribunda, D.deltoidea, D.composita belonging to the family Dioscoreaceae.



Structure of Diosgenin

Uses:

- It is mainly used as a precursor for synthesis of several corticosteroids, sex hormones and oral contraceptives.
- Also used in treatment of rheumatic arthritis.

ISOLATION PRINCIPLE:-

Diosgenin can be isolated by two methods.

1. Alcoholic extraction method
2. Acid hydrolysis method.

PROCEDURE FOR ISOLATION:-

Alcohol extraction method:

- Alcohol tubers are chopped into chopped into small pieces and air dried.
- It is finely grounded and extracted twice with boiling ethanol or methanol.
- The filtered alcoholic extract is concentrated to dark syrup
- Syrup is hydrolysed by heating with 2N HCl for 2-12 hour

The crude diosgenin is precipitated and is recovered by filteration. It is purified by recrystallization from alcohol or chlorinated hydrocarbon.

Acid hydrolysis method:

- The properly dried tubers are grounded to a powder.
- The powdered drug is refluxed with 2N mineral acids (HCl or H2SO4) for a period of 2 -6 hours.
- The crude hydrolysate obtained on filteration is washed with water and lime till neutral.
- Then it is exhaustively extracted with hydrocarbon solvent such as toluene, n-hexane etc., for 6 hours.
- Crystals of diosgenin obtained.

Identification tests:

By TLC:

Sample is prepared by dissolving 1mg of diosgenin in 1ml methanol.

Adsorbent: Silica gel G

Solvent system: Toluene : Ethylacetate (7:3)

Detection: Spray with Anisaldehyde-sulphuric acid reagent and heat at 110^{o}C for 10min.

Rf value: 0.37 (A dark green spot).

REPORT: Diosgenin was isolated from Disocorea and the percentage yield was found to be

Experiment-10

ISOLATION OF SENNOSIDE FROM SENNA

AIM: To extract Sennosides from leaflets of Senna.

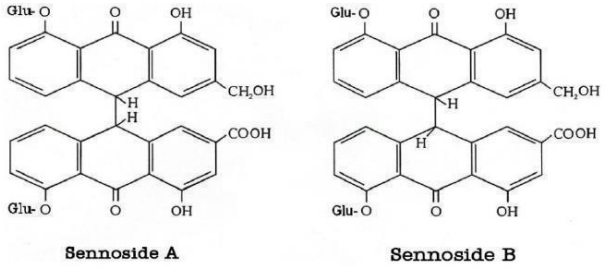
REFERENCE:

- C. K. Kokate, Text Book of Pharmacognosy, 51 ed, Nirali Prakashan, Pune. 2015: 9.24.
- Shah B, Seth AK. Isolation of phytopharmaceuticals, Text book of Pharmacognosy and Phytochemistry, 2nd ed. CBS publishers & Distributors. 2014: 460.

MATERIALS & APPARATUS: Senna leaflet powder, benzene, methanol, dil.HCl, calcium chloride, methanolic ammonia, gluconic acid and hydrobromic acid.

DISCUSSION:-

Sennosides are the dimeric anthraquinone glycosides obtained from the leaves and pods of Cassia angustifolia and C.acutifolia, belonging to family Leguminosae.



Structure of Sennosides

Uses:

- Purgative for habitual constipation problems.

IDENTIFICATION TEST:-

- Borntragger's test:Drug is boiled with dil. Sulphuric acid. Filtered. Filterate is partitioned with benzene or ether. Organic layer is separated and treated with ammonia. Ammoniacal layer shows pink or red colour due to anthraquinone glycosides.

PROCEDURE FOR ISOLATION:-

Method I:

- Powdered leaves are extracted with benzene on an electric shaker for 2 hours.
- Solvent is discarded and dried marc is extracted with 70% methanol for 4-6 hours.
- The marc is re-extracted again with 70% methanol.
- Extracts are combined and concentrated.
- Acidified with dil. HCl to pH 3.2 and set aside for 2 hours (to remove the resinous matter)
- Solution is filtered and to the filterate anhydrous CaCl2 in spirit is added and filtered.
- To the filterate, methanolic ammonia is added (pH-8) and set aside for 2 hours and precipitate is obtained.
- The precipitate is washed with methanol and dried.
- The precipitate containing Sennoside is dissolved in methanol and acidified with gluconic acid at 40^{o}C.
- The acidified extract after filteration yield precipitate containing yellow mass of sennoside A and the filterated treated with methanolic hydrobromic acid which yield Sennoside B.

Method II:

- 100g of powdered drug is mixed with 3 different solvents such as
  a. 70% acidic methanol
  b. 5%w/w tween-80 in 70% methanol
  c. 0.5%w/w PEG in 70% ethanol.

  and allowed to stand for 2 hours.
- The solutions are filtered and the filterate is treated with CaCl2 and washed with ethanol and crystallized.
- The percentage yield of Calcium Sennoside was determined as
  a. % yield was 51.2%
  b. % yield was 73.94%
  c. % yield was 63.88%

REPORT: Sennosides was isolated from Senna leaflet powder and the percentage yield was found to be_____.

Experiment-11

ISOLATION OF ATROPINE FROM BELLADONNA

AIM: To isolate Atropine from Belladonna.

REFERENCE:

- Shah B, Seth AK. Isolation of phytopharmaceuticals, Text book of Pharmacognosy and Phytochemistry, 2nd edition. CBS Publishers & Distributors Pvt. Ltd. 2014: 447.

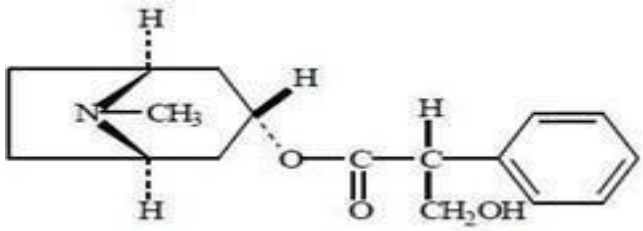
MATERIALS & APPARATUS: Belladonna powder, **ether**, benzene,sodium carbonate, acetic acid, sodium sulphate, alcohol, sodium hydroxide, nitric acid and potassium hydroxide.

DESCRIPTION:

It occurs as white crystals, usually needle-like, or as a white, crystalline powder. It is highly soluble in water with a molecular weight of 289.38. Atropine, a naturally occurring belladonna alkaloid, is a racemic mixture of equal parts of d-and l-hyoscyamine; its activity is due almost entirely to the levo isomer of the drug.

DISCUSSION:

Atropine is the major constituent obtained from the herb belladonna which is the dried herb of Atropa belladonna Linn. belonging to the family Solanaceae. The chemical constituents of the drug include an alkaloid, l-hyoscyamine. Atropine is the racemate form of l and d form of hyoscyamine. Chemically, atropine is designated as 1 H,5 H-Tropan-3 –ol (±) -tropate. Its empirical formula is $C_{17}H_{23}NO_3$ and its structural formula is:



Structure of Atropine

PROCEDURE:

Isolation of Atropine:

From the juice of the powdered form of drug, atropine is isolated. By using the aqueous solution of sodium carbonate, the powdered drug material is thoroughly moistened and extracted by using ether and benzene. From the solvent, the free base of alkaloid is extracted with acetic acid. To remove the colouring matter the acid solution is shakenwith ether and precipitation of alkaloid is carried out by using sodium carbonate. Then the solution is filtered, washed and dried. Again the dried sample is dissolved in acetoneor ether and dehydrated by using anhydrous sodium sulphate before filteration. A crude crystal of atropine is obtained after the concentration of filterate following cooling of the solution. For the complete racemisation of hyoscyamine to atropine, we have to dissolvethe crude crystalline sample in alcohol and sodium hydroxide solution.

IDENTIFICATION TEST:

Vitali Morin reaction:

The sample solution is diluted and treated with concentrated nitric acid. Evaporation of the mixture takes place for the drying of sample on the water bath and it produces a pale yellow colour residue. A drop of freshly prepared KOH is added to the yellow colour residue and the colour changes to violet

Uses: Atropine, an anticholinergic agent (muscarinic antagonist) and mydriatic activity.

REPORT: Atropine was isolated from Belladonna and the percentage yield was found to be_____.

CHROMATOGRAPHY

CHROMATOGRAPHY

Definition:

Chromatography is a method for separating the constituents of a solution (gas or liquid) by exploiting the different bonding properties of different molecules. Used in qualitative and quantitative analysis of biological and chemical substances, this technique employs two immiscible substances. One substance (a gas or liquid, called the mobile phase) transports the solution being analyzed through the other substance (a liquid or solid, called the stationary phase). The stationary phase absorbs or impedes different components of the solution to different degrees and, thus, causes their separation as different layers. Invented in 1906 by the Russian botanist Mikhail Tsvet (1872-1919).

Chromatographic techniques can be classified into 5 types based on the equilibration process. These are

- Adsorption
- Partition
- Ion exchange
- Permeation
- Affinity chromatography

Adsorption Chromatography:

The stationary phase is a solid on which the sample components are adsorbed. The mobile phase may be a liquid or a gas. The components distribute between the two phases through a combination of sorption and desorption processes.

- Gas-Solid Chromatography
- Liquid Column Chromatography
- High Performance Liquid Chromatography
- Thin Layer Chromatography

Partition Chromatography:-

The stationary phase is a liquid supported on an inert solid. Again the mobile phase is

liquid or a gas. Paper chromatography is a type of partition chromatography in which the stationary phase is a layer of water adsorbed on a sheet of paper. In the normal mode of operation of liquid-liquid partition a polar stationary phase is used with a non polar mobile phase. This favours retention of polar compounds and elution of non polar compounds and is called **Normal phase** chromatography. If a non polar stationary phase is used along with a polar mobile phase then non polar solutes are retained favouring elution of polar solutes. This is called **Reverse phase** chromatography.

- Gas-Liquid Chromatography
- Supercritical Fluid Chromatography
- Liquid-Liquid Chromatography
- Paper Chromatography
- High Performance Liquid Chromatography

Ion Exchange Chromatography:

Ion Exchange Chromatography, High Performance Liquid Chromatography (HPLC)

Permeation Chromatography: Size Exclusion Chromatography

Affinity Chromatography: DNA Affinity Chromatography

Electrophoresis Chromatography: Capillary Electro Chromatography

Paper Chromatography:

A form of chromatography in which a sheet of special paper is substituted for the adsorption column. After separation of the components as a consequence of their differential migratory velocities, they are stained to make the chromatogram visible. In the clinical laboratory paper chromatography is employed to detect and identify sugars and amino acids.

Thin-layer chromatography:

In this, the stationary phase is a thin layer of an adsorbent such as silica gel coated on a flat plate. It is otherwise similar to paper chromatography.

Experiment-12

THIN LAYER CHROMATOGRAPHY OF HERBAL EXTRACTS

AIM: To separate and identify the mixture of alkaloids by Thin Layer Chromatography.

REFERENCE: **Biren Shah & Seth AK.** Text Book of Pharmacognosy & Phytochemistry.2nd ed. 2010: 447-460.

MATERIALS & APPARATUS: Toluene, ethyl acetate, acetic acid, strong ammonia solution, toluene and acetone & methanol, TLC plates, development chamber, Spraying reagent - Dragendorff's reagent.

DISCUSSION:

The term chromatography refers to a number of highly efficient techniques used for the separation and purification of wide variety of components ranging from inorganic to biopolymers. TLC is mainly used qualitatively for screening of different plant extract, which serves as an important tool in overall phytochemical research. It is a method in which the mobile phase moves by capillary action across uniform thin layer of finely divided stationary phase, which is bonded onto a plate. When a mixture of drugs are applied on the plate and developed by a mobile phase, the drugs move across the plate at different rates depending on solubility, Pka values, capability of hydrogen bonding and gets separated. The separated spots are visualized using different reagents and identified by calculating the R_f value.

Stationary Phase:

Silica gel-G is used for the experiment. Other adsorbents that can be used are silica gel-G, alumina, cellulose, keiselguhr, polyamide, activated charcoal, starch and insulin etc. Activation of adsorbent is done in order to remove the impurities and moisture from the plates by heating the plates at 100-115 °C for 1hr.

Mobile Phase:

The choice of the mobile phase depends mainly on the material to be separated and the stationary phase to be used. Selection of the solvent system is done based on cost, availability, and stability. It should not require with the substance to be separated. The solvent systems for alkaloids are:

n- Butanol: acetic acid: water (4: 13)

- Toluene: ethyl acetate: diethyl amine (7:2:1)
- Chloroform: dimethylamine (9:1)
- Ethyl acetate: methanol: water (10: 13, 5:10)

Sample application:

For quantitative work, micropipettes are used for qualitative work, capillary tubes are used.

Saturation of chamber with the mobile phase:

Glass chamber with a lid is used. After sample application the plate is placed in the development chamber at an angle of 45 °C. The bottom of plate 19 dipped 2mm in the mobilephase. Three side of a tank are lined with solvent impregnated paper to get a uniform saturation of the chamber with a mobile phase. Throughout the development, tank top if closed with a lid.

Detection of components:

By using visualizing reagents.

Evaluationof chromatogram:

The number of spots and their colour are noted and R_f values are calculated.

PROCEDURE:

Caffeine:

- Dissolve 1mg of caffeine in 1 ml of chloroform or methanol.
- Sample is spotted on the TLC plate.
- Eluted with Ethyl acetate : Methanol:acetic acid (80:10:10).
- Visualized the dried TLC plate by exposure to iodine vapour.
- Caffeine develops a spot of Rf value 0.41.

Sennosides

- Sample spotted in silica gel-G plates.
- Mobile phase: Ethylacetate:methanol:water (100:16.5:13.5).
- Visualized by spraying first with 25% nitric acid and then with alcoholic potassium hydroxide.
- Red coloured spots appear with nitric acid and turns to yellow when sprayed after drying with alcoholic potassium hydroxide solution.

Diosgenin:

- The sample is dissolved in methanol and spotted in silica gel plates
- Mobile phase: Toluene: Ethylacetate (7:3).
- Visualizing reagent: Anisaldehyde –sulphuric acid reagent.
- Dark green spot of diosgenin appear with the Rf value of 0.37.

Atropine:

- 1% solution of atropine dissolved in 2N acetic acid and spotted over silica gel-G plate.
- Mobile Phase: Strong Ammonia solution : methanol (1.5:100) and Acetone : 0.5M Sodium chloride.
- Visualizing agent: Acidified iodoplatinate solution and Dragondorff's reagent..
- Atropine is visible as a spot with an Rf value of 0.18 with MP-1 and 0.70 with MP-2.

REPORT: The given sample No._____was found to be_____ with R_f value_____.

Experiment-13

SEPARATION AND IDENTIFICATION OF AMINO ACIDS BY PAPER CHROMATOGRAPHY

AIM: To separate and identify the mixture of amino acids by Paper Chromatography.

REFERENCE: N.Rajesh Kumar, S.N.Meyyanathan, A practical approach to Pharmaceutical Analysis, Pg. No. 146.

MATERIALS & APPARATUS: Standard aminoacids, sample mixture, n-butanol, acetic acid, ninhydrin reagent, glycerin, aspartic acid, beakers, chromatographic chamber, whatmann filter paper, spraying gun.

PRINCIPLE:

The paper chromatography (PC) technique is a type of partition chromatography in which the substrates are distributed between 2 liquids i.e. one is the stationery phase liquid which is held in the fibres of the paper and is called as the Stationary phase and the other is a moving liquid (or) developing solvent called the Mobile phase. The components of the mixture migrate at different rates and appear as spots at different positions on the paper.

PROCEDURE:-

- Whatmann filter paper was taken and cut out to get the proper dimensions inorder to fit into the developing chamber.
- Then the mobile phase was prepared by mixing n-butanol: acetic acid: water in the proportion (4:1:5)
- This mobile phase was then transferred to the developing chamber upto a height of 1- 2cm from the bottom and the lid was closed to saturate the chamber.
- Then the amino acid samples were prepared by dissolving in appropriate solvent.
- Then using capillary tubes amino acids were spotted on the paper maintaining a distance of about 1.5 inch from one end of the paper and leaving a gap a of 2 - 3cm between 2 sample spots.

.

- The spots on the paper were allowed to dry and then the paper was placed in a saturated chamber in such a way that the spots do not dip into the mobile phase.
- Paper was then dried and the spots were visualized by spraying ninhydrin reagent. The migration parameter was calculated by using R_f value.

REPORT: The given sample No._____was found to be a mixture of___&_____with their R_f values____ _____&_____respectively

.

Experiment-14

DISTILLATION OF VOLATILE OILS AND DETECTION OF PHYTOCONSTITUENTS BY TLC

AIM: To distillate volatile oils and identify by TLC Chromatography.

REFERENCE: N.Rajesh Kumar, S.N.Meyyanathan, A practical approach to Pharmaceutical Analysis, Pg. No. 146.

MATERIALS & APPARATUS: Standard aminoacids, sample mixture, n-butanol, acetic acid, ninhydrin reagent, glycerin, aspartic acid, beakers, chromatographic chamber, whatmann filter paper, spraying gun.

PRINCIPLE:

The paper chromatography (PC) technique is a type of partition chromatography in which the substrates are distributed between 2 liquids i.e. one is the stationery phase liquid which is held in the fibres of the paper and is called as the Stationary phase and the other is a moving liquid (or) developing solvent called the Mobile phase. The components of the mixture migrate at different rates and appear as spots at different positions on the paper.

PROCEDURE:-

- Whatmann filter paper was taken and cut out to get the proper dimensions inorder to fit into the developing chamber.
- Then the mobile phase was prepared by mixing n-butanol: acetic acid: water in the proportion (4:1:5)
- This mobile phase was then transferred to the developing chamber upto a height of 1- 2cm from the bottom and the lid was closed to saturate the chamber.
- Then the amino acid samples were prepared by dissolving in appropriate solvent.
- Then using capillary tubes amino acids were spotted on the paper maintaining a distance of about 1.5 inch from one end of the paper and leaving a gap a of 2 - 3cm between 2 sample spots.
- The spots on the paper were allowed to dry and then the paper was placed in a saturated chamber in such a way that the spots do not dip into the mobile phase.
- Paper was then dried and the spots were visualized by spraying ninhydrin reagent. The migration parameter was calculated by using R_f value.

CHEMICAL TESTS

Experiment-15

CHEMICAL TEST FOR ASAFOETIDA

Aim: To carry out the chemical tests for Asafoetida.

Biological source: It is obtained by incision from the rhizomes and roots of *"Ferula foetida"* and *"Ferula rubricaulis".*

Family: Umbelliferae

Description:

Colour: yellowish white

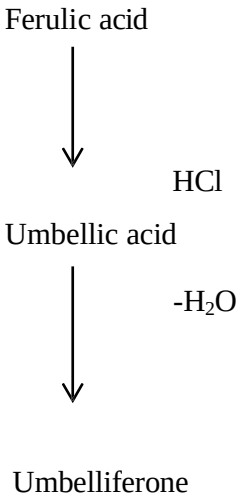
Odour: intense and characteristic

Taste: bitter, acrid

State: solid

Chemical constituents:

It contains resins (40 to 65 %), gum (20 to 25 %), and volatile oil (4 to 20 %). It mainly contains asaresinotannols in free or combined form with ferulic acid.



Oil of asafoetida obtained by steam distillation of oleo gum resin. The oil contains secondary butyl propanyl disulphide. Other constituents of oil are di and tri sulphides, pinene, and other terpenes.

Uses: carminative, nervine stimulant, flavouring agent.

Chemical tests:

Sl. no	Experiment	Observation	Inference
01.	**Solubility test:** a) To small qty of sample + water b) Sample + alcohol	Insoluble Insoluble	May be asafoetida
02.	Triturate sample with water	Orange colour	May be asafoetida
03.	A piece of freshly cut surface of asafoetida in china dish. To this add 1-2 drops of conc. H_2SO_4 and wash.	Colour changes to violet	May be asafoetida
04.	Take a piece of freshly cut asafoetida in a watch glass. Add few drops of conc. HNO_3	Green colour seen	Maybe asafoetida
05.	**Umbelliferone test:** 0.5g of drug + 5ml of 90 % alcohol for 2 mins and cool. Filter. Add 0.5 ml of 10 % NH_3.	No blue fluorescence seen	Asafetida present
06.	**Combined umbelliferone test:** 0.5g of drug + 3ml of HCl + 3ml of water. Boil for 5-10 mins. Filter and to the filterate add equal amt of alcohol and strong solution of NH_3 in excess.	Blue fluorescence obtained	Asafoetida confirmed

Report: The given drug was identified to be Asafoetida.

CHEMICAL TEST FOR BENZOIN

Aim: To carry out the chemical tests for Benzoin.

Biological source: It is a balsamic resin obtained from *Styrax benzoin, Styrax paralleloneurus*

Family: Styraceae.

Description:

Colour: Greyish- brown

Odour: Aromatic and characteristic

Taste: Sweetish and slightly acrid

State: solid

Chemical constituents: Free balsamic acids (benzoic and cinnamic acids). Coniferyl benzoate (76%), styrol, vanillin, phenyl propyl cinnamate.

Sl.No	Chemical test	Observation	Inference
1	Heat 0.5 gm of powder slowly in a dry test tube.	Melts and white fumes are evolved which condense on the walls of the test tube to form crystalline sublimate.	Benzoin present.
2	Warm 1 gm of powder with 5 ml of Potassium permanganate .	Distinct odour o almonds)	Benzoin present.
3	Triturate 0.1 gm of powder with 95% alcohol (5 ml) and filter. To the filterate, add 5 ml of 5% w/v Ferric chloride solution in alcohol.	Absence of green colour.	Sumatra benz oin is present.
4	Digest benzoin with 5 ml of ether for 5 minutes. Pour the ethereal solution in a china dish containing 2-3 drops of Conc. H_2SO_4 and rotate the dish.	Reddish bro wn colour is produced.	Benzoin confirmed.

Report: The given drug was identified to be Benzoin.

Experiment-16

CHEMICAL TEST FOR ALOES

Aim: To carry out the chemical tests for Aloes.

Biological source: It is the mucilage obtained from obtained from *Aloe barbadense*

Family: Liliaceae.

Discussion:

Aloe contains a mixture of crystalline glycosides known as aloin 4 -5% in cape Aloe 18-25% in Curacao Aloe, resin(16-37%),emodin and volatile oil. It also possess the anthraquinone glycoside like barbaloin(aloe-emodin anthrone C-10 glucoside),chrysophanic acid,B-barbaloin and iso-barbaloin.

Uses:

Aloe is Pharamaceutic aid for compound benzoin tincture and a cathartic.When used as a cathartic it acts chiefly on the large intestine. Aloe glycoside elicit a relatively advocate a preferential use of other cathartic substances. Aloe available with cascara sagrada in nature's Remedy.

Chemical Tests

Sl. No	Chemical test	Observation	Inference
1	Bromine Test: To 2ml of solution of aloes add 2ml of freshly prepared solution of bromine.	A pale yellow precipitate i f tetrabarbromaline is produced.	This test is not specific for aloes.
2	Nitric acid test to 5 ml of solution of aloes gives a brownish color rapidly changing to green	Barbados a deepbrownish- red. Socotrine a pale brownish- yellow. Zanzibar a yellow brown color.	Aloes present

Report: The given drug was identified to be Aloes.

CHEMICAL TEST FOR MYRRH

Aim: To carry out the chemical tests for Myrrh.

Biological source: It is an oleo-gum resin obtained from stem and branches of Commiphora mukul.

Family: Burseraceae.

Uses:

Antiseptic, stimulant, used in incense sticks and perfumes, astringent to mucous membrane and carminative.

Sl.No	Chemical test	Observation	Inference
1	When triturated with water	Yellow emulsion formed	Myrrh present
2.	Sample when triturated with washed sand in presence of ether, filtered and evaporated on a water bath. Thin film obtained is exposed to bromine vapours in a closed dessicator.	Violet colour obtained.	Myrrh present and confirmed.

Report: The given drug was identified to be Myrrh.

CHEMICAL TEST FOR COLOPHONY

Aim: To carry out the chemical tests for Colophony.

Biological source: Colophony is the solid residue obtained after distilling the oleo- resin from various species of *Pinus palustris*.

Family: Pinaceae.

Chemical constituents:

Resin acids- Resin acid or diterpene acids like abietic acid

Neutral inert substance

Resins. Esters of fatty acids

Uses:

1. It is used in preparation of like plasters and ointment.
2. It is also used in manufactures of varnishes and disinfecting liquids.

Chemical test:

Sl. No	Chemical test	Observation	Inference
1	0.1 gm of powdered drug sample (**colophony**) is dissolved in 10 ml of acetic acid and one drop of sulphuric acid (concentrated) is added in a dry **test** tube.	An alcoholic solution of **colophony** is acidic to litmus which turns the blue colour litmus to red colour.	Colophony present
2.	Powdered sample dissolved in 2-3ml of acetic anhydride in a test tube and a drop of con.sulphuric acid is added	Purple to violet colour obtained	Colophony present
3.	Colophony dissolved in light petroleum and filtered. To the filterate 2-3 times its volume, copper acetate solution is added.	Emerald green colour seen in petroleum layer.	Colophony present and confirmed.

Report: The given drug was identified to be Colophony.

Vision and Mission of the Institution

Vision

The East Point College of Pharmacy aspires to be a globally acclaimed institution, **recognized** for **excellence in** pharmaceutical education, research and nurturing students for **holistic development.**

Mission

M1 Create pharmacy graduates through **quality education**

M2 Promote innovation, **creativity**, and excellence **in teaching**, learning, and **research**

M3 **Inspire** integrity, teamwork, critical thinking**, personal** development, and ethics in **students** and lay **the** foundation for lifelong learning

M4 **Serve** the **healthcare, technological, scientific**, and **economic** needs of then **society**.