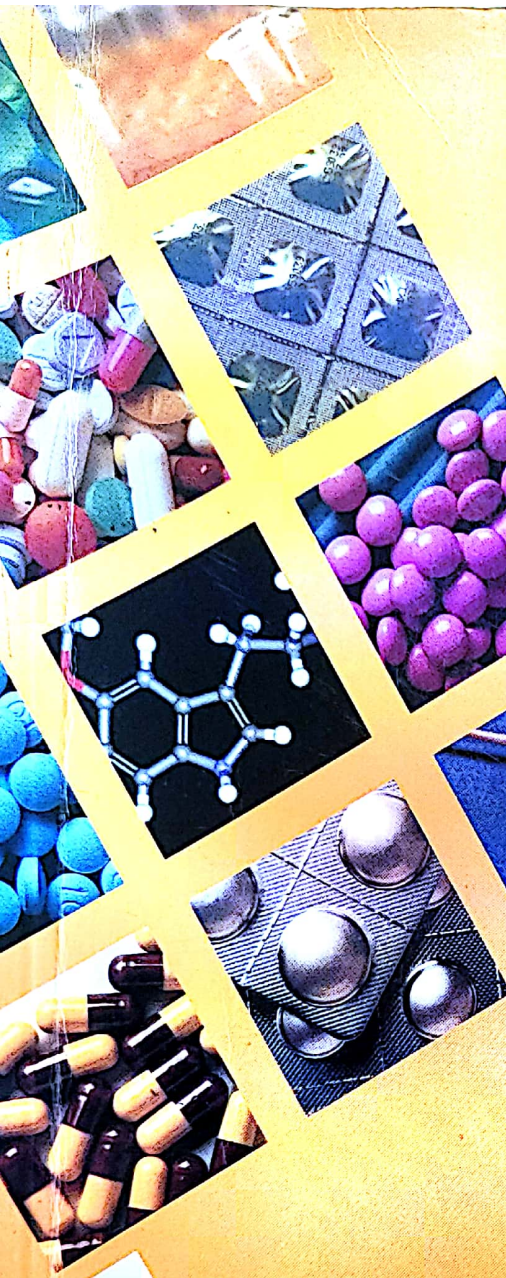

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Chopda - 425107 Dist - Jalgaon (M.S.)

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Subject : Pharmacognosy - V (P.3.6.4)

Name : Songire Harshali Ramesh Roll No. 02

Class : T.Y.B.Pharm (Sem VI th) Batch 'A'

Examination Seat No. : 507546 Pages : 250

Vision

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Mission

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Chopda - 425 107, Dist. Jalgaon (M.S.)

Exam. Seat No. 507546 *Date :* 04/04/2019

CERTIFICATE

This is Certify that

Shri /Kum. Songire Harshali Ramesh.

has satisfactorily completed the course of study as laid down by North Maharashtra University, Jalgaon for practicals in Pharmacognosy-IV *of* VI^th *Semester B. Pharm. during the Academic year 2018 -2019 & this laboratory record represents the work done by him / her within the four walls of this Institute.*

LECTURER - IN - CHARGE
(Dr. Md. Rageeb Md. Usman) sir.

HOD
(Dr. G. P. Vadnere)

PRINCIPAL
Mahatma Gandhi Shikshan Mandal's
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College of Pharmacy,
Chopda Dist. Jalgaon (M.S.)

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INDEX

Sr. No.	EXPT. No.	NAME OF THE EXPERIMENT	DATE	PAGE. No.	MARKS 10	SIGNATURE	REMARKS
		Introduction		1			
1)	1.	To perform isolation of chemical constituents and pharmaceutical significance of 'senna'	19/12/18	7	18		
2)	2.	To perform isolation of chemical constituents and pharmaceutical significance of 'ephedra'	26/12/18	11	18		
3)	3.	To perform isolation of chemical constituents and pharmaceutical significance of 'cinchona'	2/1/19	15	18		
4)	4.	To perform separation of chemical constituents of pharmaceutical significance (senna)	9/1/19	22	18		
5)	5.	To perform separation of chemical constituents of pharmaceutical significance (ephedra).	16/1/19	26	18		
6)	6.	To perform separation of chemical constituents of pharmaceutical significance (cinchona).	23/1/19	30	18		

INDEX

Sr. No.	EXPT. No.	NAME OF THE EXPERIMENT	DATE	PAGE. No.	MARKS 10	SIGNATURE	REM.
7)	7.	To perform purification of chemical constituent of pharmaceutical significance (senna)	30/1/19	37	18		
8)	8.	To perform purification of chemical constituent of pharmaceutical significance (Ephedra)	6/2/19	40	18		
9)	9.	To perform purification of chemical constituent of pharmaceutical significance (cinchona)	13/2/19	43	18		
10)	10.	To perform spectroscopic of chemical constituent of pharmaceutical significance (senna)	20/2/19	49	18		
11)	11.	To perform spectroscopic analysis of chemical constituent of pharmaceutical significance (ephedra)	27/2/19	52	18		

INDEX

Sr. No.	EXPT. No.	NAME OF THE EXPERIMENT	DATE	PAGE. No.	MARKS 10	SIGNATURE	REMARKS
12)	12.	To perform the spectroscopic analysis of chemical constituents of pharmaceutical significance (Cinchona)	6/3/19	57	18		
13)	13.	To perform estimation of total flavonoids	13/3/19	60	18		

Avg = 234 / 13

Avg = 18

Introduction

Extraction -

Extraction may be defined as the process in which the animal or plant tissue are treated with specific solvent thereby the medicinally active constituents are dissolved out. cell tissue & most of inactive or inert components remains undissolved.

Menstrum -

Solvent used for extraction purpose is known as menstrum.

Marc -

Residue left after extracting the desired constituents is known as marc.

List of solvent used for extraction

1) Water
2) Ether
3) Chloroform
4) Petroleum ether
5) Ethanol

Extraction Process -

1) Infusion -

The method is used for those drugs which are soft in nature so that water may penetrate

easily to tissue.

2) Decoction -

Decoction is a process in which the water soluble and heat stable constituents of hard and woody crude drugs are extracted out.

3) Digestion -

The process is modified form of maceration in which the extraction of drugs is carried out by applying gentle heat to the substance being extracted.

4) Maceration -

In this process the extraction of drugs is carried by placing the solid drugs. Fine powders are not used because there will be modified or moderately coarse powder water or alcohol are the used as menstrum and closed vessel is used to prevent evaporation.

5) Percolation -

Percolation also known as simple percolation is another method of extractive of active constituents from the drugs used in preparation of tincture of liquid extraction in this process the suitable communicable drug is moistened with sufficient quantity of menstrum.

6) Continuous Hot Percolation Process / Soxhlet Extraction

Continuous hot percolation process is used for those drugs where the penetration of menstrum into the cellular tissue is very slow & solute is not readily soluble.

Factors affecting selection of extraction process –

1) Nature of Drug :–

The selection of extraction process is mainly depend upon the physical process / nature of drug. Hard and woody drug are extracted by percolation process whereas soft drugs extracted by maceration.

2) Cost of Drug :–

Costly drug are extracted by percolation where as cheaper drugs may extracted by the maceration.

3) Stability of Drug :–

Continuous hot extraction should not be used for those drugs are the active constituents of which are thermolabile.

4) Therapeutic value of Drug :–

The drugs containing flavouring agent bitter etc which does not have therapeutic value may be extracted by maceration.

Procedure –

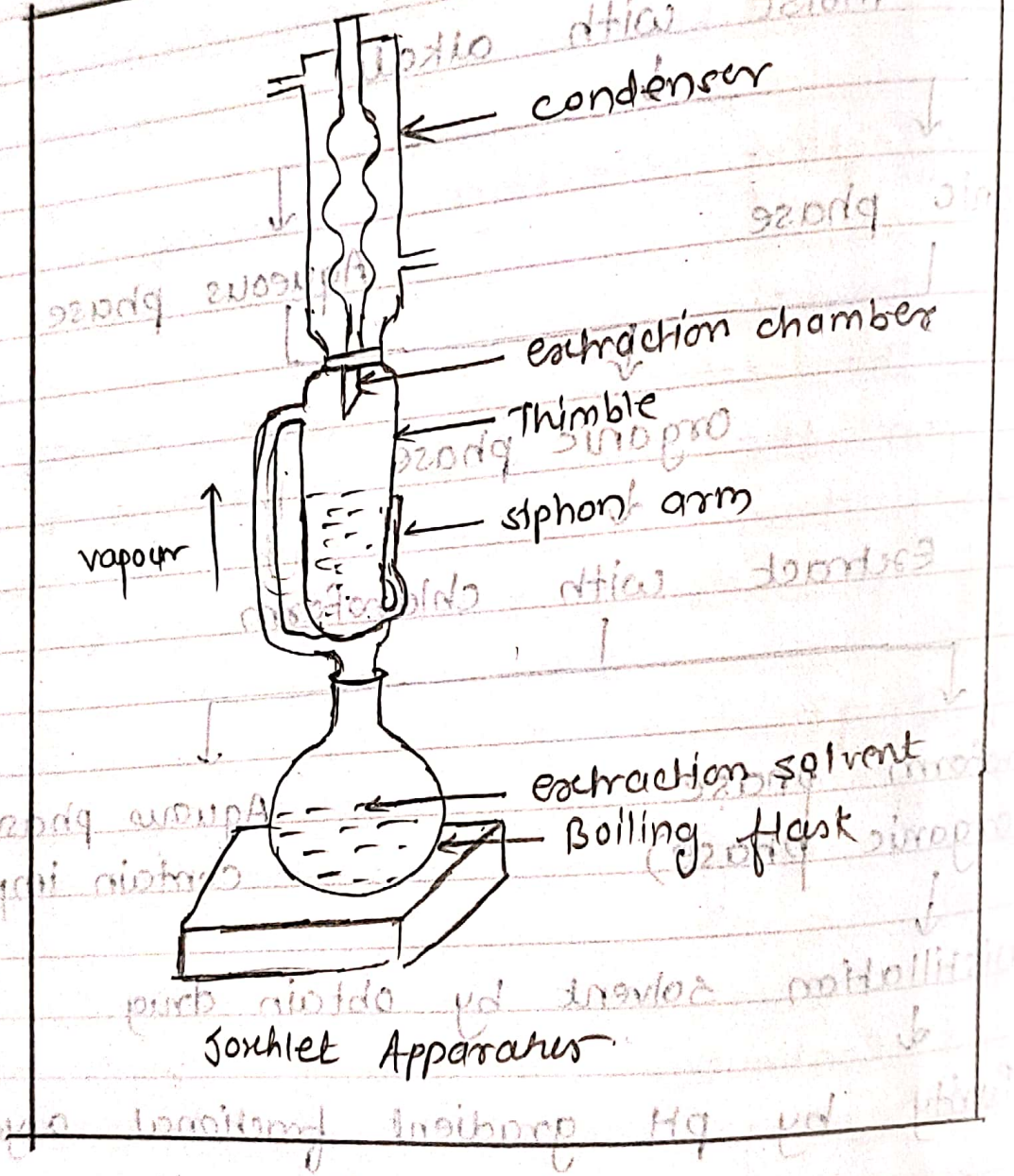
Powdered Drug
↓
moist with alkali
↓
Organic phase | Aqueous phase
↓
organic phase
↓
Extract with chloroform
↓
Chloroform phase (organic phase) | Aquous phase contain impurity
↓
Distillation solvent by obtain drug
↓
Purity by pH gradient fractional crystallization

Isolation –

Isolation is a process of separation of active chemical constituents from crude drug or extract.

Principle –

The extraction & isolation of an element from its combined form involves various principles of chemistry



Soxhlet Apparatus

A particular element may occur in a variety of compounds. The process of metallurgy & isolation should be such that it is chemically feasible & commercially viable.

Method use for isolation :-

Soxhlet Apparatus -

1) Soxhlet extraction is the process of continuous extraction in which the same solvent can be circulated through the extractor several times.
2) The process involves extraction followed by evaporation of the solvent.
3) The vapours of the solvent are taken to a condensor and the condensed liquid is returned to the drug for continuous extraction.
4) Soxhlet apparatus designed for such continuous extracti-on consist of the body of the extractor attached with a side tube & siphon tube.
5) The extractor from the lower side can be attached to distillation flask and the mouth of the extractor is fixed to a condensor by standard joints.
6) The crude drugs powder is packed in the soxhlet apparatus directly or in a thimble of filter paper or fine muslin.
7) The diameter of the thimble correponds to the internal diameter of the soxhlet extractor.

Advantages -

1) Soxhlet extraction is advantageous in a way that less solvent is needed for yielding more concentrated products.

2) The extraction can be continued until complete exhaustion of the drug.

Disadvantages—

The main disadvantage is that this process is restricted to pure boiling solvents or to azeotrops.

Senna

Synonym - senna leaf

B.S - leaflets of the plant cassia angustifolia

Family - Leguminoceae

macroscopy-

Colour - Yellowish green

odour - slight

Taste - Bitter

C.C - anthraquinone, sennaside A & B

user - Purgative

Substituent & Adulterants:-

Dog senna & pathe senna.

Experiment No - 01 Date - 19/12/18

Page No.: 7

im To perform the isolation of chemical constituents and pharmaceutical significance (Senna)

ference-

1) Kokate C.K, Purohit A.P, Gokhale S.B, Book of Pharmacognosy, Nirali Prakashan, 48th edition, Page no - 9.24-9.27
2) Dr. Khadbadi S.S, Dr. Deore S.L. Baviskar, Book of experimental pharmacognosy, Nirali Prakashan, 6.2-6.16
3) Rangari vindo. D. (2009) Nasik, Pharmacognosy & phytochemistry, Vol-II, 2nd edition, Career publication. 223-228.

quirements-

Apparatus - Soxhlet apparatus, reflux condenser, heating mantle, RBF, beaker, measuring cylinder.

Chemicals - Drug powder, alcohol, water etc.

Principle -

Soxhlet extraction process is used for isolation of active chemicals constituents of senna drug. Soxhlet process involves the material which is to extracted is placed in a thimble made up of cellulose in the centre of assembly of soxhlet apparatus. It involves 8:7 in/by from of soxhlet is originally designated extraction of the liquid.

* Calculation -

wt of empty dish = 63.27 gm

wt of crucible + dried sample = 69.75 gm

wt of sample = 6.48 gm

Total wt = 6.38 + 6.48

= 12.86 gm

Percentage yield -

$23 = 100\%$

$12.86 = x$

$$x = \frac{12.86 \times 100}{23}$$

$$\boxed{x = 55.91\%}$$

SENNA

Synonym - Senna leaf, Sennai-ki-patti etc.

Biological source -

It consist of dried leaflets of casia angustifolia or cassia senna vahl. (1)

Family - Leguminaceae. (1)

Geographical source -

Indian senna is calculated in India. It's cultivation is done in Tinnevelley, Madurai and Ramnathpuram district of Tamil Nadu. (1)

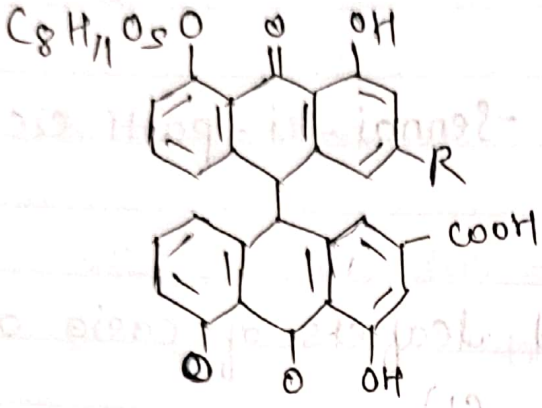
Macroscopic character -

Colour - Yellowish - Green
Odour - Slight
Taste - Mucillageneous, bitter & characteristics.
Size - 1-8 mm in width & 25-60 mm length.
Shape - Leaves are lanceolate, entire apex is acute with spine at the top. (2)

Chemical constituents -

1) Senna contain mainly two anthraquinone glycosides called as sennoside A & sennosides B
2) Tutin in 1913 isolated rhein and aloe-emodin
3) Sennoside A & B are stereoisomer of each other.
4) They are dimeric glycoside with rhein dianthrone

(2)

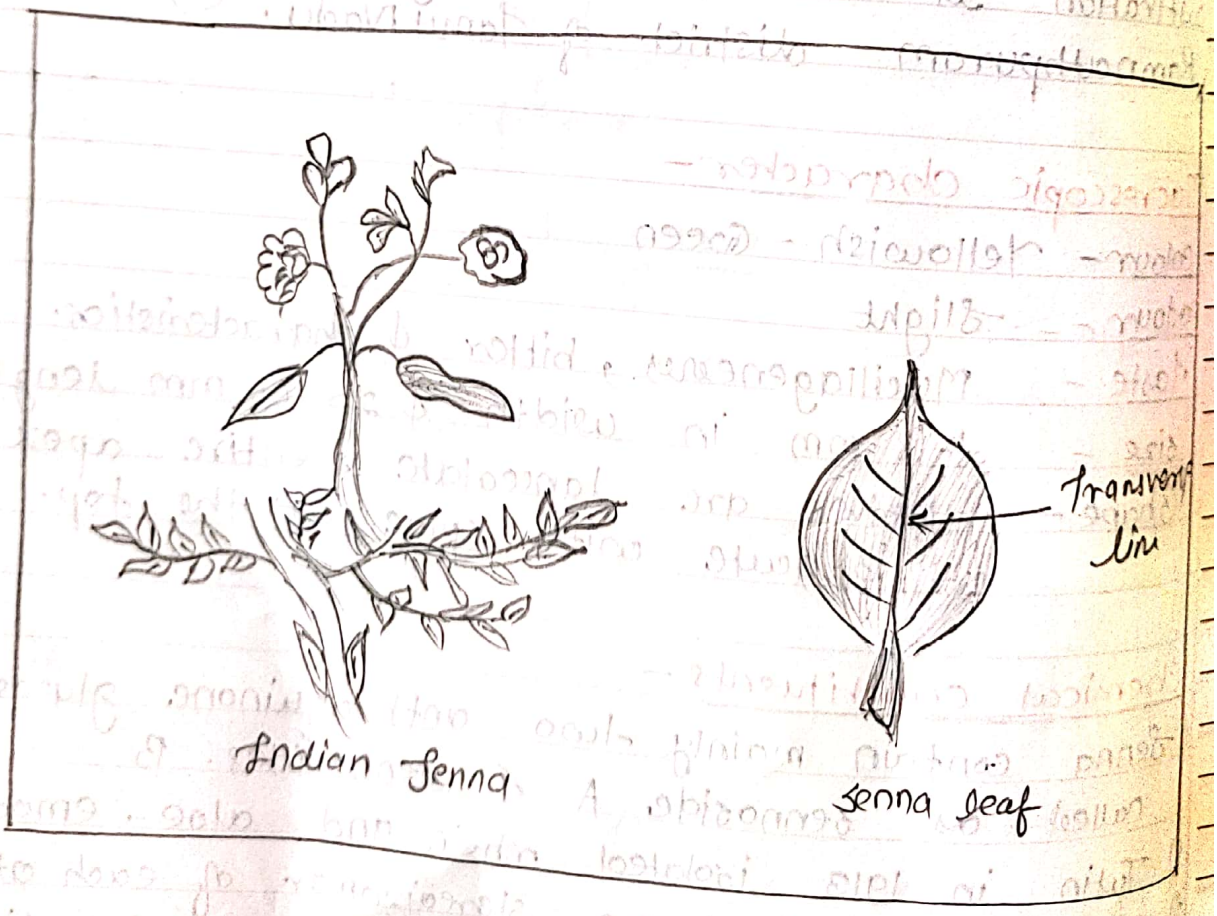


Sennoside A & B—

R = COOH

Sennoside C & D

$R = CH_2OH$



as aglycone.

5) Senna leaf also contain sennosides C, D, rhein 8-glycoside, aloe-emodin. (1)

Uses –

1) Senna and its preparation are used as purgative in perpetual constipation.
2) Senna increases the peristalsis also causes reduction in the water absorption. This results in bulky & soft faeces.
3) Gripping effect caused by senna due to its resin & emodin content. (1)

Chemical Test –

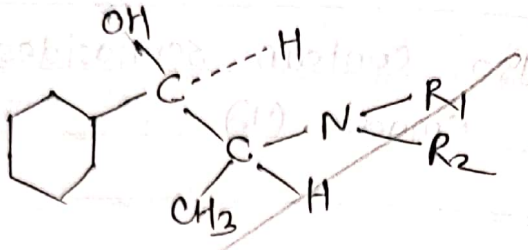
Borntrager's Test –

The drug is boiled with dilute H_2SO_4, filtered and to the filtrate, benzene and ether is added and shaken well.

The organic layer is separated to which ammonia added slowly. The ammonia layer shows pink to red colour due to presence of anthraquinone glycosides. (2)

Substituents & Adulterants –

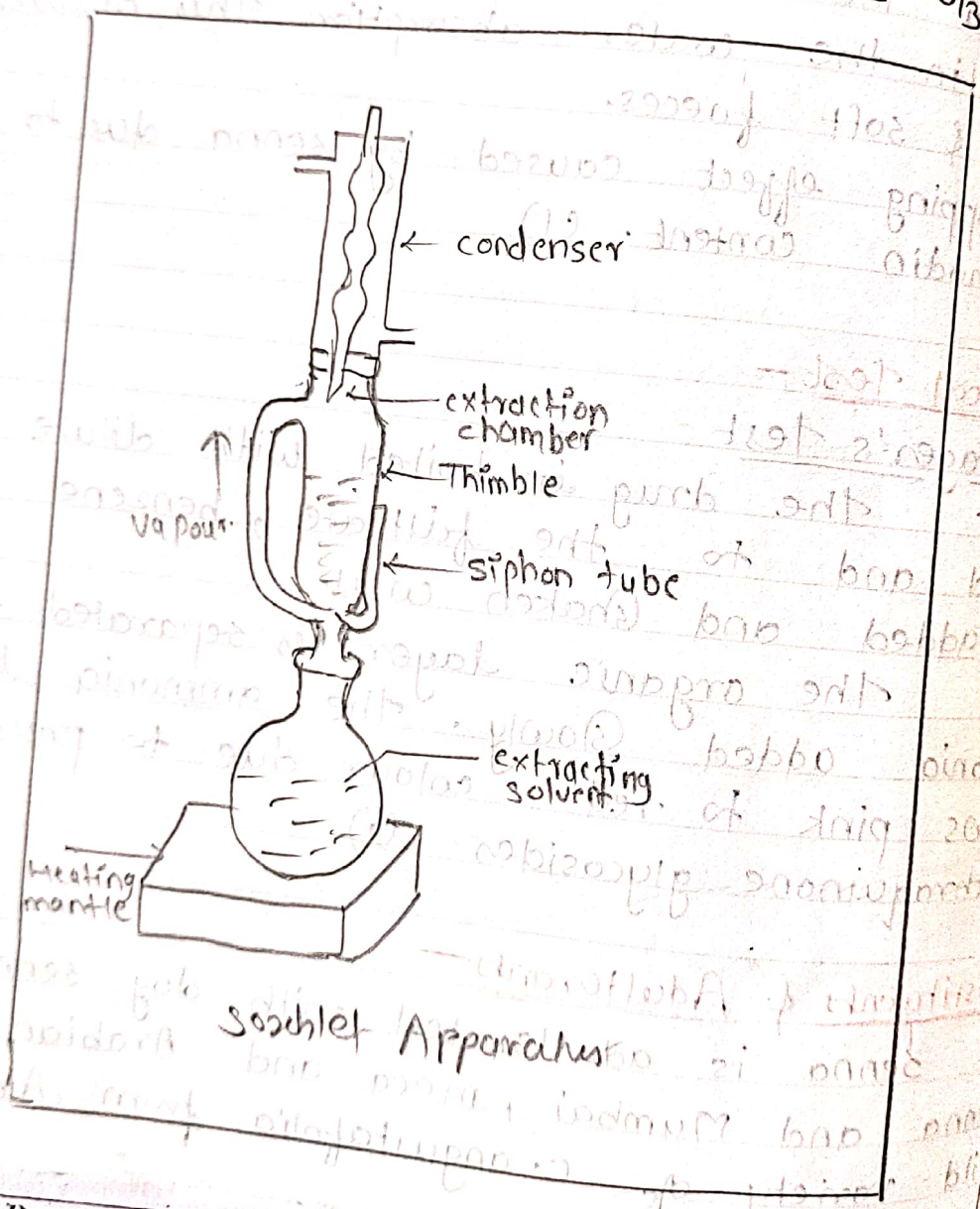
Senna is adulterated with dog senna, pathe senna and Mumbai, meca and Arabian senna mild variety of C. angustafolia from Arabica. (2)



(-) ephedrine — $R_1 = H$, $R_2 = CH_3$

(-) norephedrine — $R_1 = H$, $R_2 = H$

(-) n-methyl ephedrine — $R_1 = CH_3$, $R_2 = CH_3$



Soxhlet Apparatus

Laboratory Record/Performance (08)	Attendance (08)	Viva-Voce (04)	Total (20)	Signature of Subject Teacher
07	08	03	18	

Procedure –

1) Take the dried senna leaf and make powder form
2) Weight the 25 gm of senna powder.
3) set up the soxhlet assembly as solvent containing RBF Sample compartment and reflux condenser
4) which are separated glassware assembled together with the appropriate apparatus putting into the heating mantle.
4) The powdered senna is placed in a thimble made up of cellulose or cloth in central ~~of~~ compartment.
5) The alcohol in the RBF is heating to boiling & the vapour passes into the reflux condenser where vapours are liquifies drips into the thimble containing material i.e senna powder.
6) The condensed liquid graqually tickle down.
7) The content of thimble containing materials & extract the constituents of senna.
8) The extract containing active constituent of senna is increases and goes to siphon tube is set up liquid reaches to point of return & content of extract chamber transformed to RBF.
9) The cycle of alcohol evaporation & siphoning back into the RBF is continued as many cycles without changing ethanol with other solvent
10) The final extract of senna was collected in RBF.

Result – The isolation of chemical constituents of the senna was performed ~~successfully~~. and the % yield obtained was 55.91%.

Ephedra

Synonym - Ma-Hung

B.S - Ephedra Gerardiana Stapf.

Family - Gentaceae

Macroscopy -

Colour - Greenish Yelloco

odour - Slightly aromatic

Taste - Astringent & bitter

Shape - cylindrical

C.C - ephedrine, nor-ephe-drine, pseudoephedrine

uses - Analgesics,
Emetic,
Antirheumatic

Allied drug - Ephedra, Sinica.

Experiment No-02 Date- 26/12/18

Page No.: 11

Aim:- To perform the isolation of chemical constituent of pharmaceutical significance.

Reference:-

1) Dr. Khadbadi S.S, Dr. Deore S.L, Baviskar B.A, Book of experimental pharmacognosy, Nirali Prakashan, 1.1 - 1.9 & 5.11.

2) Kokate C.K, Purohit A.P, Gokhale S.B, Book of pharmacognosy, Nirali Prakashan, 48th edition, 15.65 - 15.66.

3) Rangari Vinod D. (2009), Nashik, Pharmacognosy & phytochemistry, vol-I, edition-II, career publication, 479-480.

Requirement:-

Apparatus - Heating mantle, soxhlet apparatus, RBF, condenser, beaker etc.

chemicals- Glycerine, Alcohol.

Principle:-

This method is used for the isolation of chemicals constituent of given drug. Soxhlet process involve the material which is to extracted is placed in thimble made up of cellulose in the centre of the assembly of the soxhlet apparatus

* Calculation

1) wt of empty porcelin dish

A = 70.79 gm

B = 63.64 gm

2) wt of porcelin dish + sample

A = 73.60 gm

B = 65.79 gm

3) wt of sample A = 73.00 − 70.79

= 2.21

4) wt of sample B = 65.79 − 63.64

= 2.25

Total wt = 2.21 + 2.25

= 4.46 gm

5) Theoritical yield = 25 gm

% yield = $\frac{4.46}{25} \times 100$

= 17.84 %

EPHEDRA

Synonym – Ephedra, Ma-Hung. [2]

Biological Source –

It consists of the dried young Stems of Ephedra Gerdiana (Wall) Staf.

Family – Gentaceae. [2]

Geographical Source –

The main source of Ephedra is from China, Pakistan, North-West part of India; Australia. [2]

Macroscopic character –

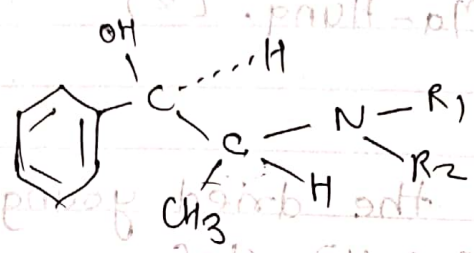
Colour – Grey to Greenish.
odour – odourless
Taste – Tasteless
size – about 5 mm in diameter, 3-3.5 cm
Shape – cylindrical. [1]

Chemical Constituents –

1) Ephedra contains the amino-alkaloid. They are ephedrine, nor-ephedrine, n-methyl ephedrine, pseudo-Ephedrine.
2) chemical ephedrine is 1-phenyl-1-hydroxy-2 methyl-amino Propane.
3) It is soluble in water, alcohol, organic solvents & oils. [2]

EPHEDRA

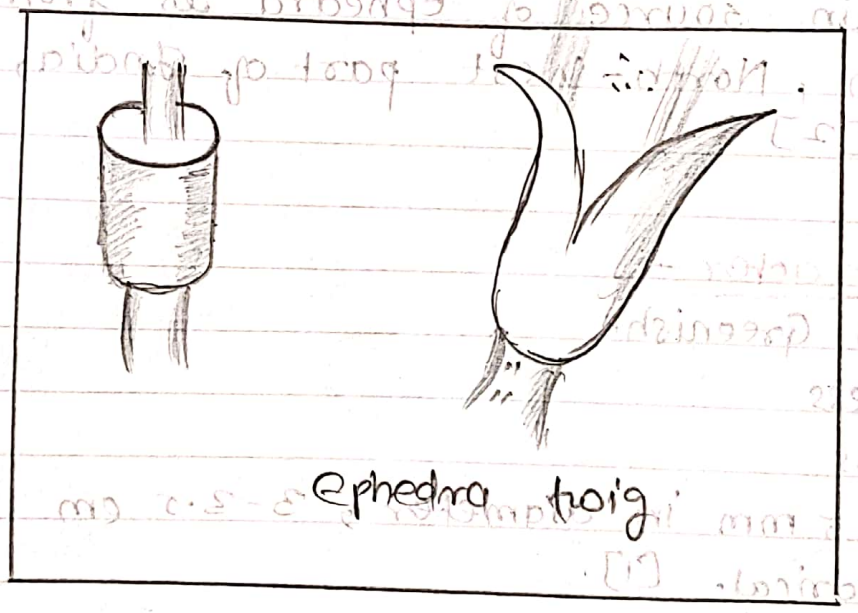
chemical constituents -



(-) Ephedrine $R_1 = H$ $R_2 = CH_3$

(-) nor ephedrine $R_1 = H$ $R_2 = H$

(-) n-methyl ephedrine $R_1 = CH_3$ $R_2 = CH_3$



Ephedra troig

Uses –

1) Ephedra shows sympathomimetic effect.
2) It is used as bronchodilator in asthma & also in treatment of allergic conditions like hay fever.
3) Ephedrine is also used to correct the low blood pressure condition, because of its peripheral contraction of arterioles.
4) Ephedralines have hypotensive effect. (2)

Chemical Test

Ephedrine is dissolved in water and dilute HCl and then treated separately with copper sulphate & sodium Hydroxide The solution given violet colour. If shaken with solvent ether, the organic layer shows purple and aqueous layer show blue colour.

Adulterants –

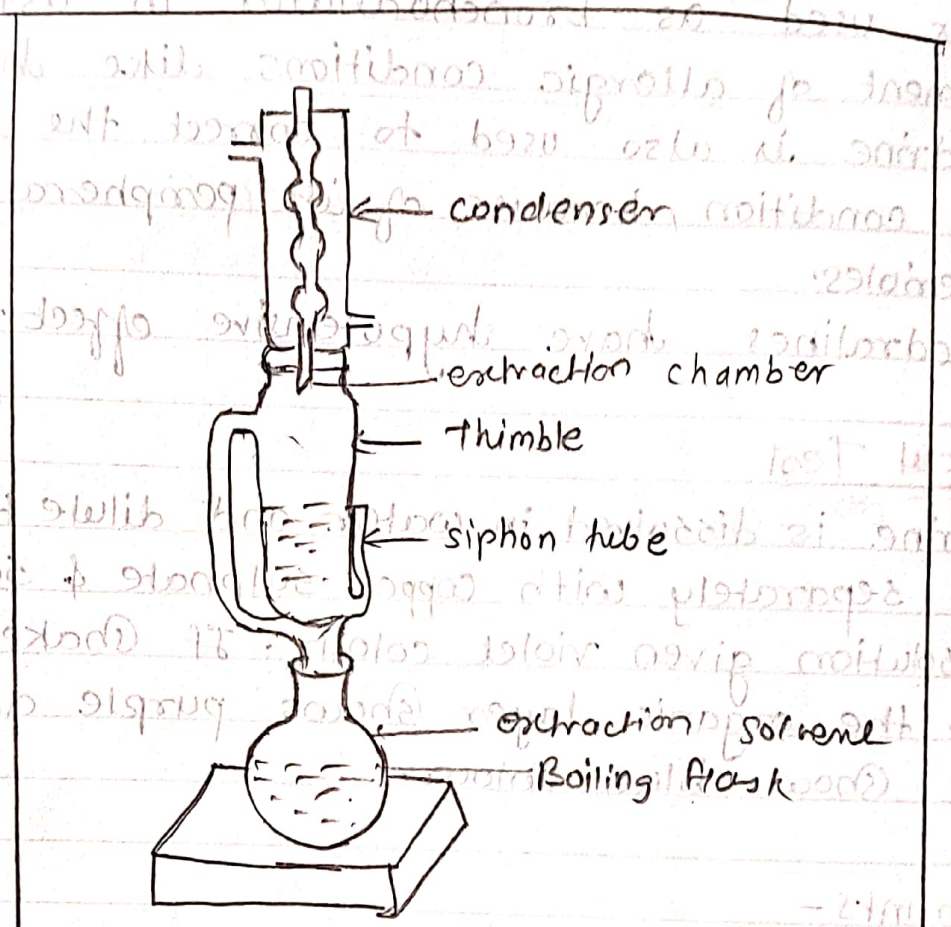
The species containing ephedrine are E. Equisetina & E. sinica. (2)

Allied Drug –

The plants having contents similar to colchicine type of alkaloids belong to different genera such as Dipidax, Gloriosa, Androcybium. (3)

Procedure –

1) Take 25 gm of powder of ephedra to isolate the active chemical constituents from drug.
2) Soxhlet assembly is set and used to extract or isolate the active constituents from drug.



Soxhlet Apparatus

3) Glycerine is used as Menstrum
4) Place the powdered drug in a thimble or extraction chamber
5) Cotton is placed at the bottom before drug is place.
6) Solvent (alcohal) is filled in flask & connect assembly
7) The alcohal in the RBF is get heated & vapour are passes through side tube to condenser.
8) At condenser, the vapour are condensed and drops into the thimble over drug.
9) Due to which the extract of active constituents of ephedra is obtained.
10) These extract is return to the RBF through siphon tube & isolation of constituents of ephedra is continued upto 3-4 cycles.
11) The final extract is collected in RBF.

Result :-

The isolation of chemicals constituents from ephedra was performed ~~successfully~~. and the % yield was obtained 14.84 %.

Laboratory Record/Performance (08)	Attendance (08)	Viva-Voce (04)	Total (20)	Signature of Subject Teacher
08	08	02	18	

Cinchona

Synonym - Jesuits Bark

B.S - Dried bark of cinchona calisaya wedd.

Family - Rubiaceae

Macroscopic characters -

colour - Yellowish brown

odour - Slight

Taste - Intensely bitter

C.C - Quinoline, quinidine, & cinchonine.

Uses - Antimalarial & Antipyretic.

Substituents & Adulterants - cuprea bark

Experiment No- 03 Date - 2/1/19

Page No.: 15

Aim- To perform the isolation of chemical constituent of pharmaceutical significance (cinchona)

Reference-

1) Kokate C.K, Purohit A.P, Gokhale S.B (2010) Pune, Pharmacognosy, volume-I, 48th edition, Nirali Prakashan 15.88 - 15.59

2) Dr. Khadbadi S.S, Dr. Deore S.L., Mr. Baviskar B.A. (2011) Experiment pharmacognosy Pune, 1st edition, Nirali Prakashan, 5.18 - 5.17

3) Rangari Vinod D. (2009), pharmacognosy & phytochemistry, vol-II, 2nd edition (2009) career publication. 458 - 469

Requirement -

Apparatus- Soxhlet apparatus, reflux condenser, RBF, Heating mantle, beaker.

Chemicals- Drug powder, Alcohol, glycerine.

Principle -

This method is used for the isolation of active chemical constituents of given drug. Soxhlet process involves the material which is to extracted is placed in thimble made up of cellulose in centre of the assembly of soxhet apparatus.

Calculation:-

Dish	empty wt of dish	empty wt of dish + sample	wt of sample
1)	64.24	67.47	3.23
2)	67.93	69.43	1.50
3)	63.08	65.82	2.74
4)	57.92	59.97	2.05
5)	52.51	53.49	00.98
6)	63.40	64.81	1.41
Total wt of sample =			= 11.91 gm

% wt of sample

50 gm ⟶ 11.91

100 ⟶ x

$$x = \frac{100 \times 11.91}{50}$$

$$x = 2 \times 11.91$$

wt % = $\boxed{x = 23.82\ \%}$

Cinchona

Synonyms - Jesuit's bark. [1]

Biological source

It is obtained from dried bark of cultivated trees of cinchona calisaya wedd. cinchona officinalis linn. [1]

Family - Rubiaceae [1]

Geographical source -

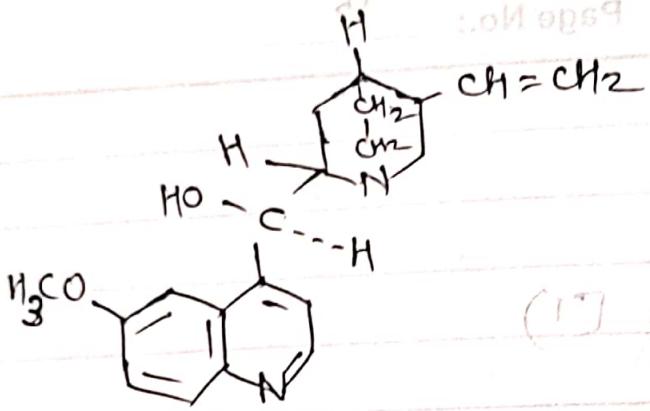
India, Bolvia, columbia, Eucador, Peru, Tanzania, Indonesia & Sri-Lanka.

Macroscopic character -

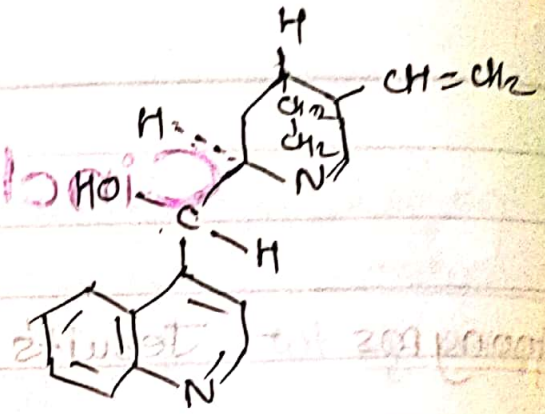
colour - Brown grey
odour - slight and characteristics
Taste - Astringent and intensely bitter
Shape - quills and curved pieces.
Size - 30 cm in length & about 2-6 cm in thickness.

Chemical constituents -

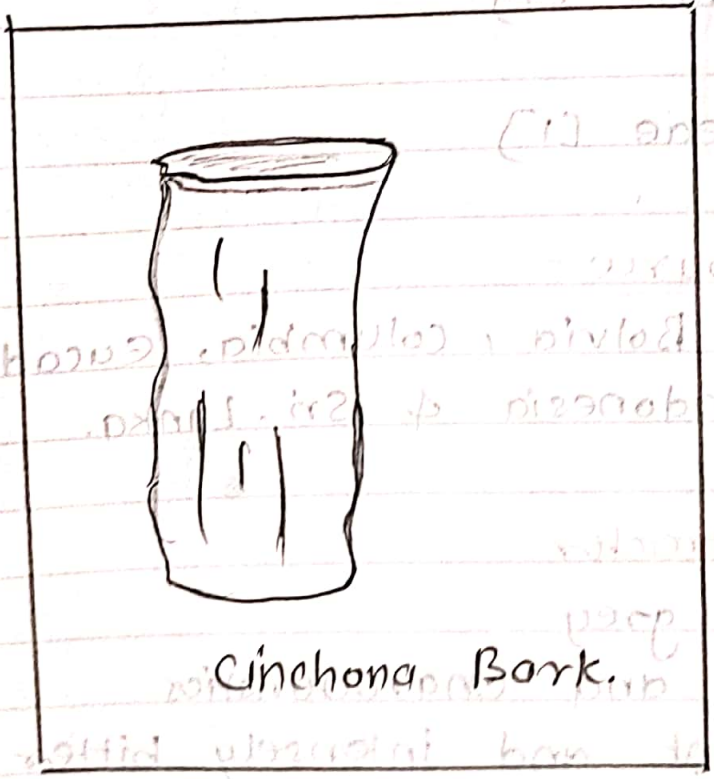
1) cinchona bark contains about 25 alkaloids, which is belong to quinoline group.
2) The important alkaloid are quinine, quinidine, cinchonine, cinchonidine.
3) Cinchona contains quinic acid & cinchotannic acid.



Quinine



Cinchonine



Uses -

1) cinchona bark is antimalarial in nature
2) The cinchona preparation like cinchona extract comp. cinchona tincture effect shows.
3) This also employed as bitter stomachics and antipyretics ~~to~~
4) Quinidine is primarily a cardiac depressant and used to prevent certain arrythmia. [2]

Chemical Test

1) Thallequine Test -

The powdered drug gives emerald green colour with bromine water & dil. the ammonia solution.

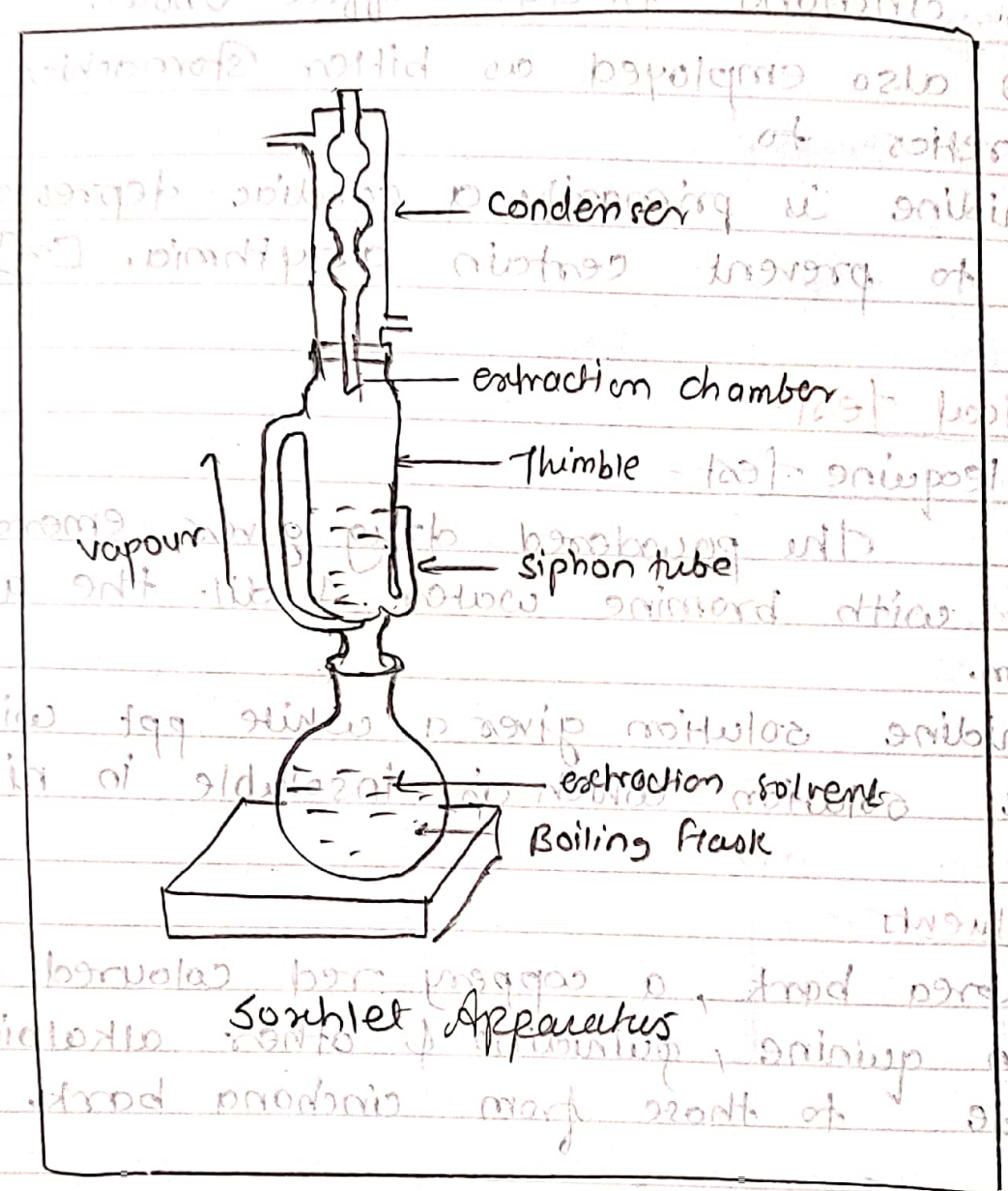
2) Quinidine solution gives a white ppt with silver nitrate solution which is insoluble in nitric acid. [1]

Substituents -

Cuprea bark, a coppery red coloured drug contain quinine, quinidine & other alkaloids which resemble to those from cinchona bark.

Procedure -

1) Take 25 gm of powdered cinchona bark to isolate the active constituent from drug
2) soxhlet apparatus are used to extract or isolate the active constituents from drug.
3) Glycerine is used as a menstrum.
4) Place the powdered drug in a thimble or extraction chamber.



Soxhlet Apparatus

5) Cotton is placed at the bottom before drug is placed.
6) Solvent (alcohal) is filled in flask & connect assembly.
7) The alcohol in RBF is get heated and vapour are passes through side tube to condenser.
8) At the condenser the vapour are condensed and drops into the thimble over drug
9) Due to which the active constituent of cinchona is extracted out.
10) These extracted is return to the RBF through siphon tube & isolation of constituents of the Continued to 3-4 ~~days~~ cycles.
11) The final extract is collected in the RBF

Result -

The isolation of chemical constituent of cinchona bark was performed successfully.

Laboratory Record/Performance (08)	Attendance (08)	Viva-Voce (04)	Total (20)	Signature of Subject Teacher
08	08	02	18	

- Introduction -

Chemical methods used for separation -

like physical methods, chemicals methods may also be used for isolation & purification of natural products These method may convert a component in recoverable OR non-recoverable derivatives OR fragments. which can later be separated & purified.

Hydrolysis -

Acid Hydrolysis -

The glycoside containing extracts can be subjected to acid treatment. This hydrolysis the glycoside into glycone and aglycone.

Enzyme Hydrolysis -

For the purpose of hydrolysis, enzyme are sometimes employed for establishing the nature of linkage of sugar to glycoside i.e α OR β also in therapy it offers a method for clearing of specific monosaccharides from glycosides.

Alkaline Hydrolysis -

Alkaline hydrolysis employ dilute alkalies such as sodium hydroxide OR pottassium hydroxide it is rarely used for the hydrolysis of glycosides. The process is used for the removal of acyl group.

Salt formation –

Most of alkaloidal bases which are soluble in organic solvents are converted to their water soluble quaternary ammonium salts.

Morphine (Base) ⇄ (HCl →, ← NH3) Morphine Hydrochloride (salt)

Eugenol (Phenol) ⇄ (KOH →, ← HCl) Potassium eugenolate (Phenoxide)

Acetylation –

It converts hydroxyl groups into their acetyl derivatives which can be isolated & crystallized easily from the mixture.

Thin Layer Chromatography

TLC is mainly used qualitatively by screening different plant extracts, which serves as a very imp tool in overall phytochemical research studies.

Preparation of TLC Plate –

1) Glass plates of diffs sizes like 20×20, 20×10, 20×5 cm are used for TLC study.
2) Adsorbent such as silica gel OR alumina is used as a stationary phase and water as a solvent for prepn of slurry.

3) Various techniques such as pouring, dipping or spreading are used for the preparation of TLC plates.
4) The prepared plates are air dried and activated in the oven at 105°C for 30 min.
5) Aluminium precoated TLC plates are also available for TLC study.

Application of samples -

Standard Borosil glass capillaries are used for applying samples on the prepared plates. Spots are applied at suitable distance & allowed to dry.

Preparation of saturated chamber -

The solvent system used as a mobile phase is selected, depending upon the nature of the phytoconstituent. The TLC chamber is allowed to saturate with the fumes of the solvents prior to use, so as to avoid unequal solvent evaporation losses from the developing plates.

The plates are generally observed under UV radiation & colour & fluorescence are noted.

General reagents, such as iodine, In iodine chamber, 1% vanillin in sulphuric acid or 50% methanol in H_2SO_4, are used as spraying reagents

RF value is calculated as follows -

$$RF = \frac{\text{Dist. Travelled by comp. from origin (X)}}{\text{Dist Travelled by solvent from origin (Y)}}$$

Experiment No - 04 Date - 9/11/19

Aim - To perform separation of chemical constituent of pharmaceutical significance (Senna).

Reference -

1) Kokate . C . K . , Purohit A . P . , Gokhale S . B (2010), Pune, Pharmacognosy , Vol - I , edition 46th , Nirali Prakashan, 8 . 9 – 8 . 14

2) Dr. Khadbadi S . S , Dr. Deore S . L . , Mr. Baviskar B . A , (2011) , pune , Experimental Phytopharmacognosy , edition - I Nirali Prakashan , 6 . 1 – 6 . 18 .

3) Rangari V . D , (2009) , Nashik , Pharmacognosy and Phytochemistry , volume – II , edition - IInd , career publication , 105 – 110 .

Requirements -

Apparatus - TLC plate , TLC chamber.
chemicals - silica gel , Senna extract.

Principle - The interaction include the physicochemical principle such as adsorption partition ion-exchange molecular or sieving & affinity.

Two phases & involves in chromatography
i) Stationary phase,
ii) Mobile phase

Senna

Synonym- Senna leaf, senna-ki-patti [1]

B.S It consists of dried leaflets of cassia angustifolia or cassia senna vahl (1)

Family - Leguminaceae. (1)

Macroscopic character-

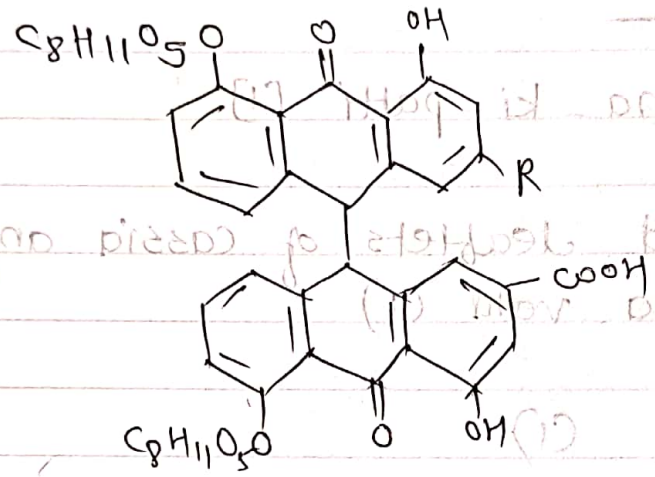
colour - yellowish-green.
odour - slight
Taste - Mucilaginous, bitter & characteristics.
size - 1-8 mm in width & 25-60 mm length.
Shape - Leave are lanceolate, entire apex is acute with spine or the top (2)

Geographical source

Indian senna is cultivated in India. Its cultivation is done in Tinnevelly, Madurai and Romanathpuram district of Tamil Nadu. (3)

Chemical constituents-

1) senna contain mainly two anthraquinone glycoside called as sennoside A & B
2) Tutin in 1913 isolated rhein and aloe-emodin.
3) Sennoside A & B are stereo-isomers of each other. [2]



Sennoside A & B

R = COOH

Sennoside C & D

R = CH_2OH

User -

1) Senna and its preparation are used as purgative in perpetual constipation.
2) Gripping effect caused by senna due to its resin and emodin content.
3) Senna increase the peristalsis also causes reduction in the water absorption. This result in bulky & soft faeces [1].

Adulterants & Substituents -

Senna is adulterated with dog senna, palthe senna and Mumbai, Mecca & Arabian senna mild variety of C. angustafolia from Arabia [1].

Procedure -

1) About 15 gm of silica gel is made into slurry with 30 ml of distilled water.
2) The chromatography plates are mixe followed with chromic acid by washing with distilled water & dried in oven with the D.W & the spreading layer of 300 µm width is coating is on the cleaned plate.
3) The coating is allowed by dry at R.T & plate are activated in an oven at 120°C for 20 min.
4) The solvent system is poured in TLC chamber to the depth of 3 cm and allow to separated for 3 hrs. The chamber is lined with paper on side to maintain equilibrium of mobile phase

Calculation -

1) Distance travelled by solvent = 15.5
2) Distance travelled by ~~solvent~~ solute from base line = 8.4

③ R.F value

$$= \frac{\text{distance travelled by comp. from origin}}{\text{distance travelled by solvent from origin}}$$

$$= \frac{8.4}{15.5}$$

$$= \text{R.F value} = 0.54$$

5) The test solution of total glycoside extract of Standard solution of sennoside are prepare in $CaCl_3$ (1 mg/ml) and 10 ml of solution are loaded on activated plates & with the aid of calibrated capillary chromatography allowed to develop for 10 cm take out the plate mark the solvent from & dry at RT that take out the plate solvent from & dry respective temp.

6) Observed the plates & dry respective temp. the glycosides Shaos flurescence spray plates with bronth triger reagent & dry the warming identify the location of glycoside which take colouration.

Result -

The RF value of chemical constituents of senna was found to be 0.54

Laboratory Record/Performance (08)	Attendance (03)	Viva-Voce (04)	Total (20)	Signature of Subject Teacher
08	08	02	18	

Experiment No - 05

Date - 18/1/19

Aim - To perform separation of chemical constituent of pharmaceutical significance (Ephedra).

Reference -

1) Kokate C.K., Purohit A.P., Gokhale S.B (2010), Pune, Pharmacognosy, volume - I, edition - 48th, Nirali publication, 15 - 65 - 15.66.
2) Dr. Khadbadi S.S., Dr. Peore S.L, Mr. Baviskar B.A. (2011), Pune, experimental pharmacognosy edition - I, Nirali Prakashan 1.1 - 1.9.
3) Rangari Vinod. D. (2009), Nashik, pharmacognosy, edition - II, career publication, 274 - 275

Requirement -

Apparatus - TLC plate, TLC chamber
chemicals - silica gel, senna extract

Principle -

The interaction include the physicochemical principle such as adsorption partition ion - exchange molecular or seiving & affinity.

Two phases involves in chromatograph
1) Stationary phase &
2) Mobile phase

Ephedra

Synonym – Ephedra, Ma-Huang (2)

Biological Source – It consist of the dried young Stem of Ephedra Gerardiana (Wall) ~~Stapf~~ Stapf.

Family – Gentaceae (2)

Geographical source.

The main source of Ephedra is from China, Pakistan, North-West part of India, Australia. (2).

Macroscopic Character-

Colour- Grey to Greenish.

odour – odourless

Taste – Tasteless

size – About 5 mm in diameter, 3-3.5 cm

Shape – Cylindrical (□

Chemical Constituents –

1) Ephedra contains the amino-alkaloid. They are ephedrine, nor-ephedrine, n-methyl ephedrine, Pseudo-ephedrine.

2) Chemical ephedrine is 1-phenyl-1-hydroxy-2-methyl amino propane. (2)

User -

1) Ephedra shows sympathomimetic effect.
2) Ephedranaline have hypotensive effect.
3) It is used as bronchodialator in asthma & also in treatment of allergic conditions like hay fever. (2)

Allied drug -

The plants having contents similar to colchicine type of alkaloids belong to different genera such as dipidax, Gloriosa, Androcybium (3).

Procedure -

1) About 15 gm of silica gel is made into slurry with 30 ml of distilled water.
2) The chromatography plates are mixe followed with chronic acid by washing with distilled water and dried in oven with the D.W and spreading layer of 30 µm width is coated on cleaned plates.
3) The coating is cleaned plates allowed to drug at R.I & plates are activated in an oven at 120° for 20 min.
4) The solvent system is poured in TLC of depth of 3 cm and allowed to saturated for 3 hrs. The chamber is lined with filter paper on 3 sides to maintained equilibrium of mobile phase

* Calculation -

1) Distance travelled by solvent = 9.5
2) Distance travelled by solute from base line = 3.6

③ R.F. Value -

$$= \frac{\text{distance travelled by comp from origin}}{\text{distance travelled by solvent from origin}}$$

$$= \frac{3.6}{9.5}$$

R.F value = 0.37

5) The test solution of total glycosides & extract of the Standard solution ephidrine are prepared in CCl_3 (1 mg/ml) & 10 ml of solution are on active plate with aid of calibrated in active ~~plate with~~ capillary chromatogram is allow to developed for 10 cm take out plates mark the solvent from & dry at respective temperature.

Result - The RF value of the ephedra extract was found to be 0.35

Laboratory Record/Performance (08)	Attendance (08)	Viva-Voce (04)	Total (20)	Signature of Subject Teacher
08	08	02	18	