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Licorice

DEFINITION

Licorice consists of the roots, rhizomes, and stolons of *Glycyrrhiza glabra* L. or *Glycyrrhiza uralensis* Fish. ex DC. (Fam. Fabaceae). It contains NLT 2.5% of glycyrrhizic acid ($C_{42}H_{62}O_{16}$), calculated on the dried basis.

IDENTIFICATION

• A. THIN-LAYER CHROMATOGRAPHIC IDENTIFICATION TEST

Standard solution: 5 mg/mL of [USP Glycyrrhizic Acid RS](#) in a mixture of alcohol and water (7:3)

Sample solution: 2 g of pulverized Licorice in 10 mL of a mixture of alcohol and water (7:3). Heat by shaking on a water bath for 5 min, cool, and filter.

Chromatographic system

(See [Chromatography \(621\)](#), [Thin-Layer Chromatography](#).)

Adsorbent: 0.25-mm layer of chromatographic silica gel mixture (TLC plates)

Application volume: 2 μ L

Developing solvent system: Butyl alcohol, glacial acetic acid, and water (7:1:2)

Analysis

Samples: *Standard solution* and *Sample solution*

Develop the chromatogram in an unsaturated chamber to a length of 10 cm. Examine the plate under 254-nm light.

Acceptance criteria: The chromatograms show a dark purple zone, among other spots, due to glycyrrhizic acid at an R_f value of 0.4.

COMPOSITION

• CONTENT OF GLYCYRRHIZIC ACID

Diluent: Alcohol and water (1:1)

Solution A: Dilute acetic acid (1:15)

Mobile phase: Acetonitrile and *Solution A* (2:3)

Standard solution: 0.25 mg/mL of [USP Glycyrrhizic Acid RS](#) in *Diluent*

Sample solution: Transfer 500 mg of Licorice, reduced to a powder, to a suitable flask. Add 70 mL of *Diluent*, shake for 15 min, centrifuge, and decant the supernatant into a 100-mL volumetric flask. Mix the residue with 25 mL of *Diluent*, shake for 15 min, centrifuge, and add the supernatant to the volumetric flask. Dilute with *Diluent* to volume, and filter.

Chromatographic system

(See [Chromatography \(621\)](#), [System Suitability](#).)

Mode: LC

Detector: UV 254 nm

Column: 4.6-mm $\times$ 15-cm; packing L1

Flow rate: 0.6 mL/min

Injection volume: 20 μ L

System suitability

Sample: *Standard solution*

Suitability requirements

Column efficiency: NLT 5000 theoretical plates determined from glycyrrhizic acid

Tailing factor: NMT 2.0 for the glycyrrhizic acid peak

Relative standard deviation: NMT 2.0%

Analysis

Samples: *Standard solution* and *Sample solution*

Calculate the percentage of glycyrrhizic acid ($C_{42}H_{62}O_{16}$) in the portion of Licorice taken:

Result = $(r_U/r_S) \times C_S \times (V/W) \times 100$

- r_U = peak area of glycyrrhizic acid from the *Sample solution*
- r_S = peak area of glycyrrhizic acid from the *Standard solution*
- C_S = concentration of [USP Glycyrrhizic Acid RS](#) in the *Standard solution* (mg/mL)
- V = final volume of the *Sample solution* (mL)
- W = weight of Licorice taken to prepare the *Sample solution* (mg)

Acceptance criteria: NLT 2.5% on the dried basis

CONTAMINANTS

- [ARTICLES OF BOTANICAL ORIGIN, Limits of Elemental Impurities \(561\)](#): Meets the requirements
- [ARTICLES OF BOTANICAL ORIGIN, Pesticide Residue Analysis \(561\)](#): Meets the requirements

SPECIFIC TESTS

• BOTANICAL CHARACTERISTICS

Macroscopic: The terrestrial stem is nearly cylindrical, 0.5–3.0 cm in diameter, and over 1 m in length; it is externally dark brown to red-brown and longitudinally wrinkled. It often has lenticels, small buds, and scaly leaves. The transverse section reveals a rather clear border between the phloem and the xylem, and a radial structure that often has radiating splits.

Microscopic: The transverse section reveals several yellow-brown cork layers, and a layer of phelloderm that is 1–3 cells thick. The cortex exhibits medullary rays, and obliterated sieve portions radiate alternately. The phloem exhibits groups of phloem fibers, which are surrounded by crystal cells, with thick but incompletely lignified walls. The vessels are accompanied by xylem fibers, which are surrounded by crystal cells, and by xylem parenchyma cells. The parenchyma cells contain starch grains, and often contain single crystals of calcium oxalate.

- [ARTICLES OF BOTANICAL ORIGIN, Foreign Organic Matter \(561\)](#): NMT 2.0%
- [ARTICLES OF BOTANICAL ORIGIN, Alcohol-Soluble Extractives, Method 2 \(561\)](#): NLT 25.0%
- [LOSS ON DRYING \(731\)](#): Dry a sample at 105° for 6 h; it loses NMT 12.0% of its weight.
- [ARTICLES OF BOTANICAL ORIGIN, Total Ash \(561\)](#): NMT 7.0%
- [ARTICLES OF BOTANICAL ORIGIN, Acid-Insoluble Ash \(561\)](#): NMT 2.0%

ADDITIONAL REQUIREMENTS

- **PACKAGING AND STORAGE:** Preserve in well-closed containers. Store in a cool, dry place.
- **LABELING:** The label states the Latin binomial and, following the official name, the part of the plant contained in the article.
- [USP REFERENCE STANDARDS \(11\)](#)
[USP Glycyrrhizic Acid RS](#)

Auxiliary Information - Please [check for your question in the FAQs](#) before contacting USP.

Topic/Question	Contact	Expert Committee
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