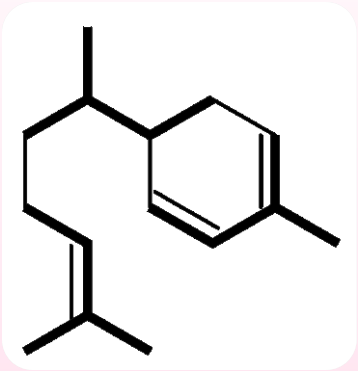
Phytochemistry

Essential oils



Amal Kabbash, *Professor of Pharmacognosy*
Vice Dean for Education and Students Affairs

ILOS of the Course:

Intended Learning Outcomes

a- Learning and Understanding

b- Intellectual Skills

c. Professional and Practical Skills

d- General and Transferable Skills

Intended Learning Outcomes of Course: ILOS

a- Learning and understanding

a1- Enumerate the several groups of alkaloids and volatile oils.

a2- Describe the botanical occurrence, extraction, isolation, identification and determination procedures.

a3- Describe the biosynthesis and the therapeutic effects of different alkaloids and volatile oils.

b- Intellectual Skills

b1-**Differntiate** between the chemical structures and uses of different groups of alkaloids and volatile oils.

b2- **Summarize** the possible leads to new drugs depending on natural product templates

b3- **Determine** the different methods used for quantitative and qualitative determination of different groups of alkaloids and volatile oils.

c. Professional and Practical Skills

c1- **Summarize** the practical phytochemical procedures.

c2- **Provides examples** of the different groups of alkaloids and volatile oils in terms of their **uses, chemical group, origin**.

c3- **Identify** different groups of alkaloids and volatile oil

c4- **Analyze** different groups of alkaloids and volatile oil

d- General and Transferrable Skills

d1- Perform search and retrieve of information

d2- Collaborate in laboratory work

d3- Collaborate in the writing of reports

Course Evaluation

- **Total Marks: 500**

Mid term: **50**

Practical exam.: **100** (include attendance, note, behavior, activity)

Oral exam.: **100** (Lecture attendance may affect)

Final written exam.: **250**

(Problem based evaluation)????


Lab
Safety

Safety measures

Do NOT

- × eat, drink or smoke in the laboratory .
- × pipette by mouth
- × leave equipment using water, gas or electricity on overnight
- × Never add water to conc. Sulphuric acid

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SAFE PERSONAL PROTECTIVE PRACTICES

- ☑ listen carefully to instructions
- ☑ wear lab gown
- ☑ tie back long hair
- ☑ wear safety glasses
- ☑ wear disposable gloves if necessary

Volatile Oils

Essential oils, Volatile Oils, Etherial oils
Terpenoids

- Definition
- Difference from fixed oils
- Distribution, localization and function
- Uses
- Physical characters
- Methods of preparation
- Quality control
- Storage
- Chemistry
- Classification

VOLATILE, ETHEREAL or ESSENTIAL OILS

- **"Volatile" or "ethereal": as they easily evapor ate on exposure to air at room temperature**
- **"Essential":** contains the "**essence of**" the plant's **fragrance** i.e: the characteristic fragrance of the plant from which it is derived
- Defination: Class of volatile compounds that give plants their characteristic odors and are used especially in perfumes and flavorings, and for aromatherapy

- **Aromatic: since most have aromatic odor**

Major Differences between volatile & fixed oils

Property	Volatile oil	Fixed oil
Volatilization at ordinary temperature	Volatile	Non-volatile
Solubility	Soluble in organic solvents (ether, $CHCl_3$) & alcohol	Limited solubility in organic solvents, almost insoluble in alcohol
Stain on filter paper	Transient	Permanent & greasy
Composition	Complex mixtures of hydrocarbons & oxygenated compounds	Triglycerides ester of fatty acids e.g. palmitic, stearic, oleic……
Response to long exposure to air & light (oxidation)	Resinification	Rancidity
Saponification with caustic alkali (KOH)	Negative	Positive

Distribution, localization and function

Distribution

In higher plants, different families as: Rutaceae, Myrtaceae, Lamiaceae (Labiatae), Lauraceae,…..

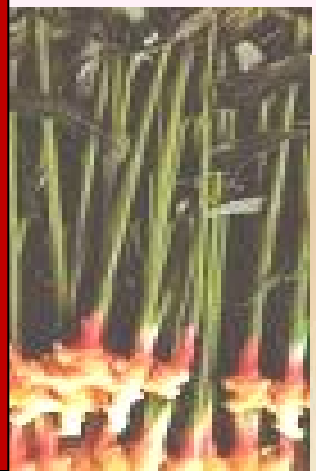
Accumulate in all types of vegetable organs:

Flowers (rose), Leaves (eucalyptus), Barks (cinnamon),

Woods (Sandalwood)

Roots (vetiver.)

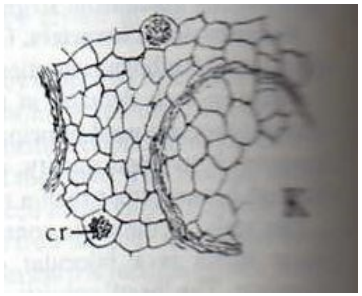
Rhizomes (ginger),

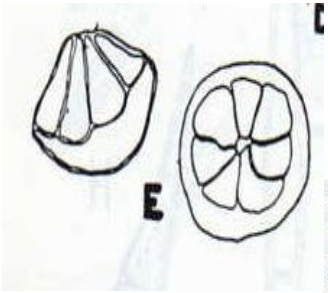


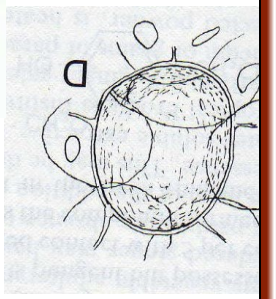
Fruits (star anise),

Seeds (nutmeg)

Localization

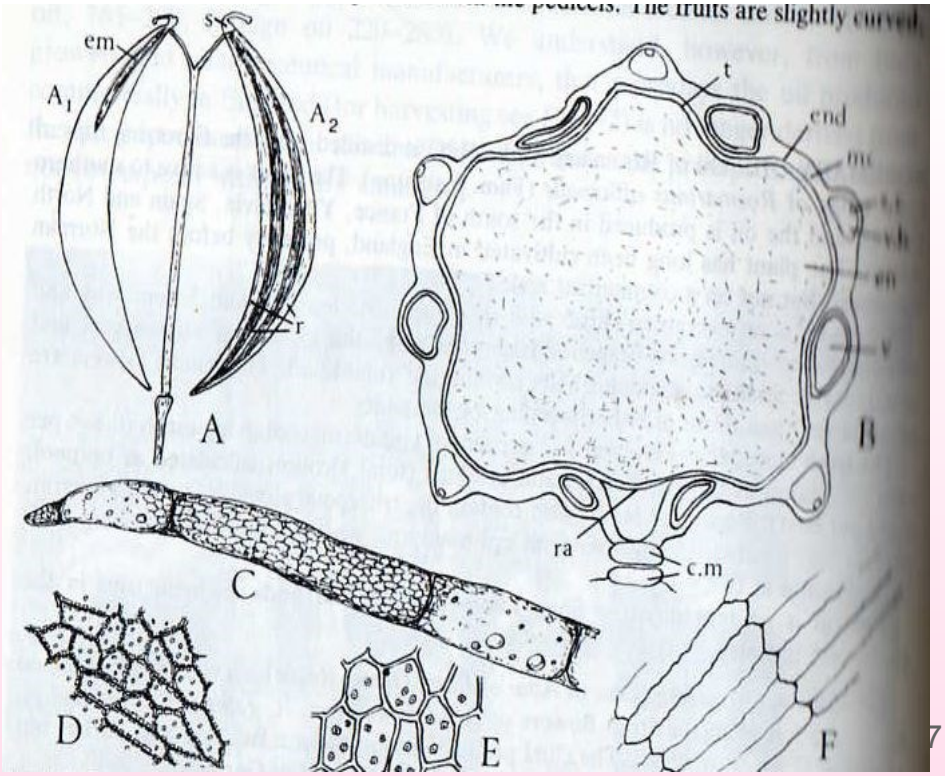






Usually in specialized histological structures

- Oil cells: Lauraceae, Zingiberaceae
- Glandular trichomes: Lamiaceae
- Secretory cavities: Rutaceae, Myrtaceae
- Secretory canals: Apiaceae (Umbelliferae).



Uses of Volatile Oils

1. **Therapeutic & medicinal uses:** local stimulants, carminatives, diuretics, mild antiseptics, local irritants, anthelmintics, parasiticides, respiratory infection, immune enhancing, anti-inflammatory,
2. **Spices & condiments:** in food (to impart aroma & flavor) or as preservatives
3. **Flavoring agents:** in food (e.g. beverages, soups, bakery products, confectionery) & pharmaceutical industries
4. **Aromatic agents:** in all types of perfume industries (cosmetics, soaps, deodorizers)

➢Function to the plants itself

- Attracting (for polination) or repelling insects
- Protection from heat or cold
- As antibacterial agents

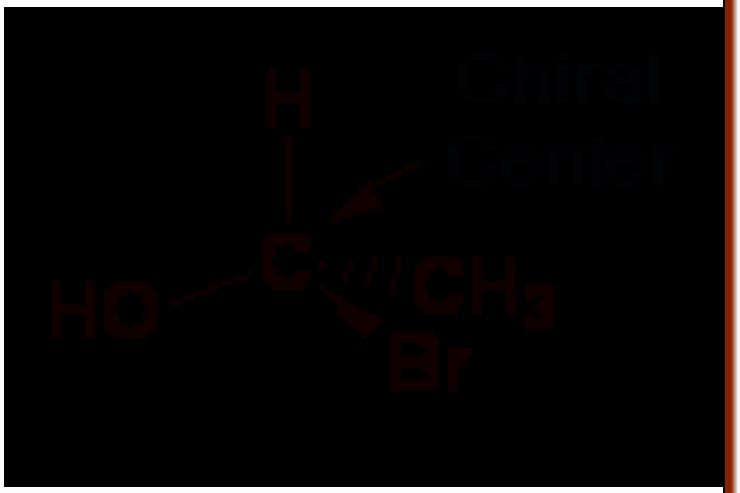
Physical properties

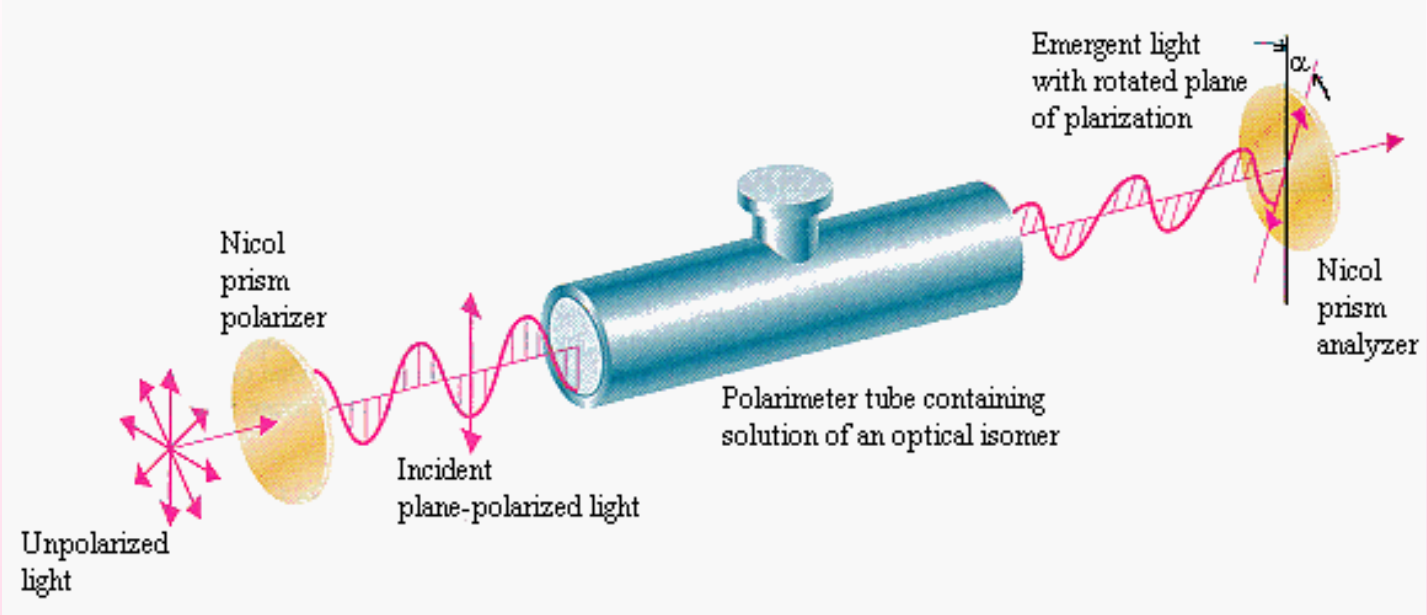
- Possess characteristic odors (lemon and lavender oils)
- Liquids and volatile at ambient temp.

Except anise oil becomes solid at 15.5°C and rose oil solidify at 18 °C

- Soluble in common organic solvents. Sparingly soluble in water.
- Mostly lighter than water, except clove, winter green and cinnamon oils are heavier (aromatic oxygenated oils).
- Most of them rotate the Plan of polarized light.

Automatic Polarimeter





Methods of preparation
there are four main methods

❖ Distillation
- Water distillation
- Water and steam distillation
- Direct steam distillation

❖ Scarification and expression
- Sponge method
- Ecuelle a piquer
- Expression of rasping

❖Extraction with solvents

❑Volatile solvents

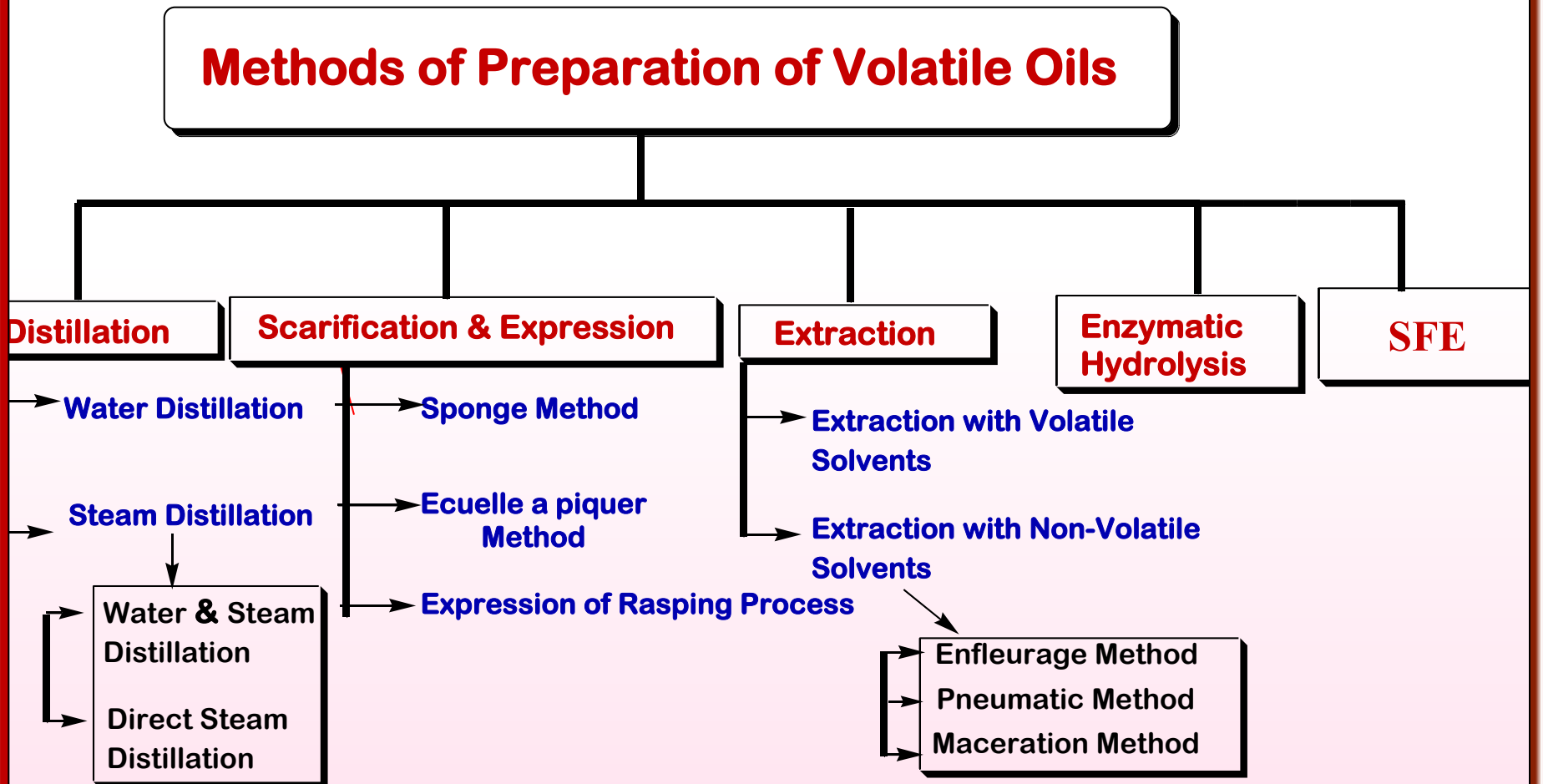
❑Non volatile solvents

- Enfleurage method
- Pneumatic method
- Maceration method

❖ **Supercritical fluid extraction**

❖**Enzymatic hydrolysis**

Methods of Preparation of Volatile Oils



Selection of the suitable method is done according to :

1. The condition of the plant material (moisture content, degree of comminution)
2. The localization of the oil in the plant (superficial or deep)
3. The amount of the oil
4. The nature of the oil constituents

Distillation methods

Application: preparation of thermostable oils, present in large amounts & not rich in esters (e.g. oils of turpentine, peppermint, ..

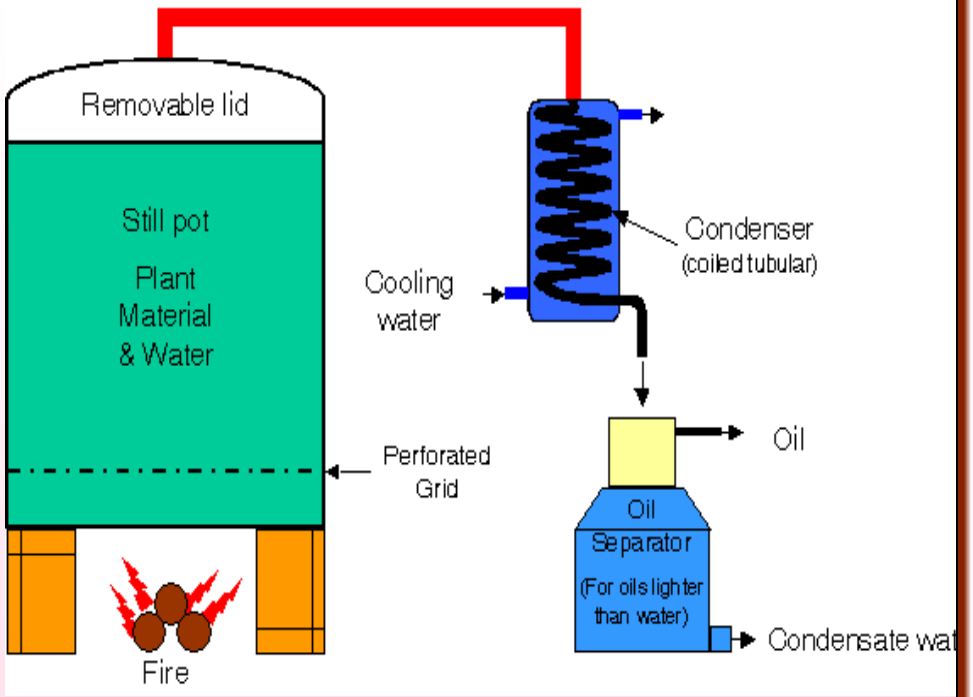
Distillation

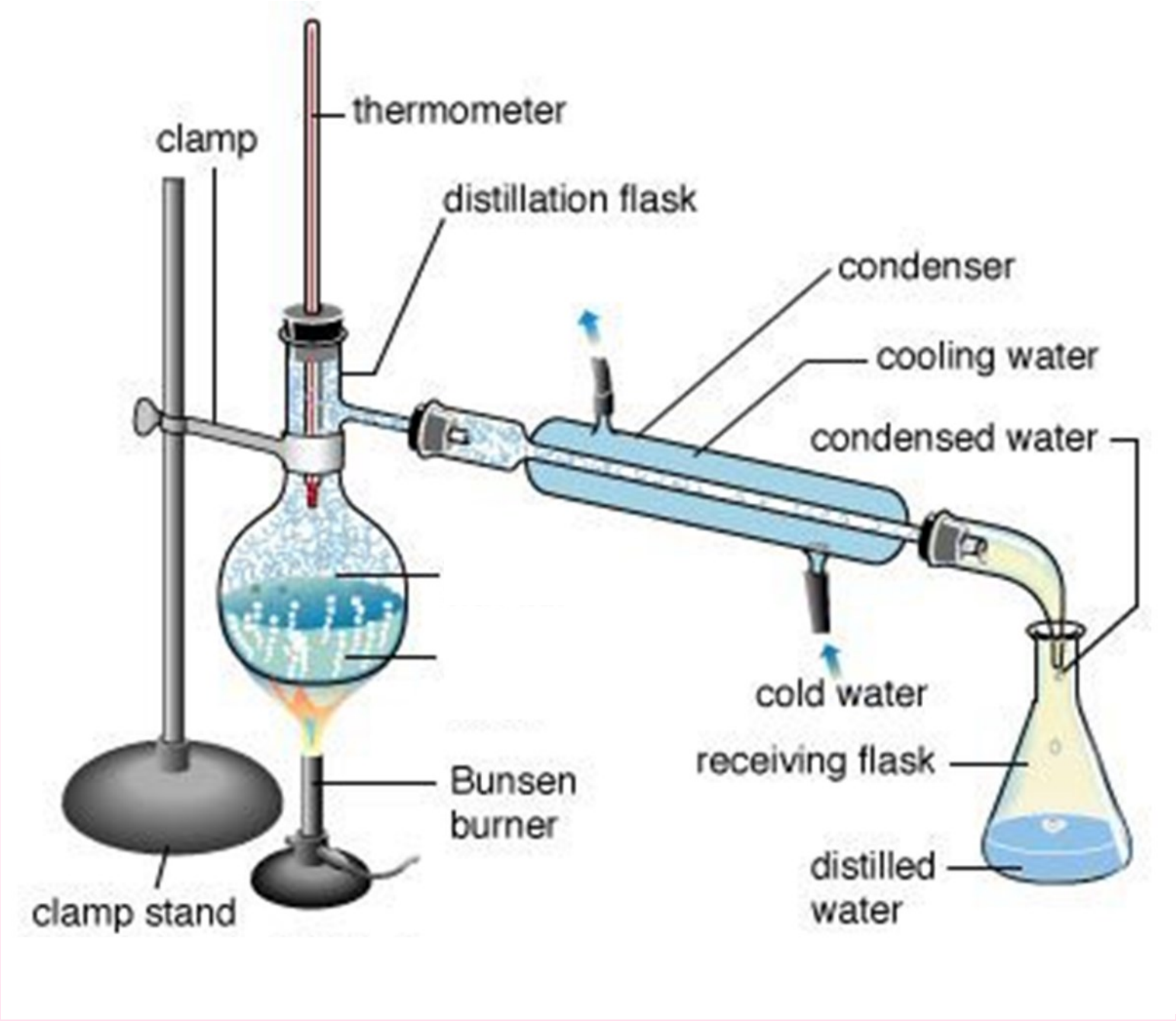
- Converts the essential oils into vapor and then condenses the vapor back into a liquid.
  - Oil of terpentine **Boils at** 160 °C, water → 100 °C
  - Oil of terpentine + water → 95.5 °C
- Advantage: **Wide Application**
- Disadvantage

Water distillation

Plant material (dried) is completely immersed in wa
er and the still is brought to the boil, when the cond
ensed material cools down ⟶ water + v. oil

e.g oil of turpentine.

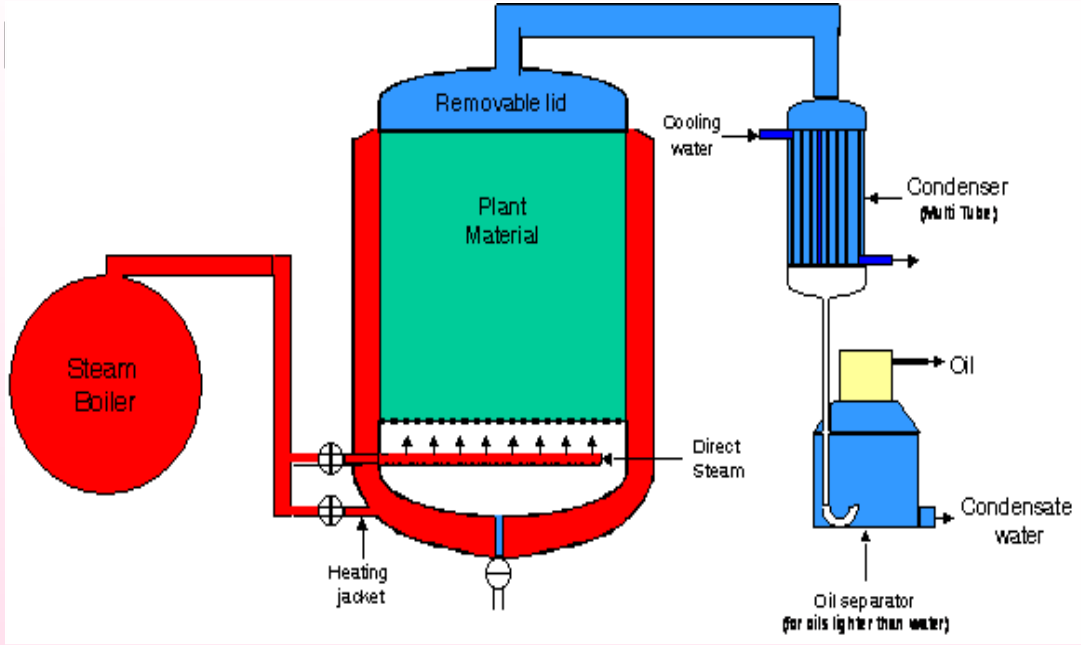



thermometer
clamp
distillation flask
condenser
cooling water
condensed water
cold water
receiving flask
Bunsen
burner
distilled
water
clamp stand

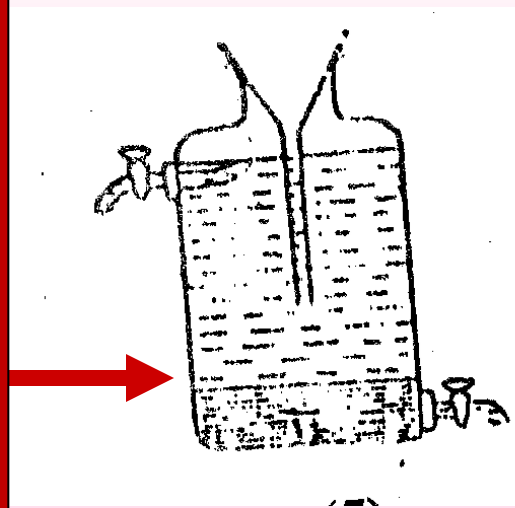
Direct steam distillation

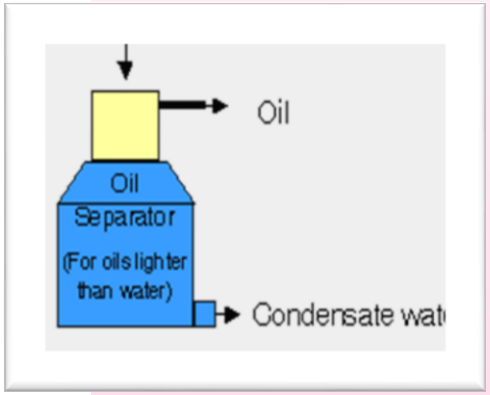
Plant material (fresh) is placed in a still and steam is forced through the material. The hot steam helps to release the aromatic molecule s from the plant material and carries them (oil droplets) to the condensation chamber.

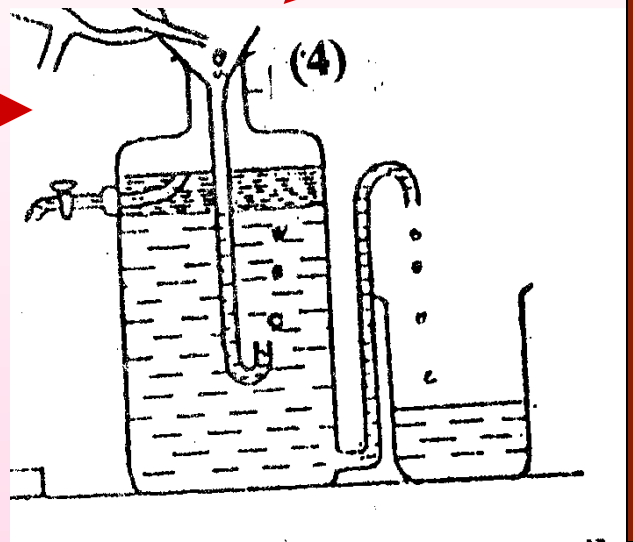
e.g oil of

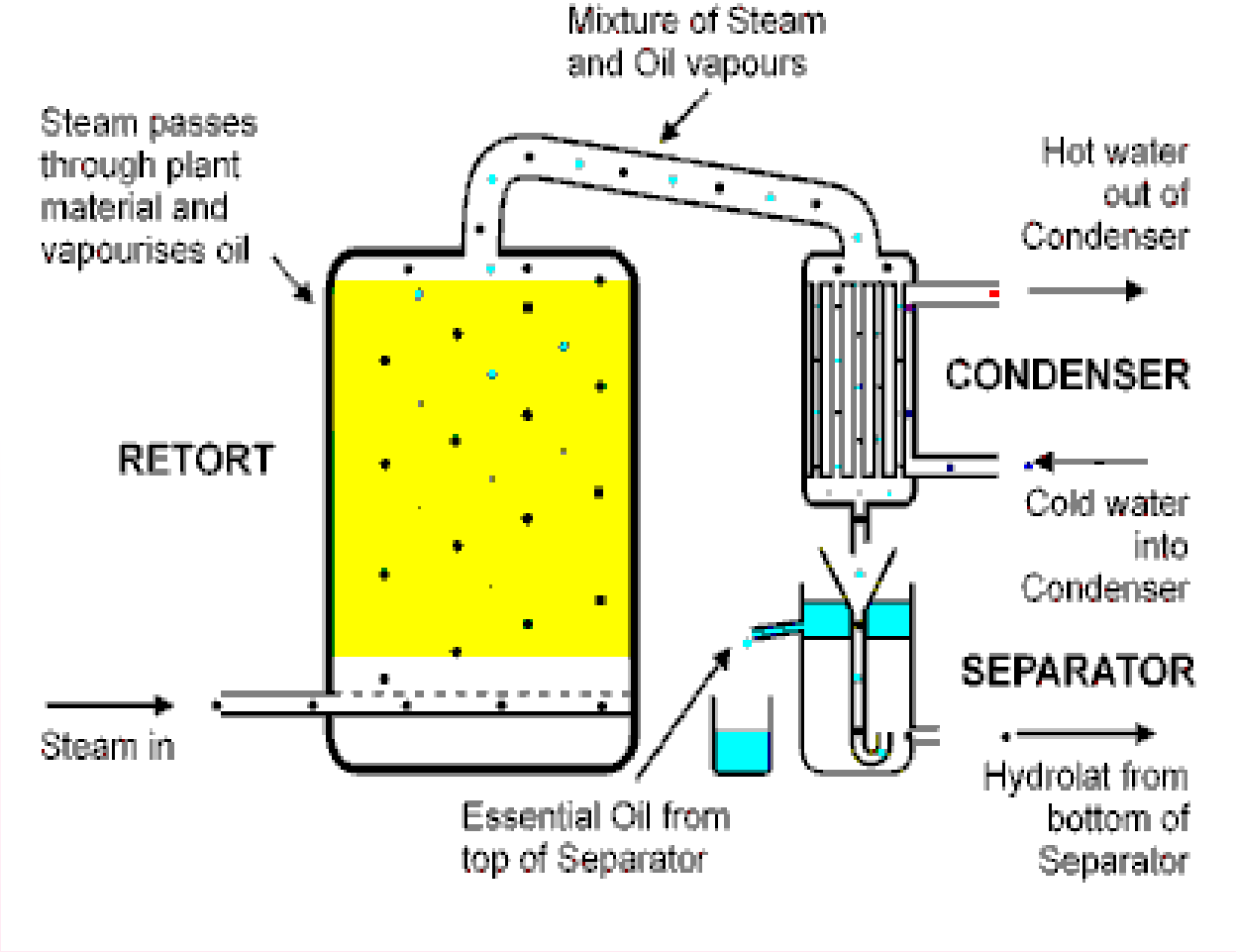


- The receiver (Florentine flask), allows the oil to col lect as a separate **upper layer** (oils lighter than wat er) or **lower layer** (oils heavier than water), while the aqueous layer saturated with oil (Aromatic wate r) is siphoned off.
- When it is automatically returned to the steam bo iler for the generation of more steam and to reco ver the dissolved oil, this is called "**Cohobation**".





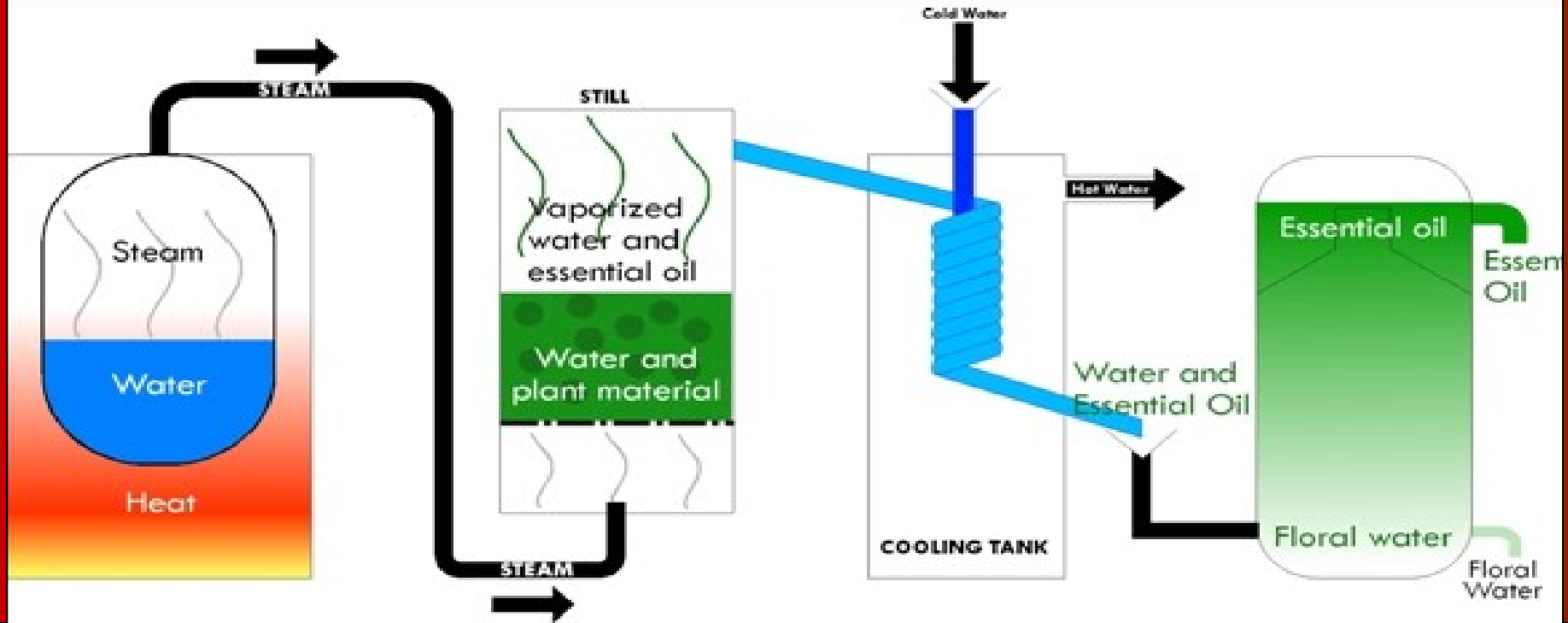


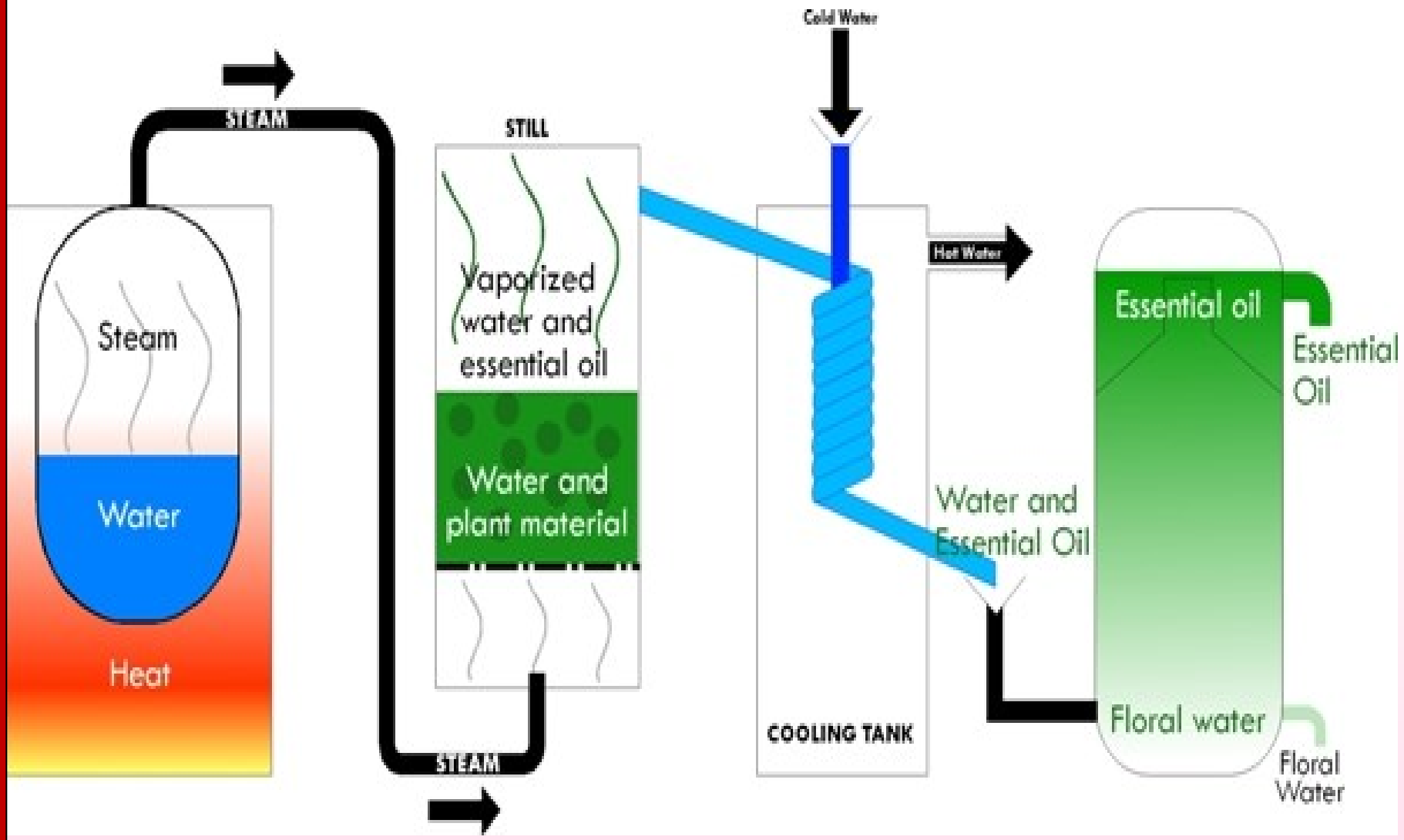


Steam distillation apparatus.

- **Water and steam distillation**

Plant material (**fresh or dried**) is immersed in water in a still, which has a heat source and **steam is fed into the mixture** (water + crushed plant).

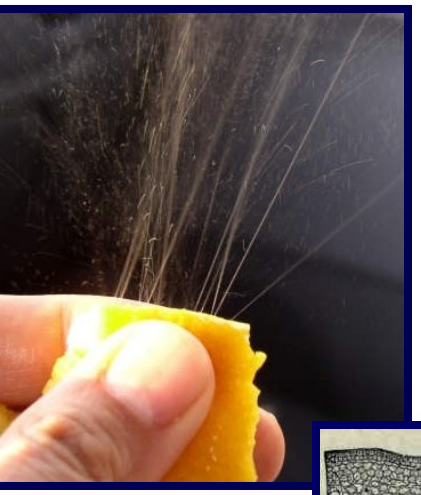


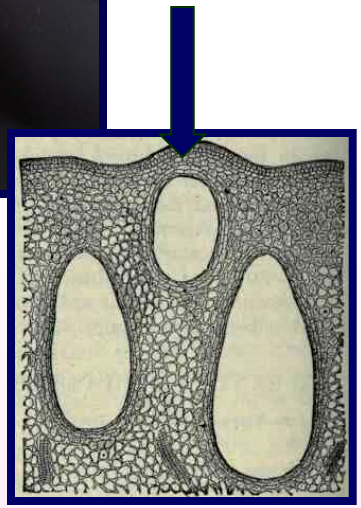

Cold Water
STEAM
STILL
Steam
Water
Heat
Vaporized water and essential oil
Water and plant material
STEAM
Hot Water
COOLING TANK
Water and Essential Oil
Essential oil
Essential Oil
Floral water
Floral Water

Steam distillation of lavender. King's Lynn, Norfolk, UK

1 pint(=0.75 ml) essential oils are obtained from ¼ ton (=225 kg) of dried lavenders by steam distillation

Scarification & Expression Methods





Principle

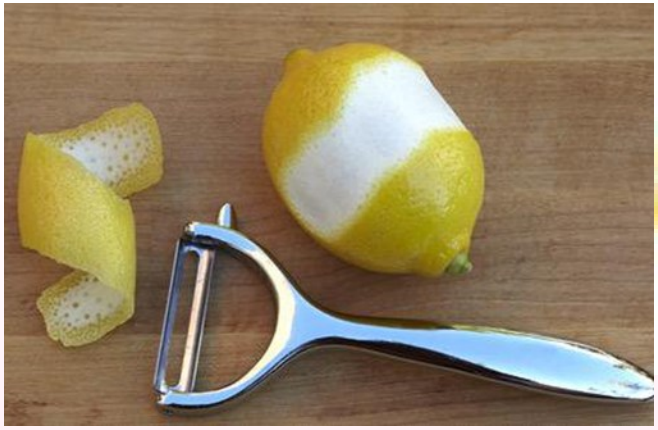
Mechanical procedures carried at room temperature & based on puncturing & squeezing of the plant material to liberate the oil, which is collected.

Applications

Preparation of **heat sensitive oils**, present in large amounts in outer peels of fruits e.g. *Citrus* fruits (Rutaceae) as orange, lemon & bergamot.

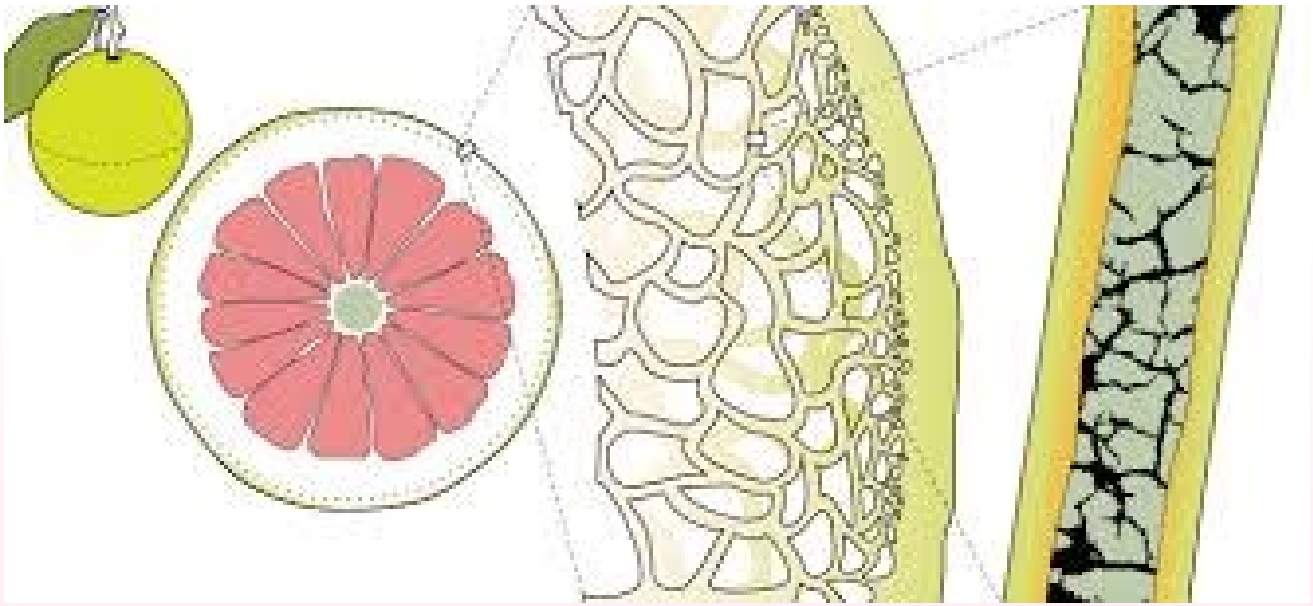
Scarification and expression

These are **cold methods of extraction**, mostly used in the extraction of citrus essential oils.



Sponge method

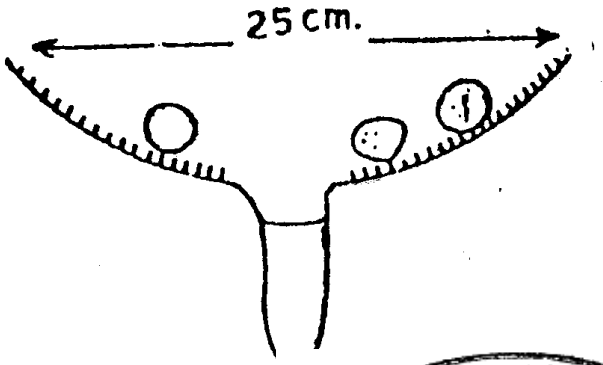
Citrus fruits are cut into halves, the juice was removed and the pericarp soaked in water, then squeezed against a sponge. The sponge i

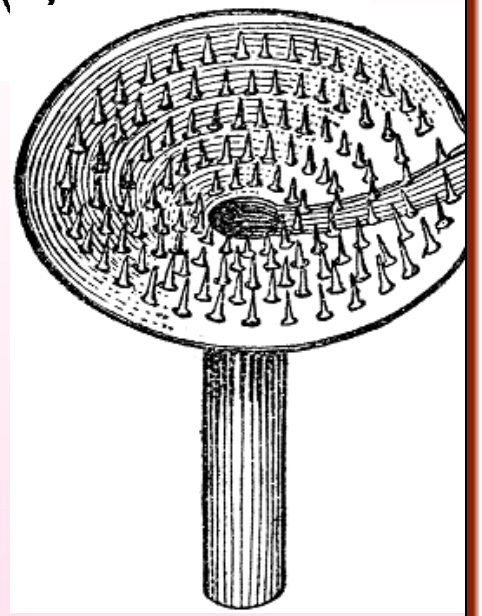


Scarification and expression

- Ecuelle a piquer

The fruits are placed in a device and rotate with puncturing the oil glands in the peel of the fruits.





- Expression of rasping

The outer region of pericarp containing oil glands is removed by a grater. The rasping is strongly pressed in horse-hair


Microplane

Extraction with solvents

For oils:

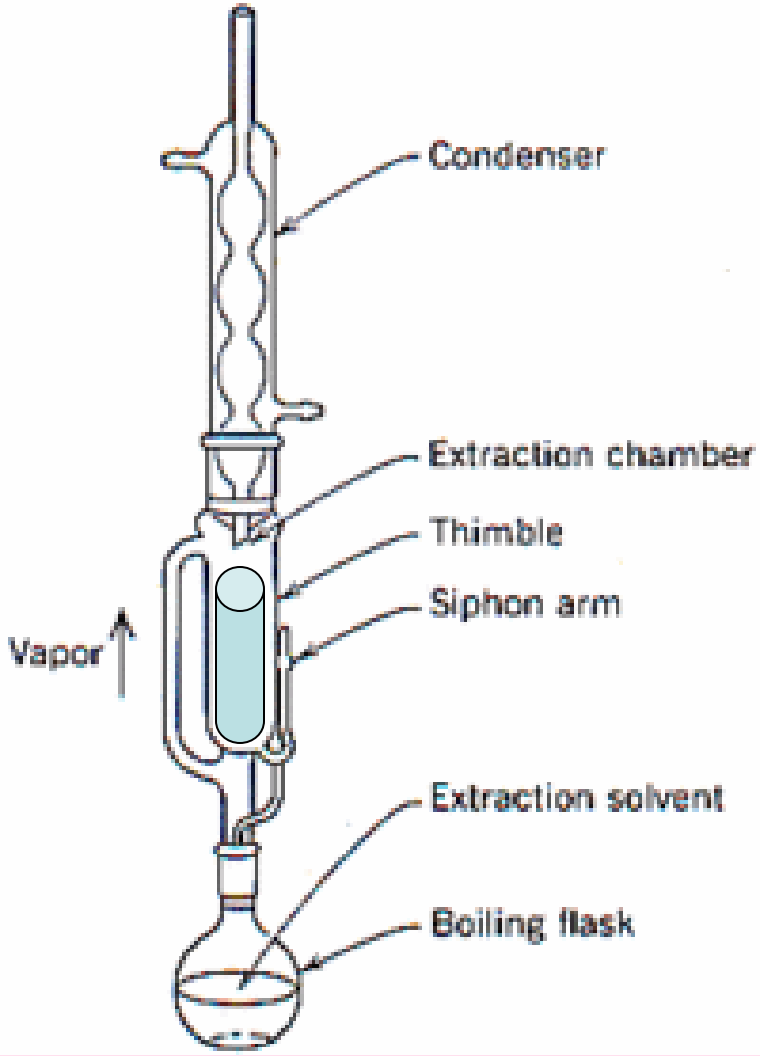
- Decompose by the action of steam.
- Present in extremely small quantities

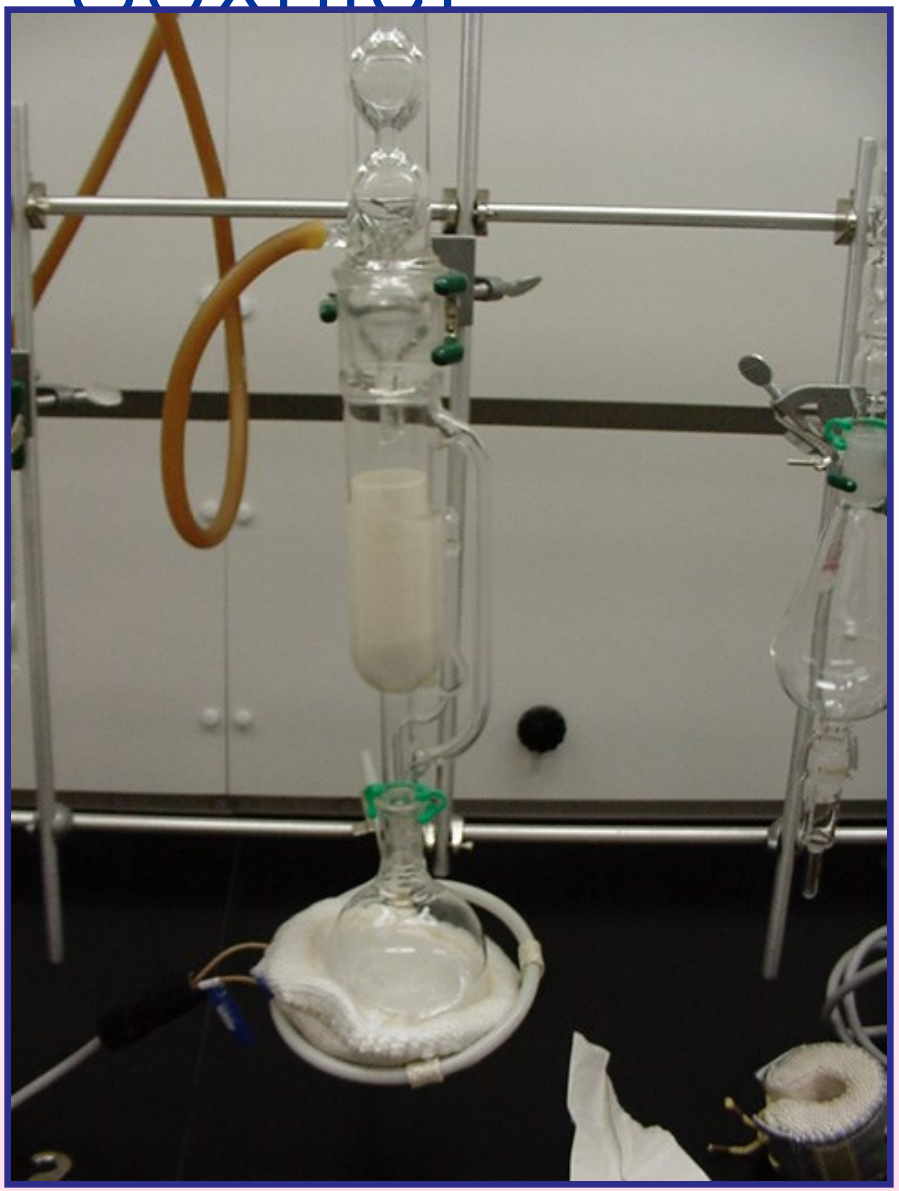
  E.g. oil of Jasmine, violet, Tuberos e……..

❖ Extraction using volatile solvents

Volatile solvents as pet. ether, hexane,.. are used by percolation or continuous extraction (Soxhlet)

Soxhlet


Condenser
Extraction chamber
Thimble
Siphon arm
Vapor
Extraction solvent
Boiling flask

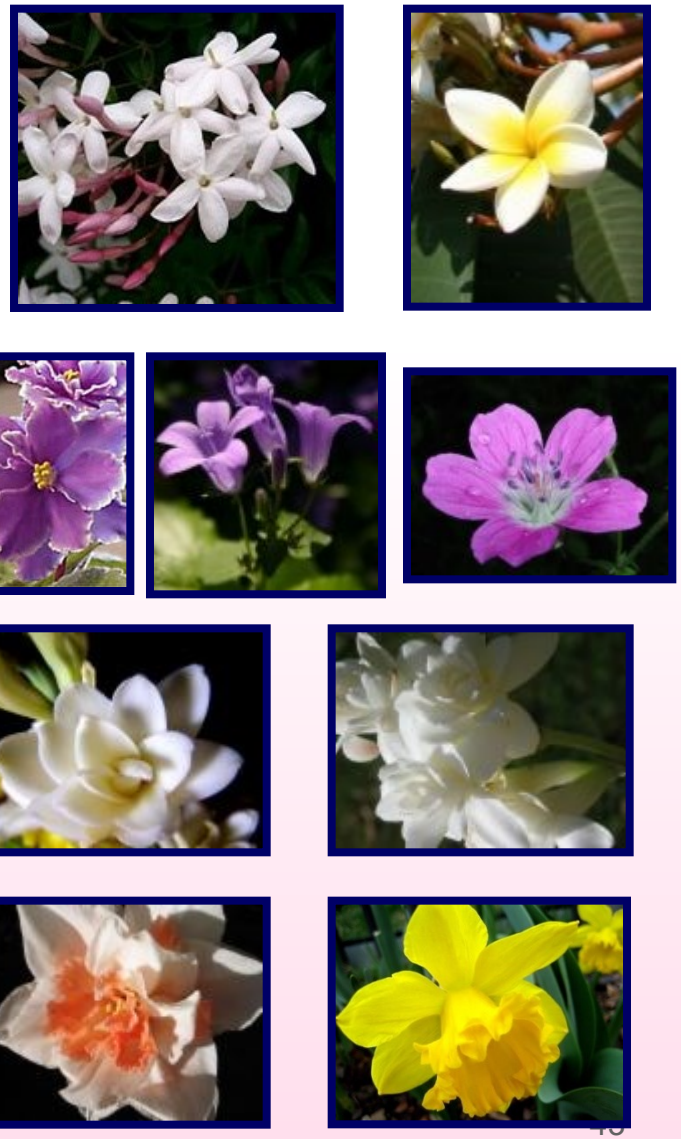


Non volatile solvent extraction methods-Application

Preparation of delicate flower oils
e.g. jasmine, violet and tuberose
which are:

1. Present in very small amounts, not easily obtained by distillation or expression

2. Oils formed of thermolabile constituents (i.e. easily decomposed by heat)



Extraction using non volatile solvents

❖Enfleurage method

Wooden frames, each enclosing a sheet of glass. Glass plates are covered with a layer of fat. The petals are spread acr oss and pressed in

The saturated fat is xtr

Enfleurage method

The oil is then recovered fro m alcohol by.

- Fractional distillation
- Evaporation under vacuum
- Dilution with water saturate d with sodium chloride.

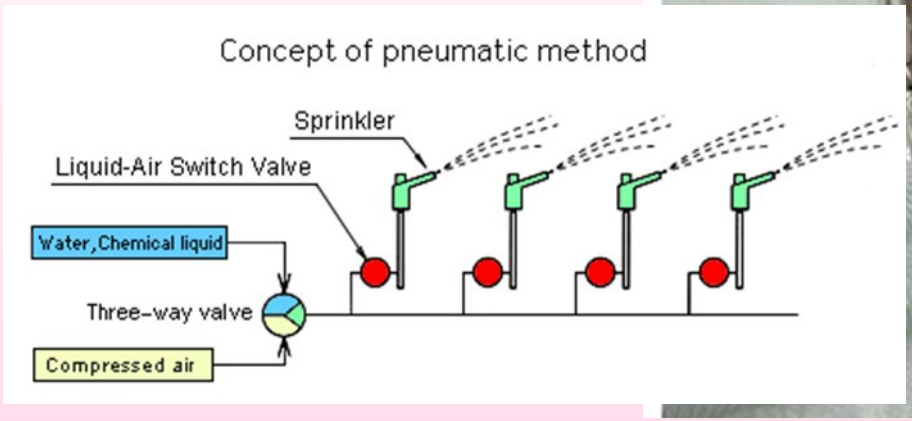
Extraction using non volatile solvents

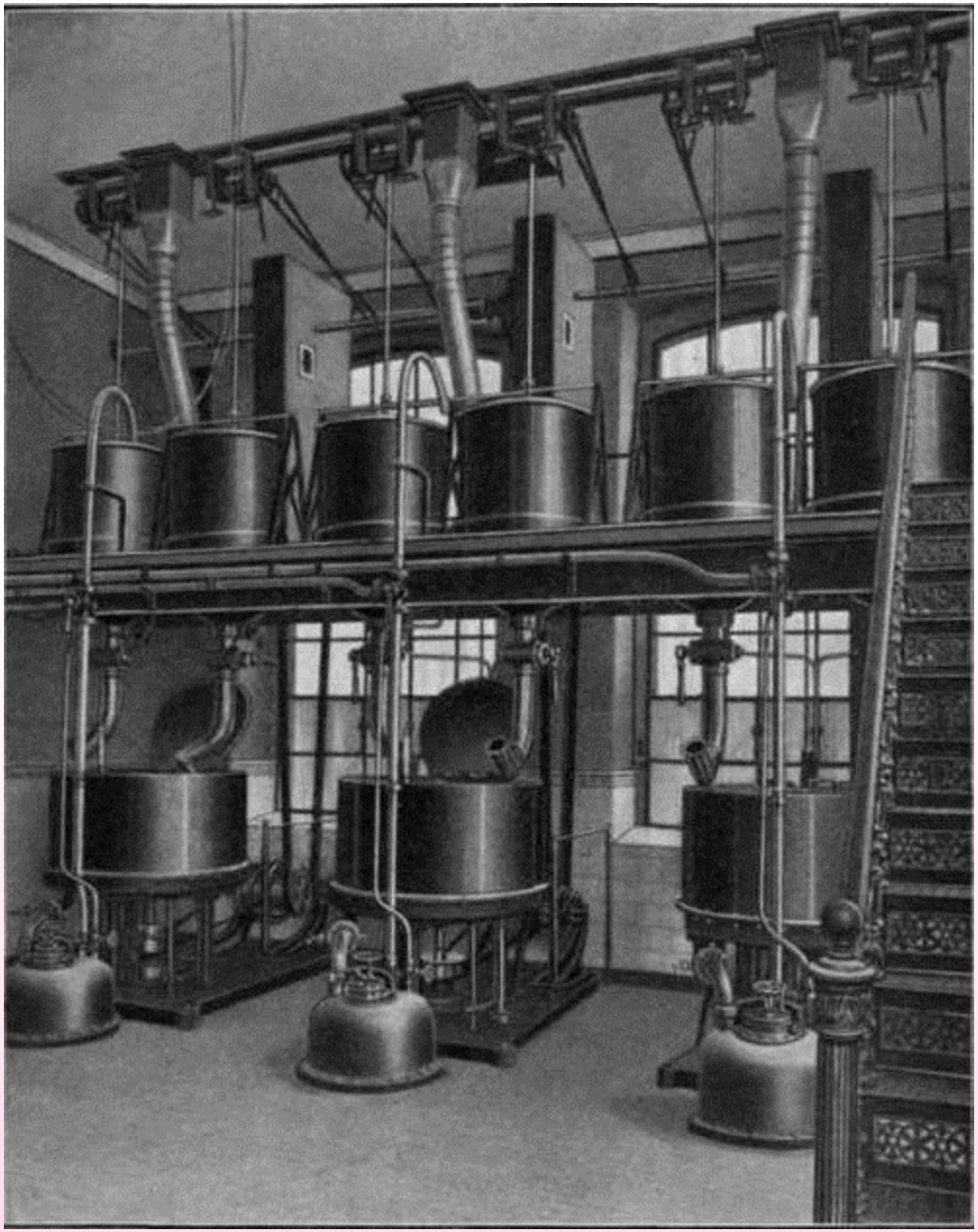
❖Pneumatic method

A current of warm air is passed through the flowers. The air laden with essential oils is passed t

melted fat ir
e oil is abs

The oil is



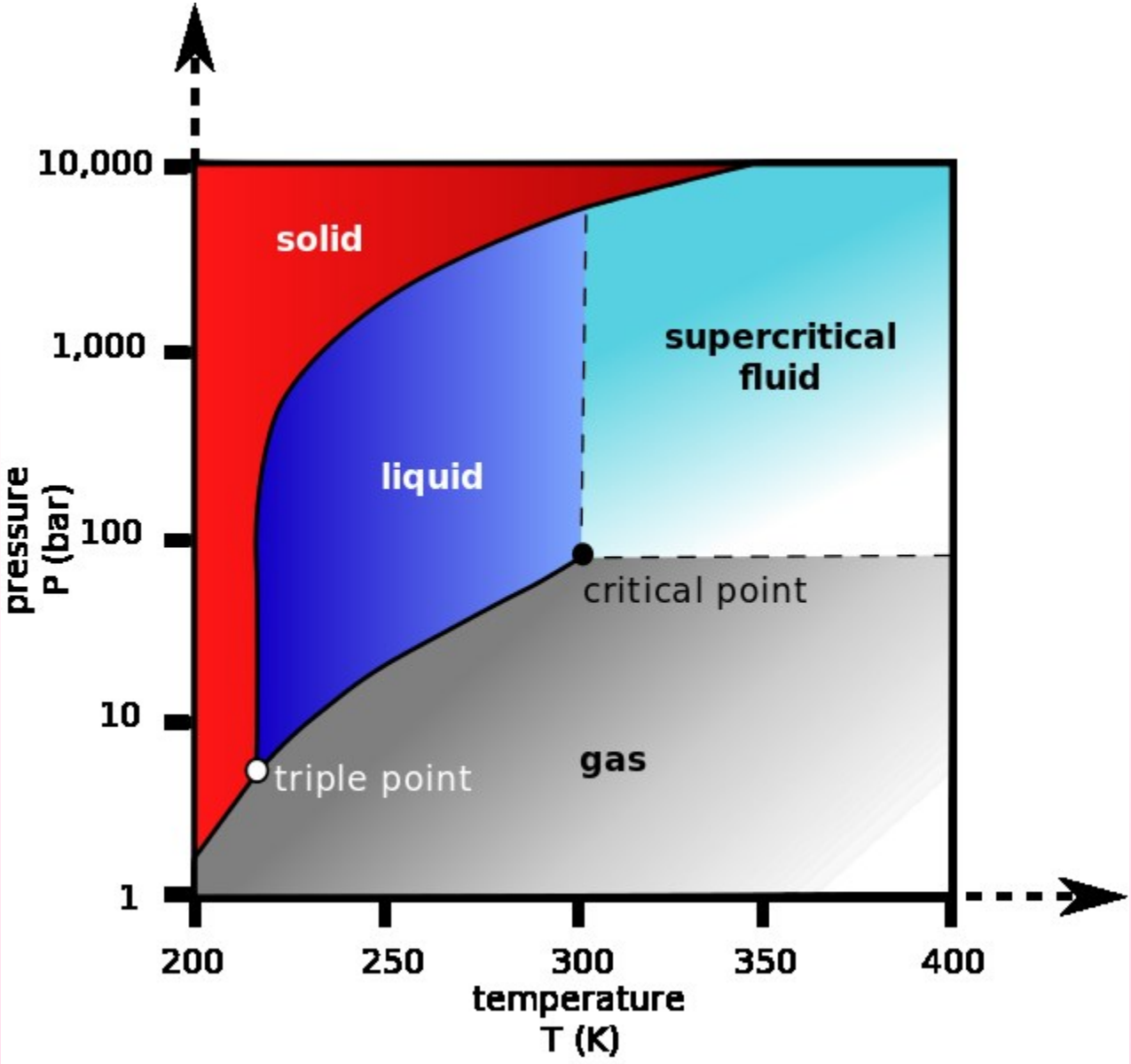


Maceration method

The flowers are soaked in hot oil to have their cell membranes ruptured, the hot oil then absorbs the essence. Exhausted flowers are removed and replaced by new ones. The oil is recovered as above

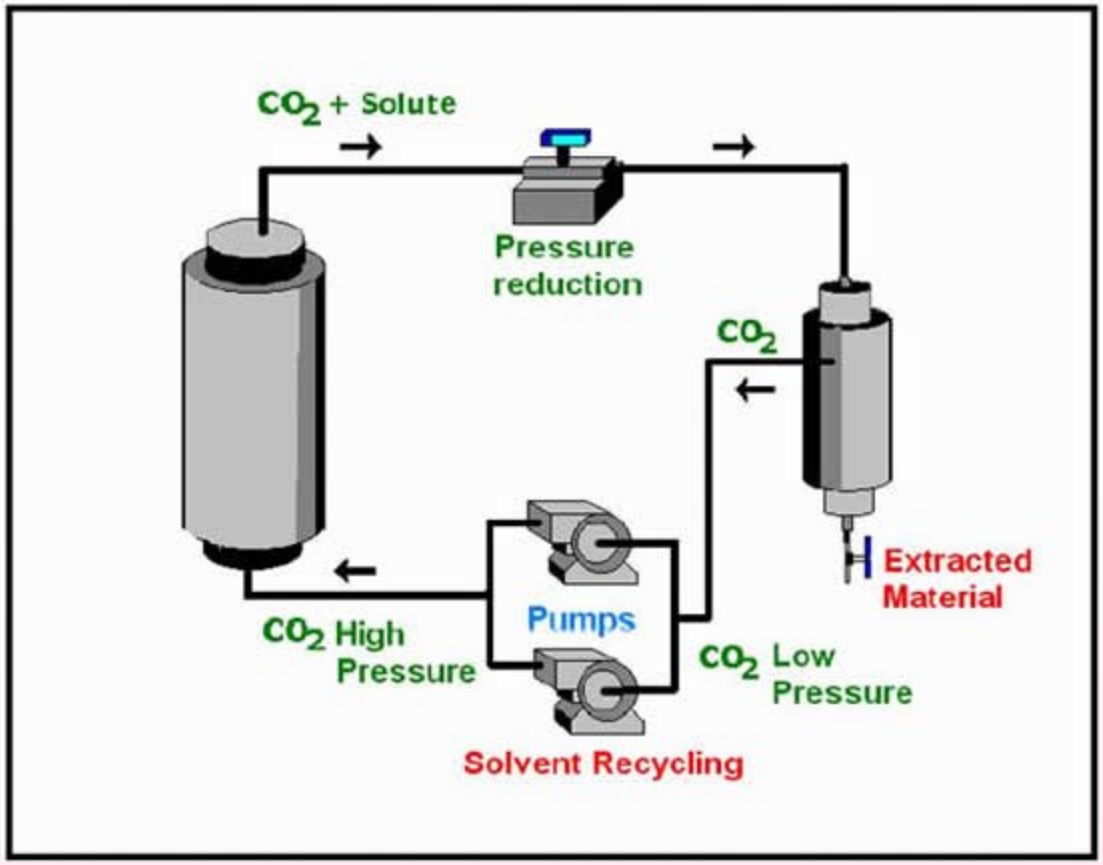
Supercritical fluid extraction

Supercritical fluid extraction



Supercritical fluid extraction

- The use of super-critical carbon dioxide extraction is a new way to extract essential oils from botanical ma terial and although a bit on the **expensive side**, doe s yield **good quality oils**
- Carbon dioxide becomes hypercritical at 33 ^{o}C, whic h is a state in which it is not really gas or liquid, but h as qualities of both, and is an **excellent solvent** to u se in the extraction of essential oils since the **low te mperature** required and the fact that the process is near to **instantaneous**.


CO_2 + Solute
Pressure reduction
CO_2
Extracted Material
Pumps
CO_2 High Pressure
CO_2 Low Pressure
Solvent Recycling

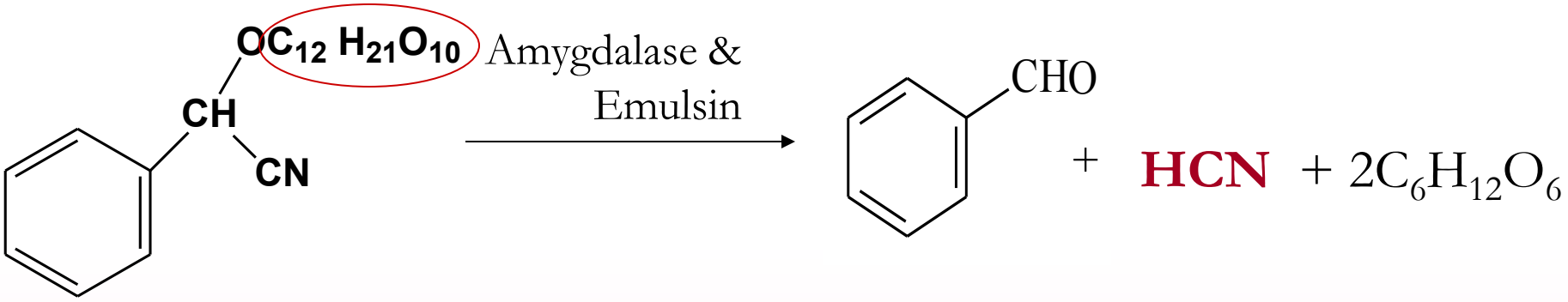

Commercial scale
scCO2 extraction plant

Enzymatic hydrolysis followed by steam distillation.

For Essential oils occur in glycosidal combination:

- Methyl salicylate (gaultherin) in wintergreen
- Eugenol (gein) in *Geum urbanum*
- Benzaldehyde (amygdalin) in bitter almond
- Allyl isothiocyanate (sinigrin) in black mustard

Preparation of essential oil of bitter alm ond



Amygdalin benzaldehyde glucose

$$\mathbf{2HCN} + Ca(OH)_2 \longrightarrow Ca(CN)_2 + H_2O$$

$$3Ca(CN)_2 + FeSO_4 \longrightarrow \mathbf{Ca_2Fe(CN)_6 + CaSO_4}$$

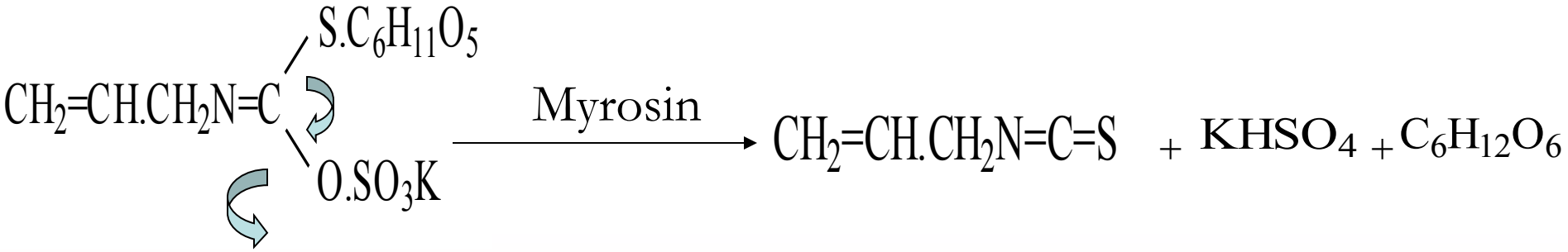
- Detection of HCN in essential oil of bitter al mond ⟶

  Oil + NaOH ⟶ aqueous layer +FeSO 4(+Fe^{3+}) ⟶

  +HCl Δ $Fe_4\{Fe$

  Prussi or (+ve HCN)

Preparation of essential oil of black mu stard



Sinigrin e

allyl isothiocyanat

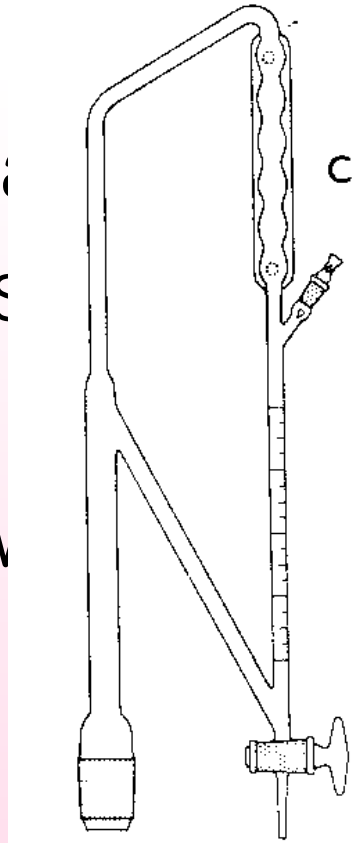
QUALITY CONTROL OF ESSENTIAL OILS

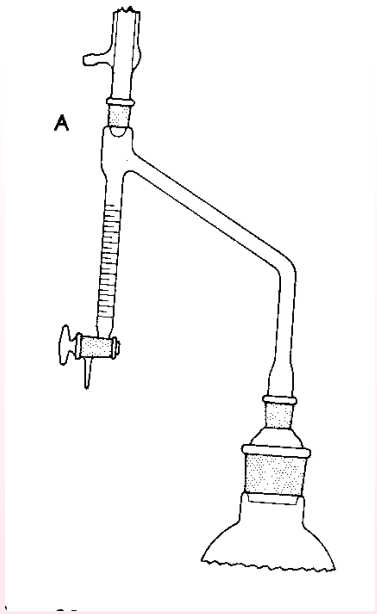
Quality control for drugs containing essential oils and quality control of essential oils

- Determination of essential oil percentage in the plant

This quantitative determina
is carried out by steam dis
in special apparatus. The
volume is read and %V/W
is calculated.

(Clavenger apparatus)





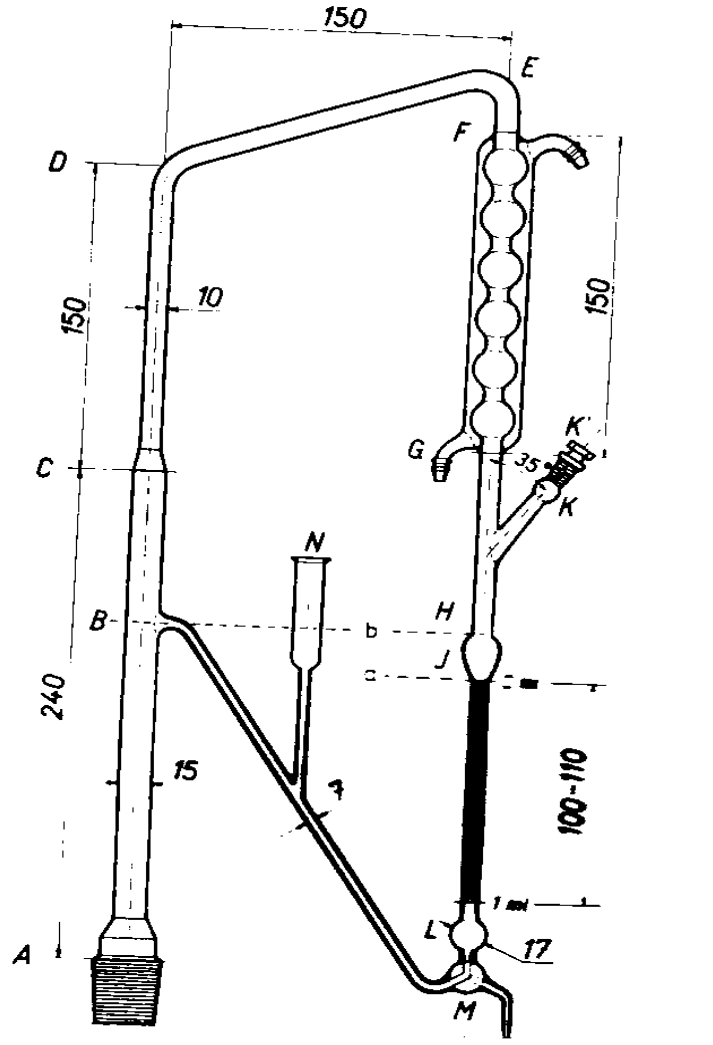
Determination of essential oil percentage in the plant

European Pharmacopoeia

Recovery of the distillate is collected in the graduated tube which already contains a known amount of xylene

Vol. of E. oil =

total vol.– vol. of xylene



Apparatus for the quantitation of essential oils in plant drugs
Dimensions in millimeters

European Pharmacopoeia 3rd edition

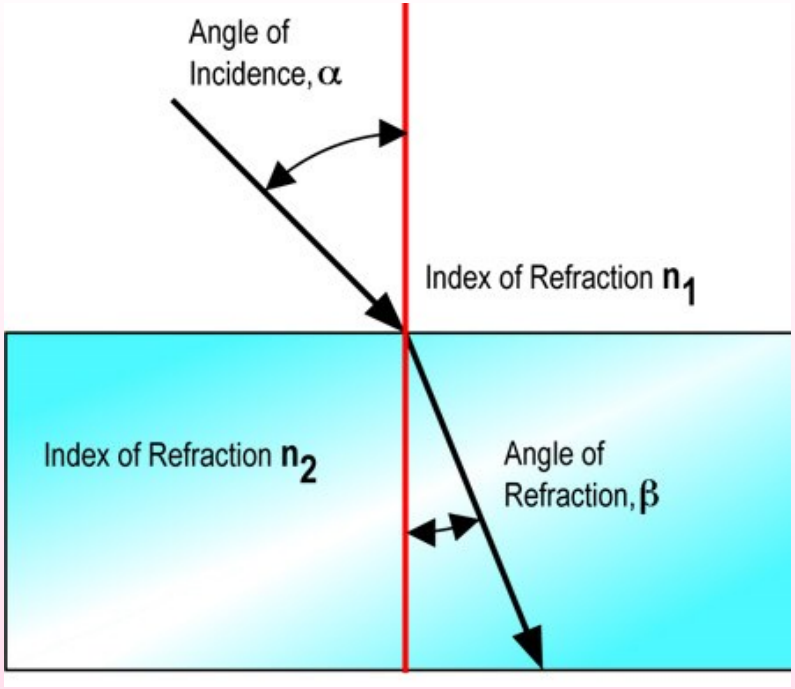
Quality control of essential oils

- Physical Examination (measurements)
  - Odor
  - Solubility
  - Weight per ml
  - Melting point
  - Optical rotation
  - Refractive index
- Analysis of the essential oils by chromatographic techniques (Gas chromatography).

Refractive index

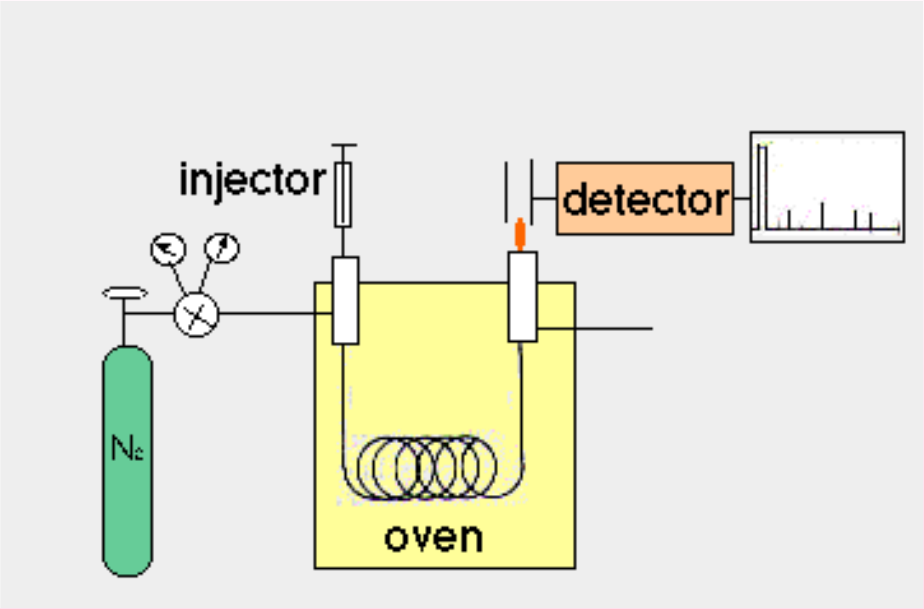
Sin of the angle of incidence = refractive index

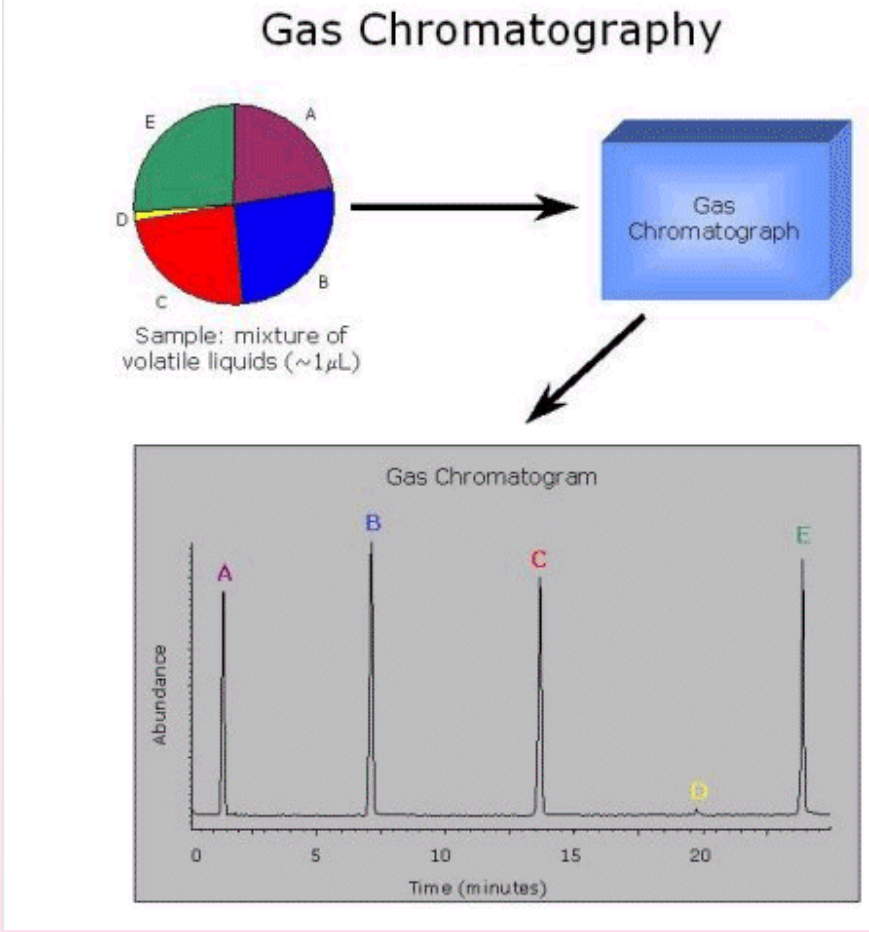
Sin of the angle of refraction



√ Gas chromatography

Is the most suitable analytical method (qu alitative and quantitative analysis).





Gas chromatography

STORAGE OF ESSENTIAL OILS

Storage of essential oils

- Storage in well-closed containers

To minimize:

- volatilization
- exposure to air
- absorption of moisture
- Various change may result from exposure to light

❑Protection of oils from light

by the use of amber- colored bottle

to prevent oxidation, hydrolysis, and polymeriza tion

❑Storage in a cool place

to minimize oxidation and volatilization.

❖Rectification of essential oils:

Purification by re-distillation in order to r emove non vol. matter and to adjust th e proportion of the constituents to the official standard.

"double-distilled" to produce oil of sta

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Thank You