PLANT LIPIDS

I. Macrmorphological tests

Ricini semen **TOXIC!**
Castor oil
Ricinus communis L. (Euphorbiaceae)

The seed is elliptical-ovoid, bean-shaped, somewhat compressed, externally it is mottled grayish and brown, varying considerably in color, smooth, with a prominent whitish caruncle.

Cucurbitae semen
Pumpkin seed
Cucurbita pepo L. (Cucurbitaceae)

The seed is covered by green pellicle. Its shape is ovoid and its upper peak is pointed.

Papaveris semen
Poppy seed
Papaver somniferum L. (Papaveraceae)

The seed of poppy plant is kidney-shaped, small, blue, grey or redish.

Lini semen Ph. Hg. VIII. Ph. Eur. szabadforgalmú
Linseed
Linum usitatissimum L. (Linaceae)

The linseed is 4 to 6 mm long, 2 to 3 mm wide, 1 to 1.5 mm thick, flat, ovate, light or dark brown, smooth, shine. Rounded on one end and acute on other.

II. PHISICAL-CHEMICAL AND CHEMICAL QUALITATIVE INVESTIGATION OF FIXED OILS

1. Determination of fixed oil content of crude drugs (oily seeds)

Cucurbitae semen (Pumpkin seed) – *Cucurbita pepo*
Helianthi annui semen (Sunflower seed) – *Helianthus annuus*
Lini semen (Linseed) – *Linum usitatissimum*
Papaveris semen (Poppy seed) – *Papaver somniferum*

Shake 5,0 g of grinded seed with 2 x 15 ml of hexane in 100 ml Erlenmeyer flask, on ultrasonic bath at room temperature for 2 x 10 minutes. Filter the solution into a round flask measured beforehand and eliminate the solvent in vacuum (Rotadest). Measure the mass of the residue and calculate the percentage (%) fixed oil content of the crude drugs.

Crude drug	Fixed oil content	Results
Cucurbitae semen	40-60%	
Helianthi annui semen	30-60%	
Lini semen	30-40%	
Papaveris semen	40-60%	

2. Identification of oils

- **Identification of different oils with TLC (thin layer chromatography)**

Layer:	Silicagel, normal phase (10 x 20 cm)	
Startpoints:	sunflower oil (diluted with chloroform)	10 µl
	linseed-oil (diluted with chloroform)	10 µl
	olive oil (diluted with chloroform)	10 µl
	castor oil (diluted with chloroform)	10 µl
	cod liver oil (diluted with chloroform)	10 µl
	<u>*Unknown oil sample*</u>	10-10 µl *(take one drop of oil with a capillar and dissolve in 1 ml chloroform)*

Developing system: Petrolether – ether – acetic acid 90 : 10 : 0,8

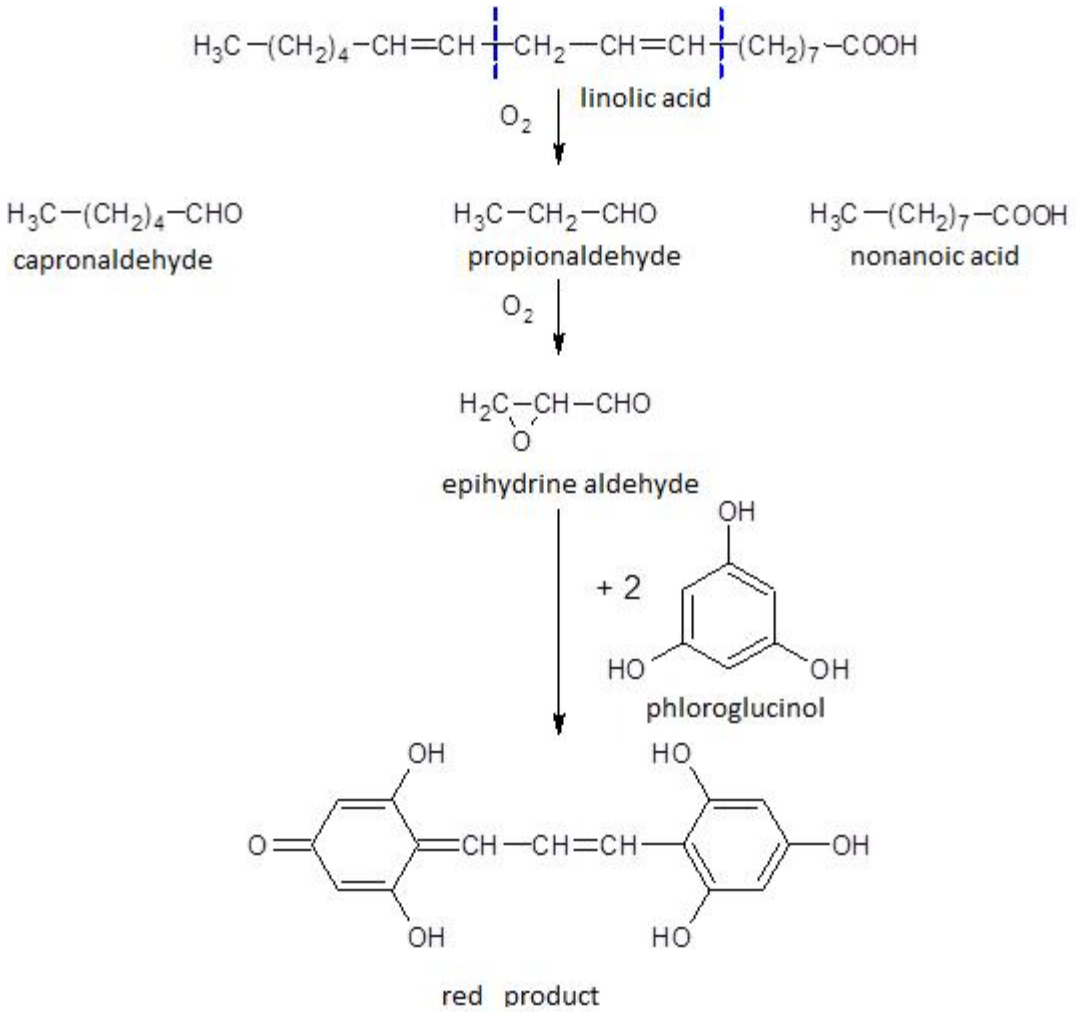
Evaluation: Put the developed layers into a glass container saturated with iodine gas and evaluate the spots. Spray with starch solution after a few minutes and evaluate again.

- **Determination of Refractive index by Abbe-refractometer**

Measure the refractive index of the oil identified with TLC according to the Ph.Eur. method with Abbe-refractometer and compare the results with the requirement of the Pharmacopoeia.

- **Investigation of rancidity**

Add 2 ml cc. HCl to the oil sample remained in the test tube (around 2 ml) under the hood. Close the throat of the test tube with cotton impregnated with 0.1% phloroglucinol/ether solution. Help the mixture of the two phases with vortex-mixer and heat the test tube with hands.



Progress of rancidness and detection

- **Determination of unsaponifiable matter of fixed oils (Ph. Eur. 2.5.7.)**

The term „unsaponifiable matter" is applied to the substances non volatile at 100-105 °C obtained by extraction with an organic solvent from the substance to be examined after it has been saponified. The result is calculated as per cent (m/m%).

Put **3 g** (*m*) of fixed oil (measure with dropper) in an Erlenmeyer flask with stopper, add **30 ml R alcohol** and **3,4 g KOH.** Heat on water bath for 1 hour with frequent shaking. Cool down and wash into a shaking funnel with **60 ml water**. Shake the aqueous extract in a separatory funnel with **3x60 ml of petroleum ether** mildly (*1-2 mild movement only, due to the risk of generating emulsion! In the case of emulsion formation, help the separation of the 2 phases with saturated NaCl solution)*. Unite the organic phases and wash (do not shake) with **3x50 ml of water** and continue the washing with **30 g/l KOH solution (25 ml).** Wash again with **5-100 ml portions of water,** until the aqueous phase will be neutral with fenoftalein indicator. Filter the petroleum ether solution on cotton covered with Na_2SO_4 sicc. into a 250 ml round flask weighed beforehand with analytical precision, wash **3 times with 5 ml petrolether.** Evaporate with Rotadest and measure the mass of the residue (*a* g, the unsaponifiable matter)

$$\text{Unsaponifiable matter} = \frac{100a}{m}\,\%$$

Investigated oils	**Requirements ofunsaponifiable matter according to Ph.Eur. (m/m%)**	**Measured (m/m%)**	**Meet or not the requirements**
Helianthi annui oleum raffinatum			
Sesami oleum raffinatum			
Iecoris aselli oleum A			
Oenotherae oleum raffinatum			

- **TLC investigation of unsaponifiable matter of fixed oils**

Dissolve the unsaponifiable, dried extract in 1 ml chloroform and investigate with TLC.

Layer:	Silicagel (normal phase)	
Startponints:	β-sitosterol/ chloroform	10 µl
	Cholesterin/ chloroform	10 µl
	Sesamin/ chloroform	10 µl
	Unsaponifiable matter/ chloroform (4)	10-10 µl

Developing system: $CHCl_3$ – diethyl-ether 9 : 1

Evaluation:

1. UV-light (254 nm)
2. Spray with 10% methanolic cc. H_2SO_4 and heat in oven (120°C) for 3 min.

Results in report:

1. Fixed oil content (m/m%)

.2. Identification and qualification of unknown oil with TLC, refractive index, rancidness

- Does it meet or not with the requirements of the Pharmacopoeia?

3. Amount of unsaponifiable matter (m/m%)

- Does it meet or not with the requirements of the Pharmacopoeia?
- Evaluation of TLC (identified components in the oil, R_f values)