

Crude drugs containing terpenophenoloids

Macroscopic investigations:

Cannabis herba

Hemp

Cannabis sativa L. (Cannabaceae)

Leaves on the lower part of the plant are digitate with five to seven folioles, whereas near the apex of the stalk only 3; the folioles are lanceolate and dentate. The smell of the drug is characteristic, the taste is bitterish.

Lupuli flos (Lupuli strobulus)

Ph. Hg. VIII.

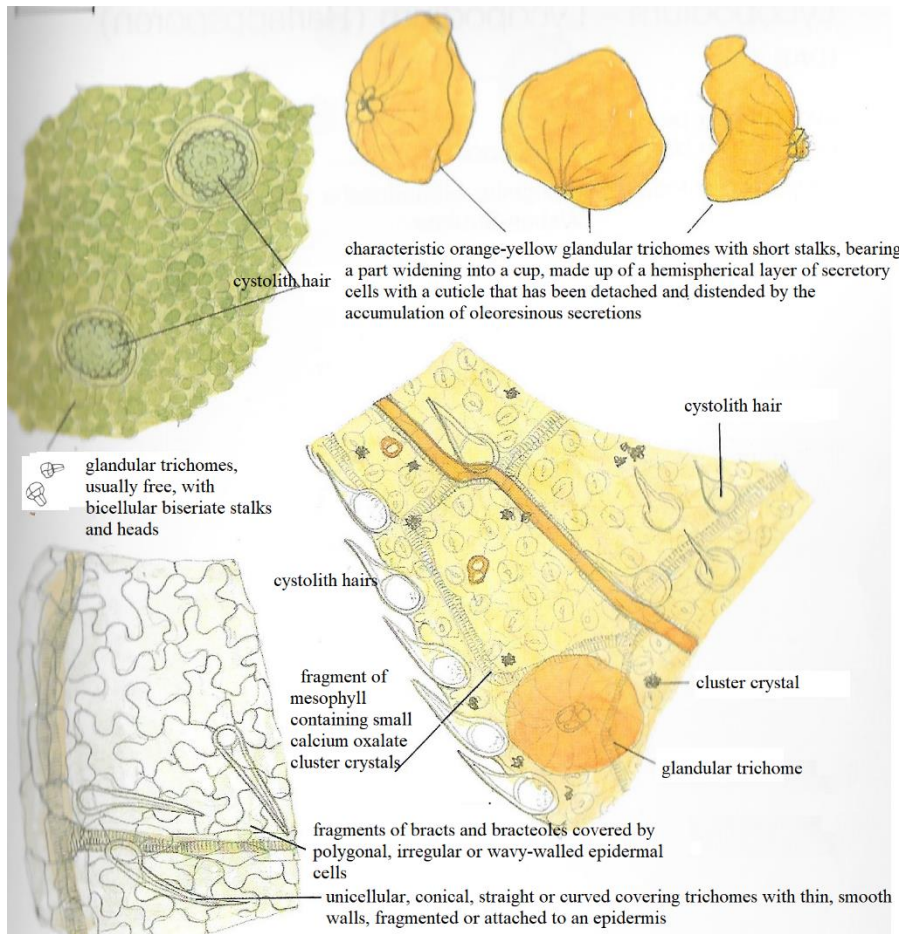
Hop strobile

Humulus lupulus L. (Cannabaceae)

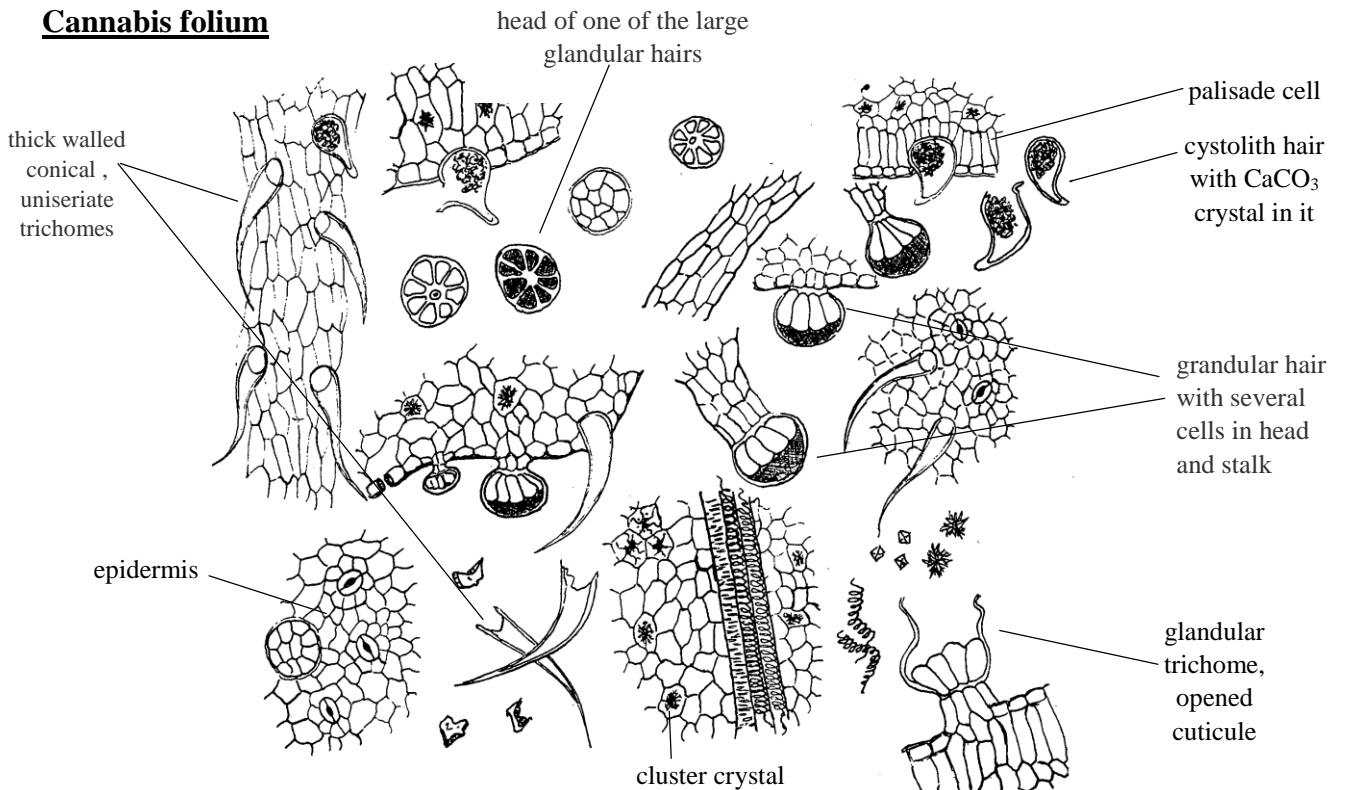
Hop strobiles are made up of many oval, greenish-yellow, sessile, membranous, overlapping bracts. Greenish-yellow flowers are at the base of the bracts. The ovary or rarely the fruit, the base of the bracts and especially the induvial fold, are covered with small orange-yellow glands. The taste is bitter.

Microscopic investigations:

Lupuli flos



Cannabis folium



1. Thin layer chromatographic investigation of *Cannabis folium*

Shake 0,3 g of powdered drug with 15 ml of petroleum ether in ultrasonic bath for 10 min, then filter the extracts (cotton) into a round flask and evaporate to 1 ml. Use 5 µl of the solution for TLC beside 2-2 µl of reference substances: 0.02% Δ⁹THC (tetrahydrocannabinol) solution and CBD (cannabidiol).

Adsorbent: Normal phase silicagel (Kieselgel 60 F254)

Developing system: n- hexane-diisopropyl-ether (5:2)

Reagent for spray: Fast blue B reagent. THC: red, CBD: orange, CBN (cannabinol): violet

2. HPLC investigation of *Cannabis indicae folium* – sample preparation

Extract 1,0 g powdered hemp with 20 ml methanol on ultrasonic bath for 20 min. Filter on paper into a round flask measured beforehand. Wash the filter paper, evaporate the solution and measure the weight of the extract. Dissolve the residue in 1.0 ml HPLC gradient grade methanol and filter on syringe filter into Eppendorf vial.

Column: RP-Select B, 5 mm

Eluent: A: triethylammonium phosphate buffer 25 mmol/l

B: acetonitrile

Isocratic separation: 36% A and 64% B,

Flow rate: 1,0 ml/min.

Injected volume: 5 µl

Detection wavelength: 210 nm

Standards: CBD (cannabidiol), CBN (cannabinol), THC (tetrahydrocannabinol), THCA (tetrahydrocannabinolic acid)

Points of the calibration line: 0,001, 0,005, 0,01, 0,02, 0,05 and 0,1 mg/ml solutions

3. Hop acids – phloroglucine derivatives – Ph.Eur. investigation

Lupuli flos, Hova tablet

- *Lupuli flos*: Extract 1 g of powdered drug with 10 ml of methanol-water (7:3) in ultrasonic bath for 10 min. and filter. Use 10 µl of the solution.
- *Lupuli flos* SFE (supercritical fluid extract) dissolved in 0,5 ml methanol; 10 µl
- *Hova tablet*: 1 tablet is powdered in a porcelaine mortar, extract with 2 ml methanol-water (7:3) for 5 min. on ultrasonic bath, filter, use 60 µl

Adsorbent. Normal phase silicagel (Kieselgel 60 F254)

Developing system: glacial acetic acid – ethyl-acetate – cyclohexane (1:19:30)

Reference: sudan-orange, curcumin, dimethylaminobenzaldehyde in methanol, 10 µl

Evaluation: UV 254 nm and 365 nm

Spray reagent: 0,2% Fast-blue reagent

4. Supercritical fluid extraction of hop strobile (SFE demonstration)

Sample weight: 0,15 g dried, powdered hop strobile

Temperature: 40°C

Pressure: 25 MPa

Extraction time: 1h

Results in the report

1. Drawings of microscopic powdered preparations
2. TLC results of Cannabis folium sample (var. ruderalis or indica)
3. HPLC investigation of Cannabis indicae folium.
4. TLC results of hop strobile (R_f values of identified components)