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White Peony Root

DEFINITION

White Peony Root consists of the dried, peeled roots of *Paeonia lactiflora* Pall. (Family *Paeoniaceae*). The roots are collected in summer or autumn and boiled in water, either before or after peeling, with the two ends and rootlets removed, then sun dried. It contains NLT 3.0% of total benzoyl monoterpene glycosides calculated as the sum of albiflorin ($C_{23}H_{28}O_{11}$), paeoniflorin ($C_{23}H_{28}O_{11}$), and benzoylpaeoniflorin ($C_{30}H_{32}O_{12}$) on the dried basis.

IDENTIFICATION

• A. [HPTLC FOR ARTICLES OF BOTANICAL ORIGIN \(203\)](#)

Standard solution A: 0.25 mg/mL of [USP Paeoniflorin RS](#) in [methanol](#)

Standard solution B: 30 mg/mL of [USP Paeonia lactiflora Peeled Root Dry Extract RS](#) in [methanol](#). Sonicate for 15 min, centrifuge, and use the supernatant.

Sample solution: 500 mg of finely powdered White Peony Root in 5 mL of [methanol](#). Sonicate for 15 min, centrifuge, and use the supernatant.

Chromatographic system

Adsorbent: [Silica gel with fluorescence indicator F₂₅₄](#), precoated plates, for HPTLC

Application volume: 8 µL as 8-mm bands

Relative humidity: Condition the plate to a relative humidity of about 33% using a suitable device.

Developing solvent system: [Methylene chloride](#), [ethyl acetate](#), [methanol](#), and [formic acid](#) (35:5:5:3)

Derivatization reagent: 10% [sulfuric acid](#) in alcohol. [NOTE—Prepare on the day of the test; it can be used for up to 1 week if stored in a tightly closed container in a cool place. Keep alcohol cold over ice, and add sulfuric acid slowly.]

Analysis

Samples: *Standard solution A*, *Standard solution B*, and *Sample solution*

Apply the *Samples* as bands and dry in air. Develop in a saturated chamber, remove the plate from the chamber, and dry in air. Treat the plate with *Derivatization reagent*, and heat at 100° for 5 min. Immediately examine under long wavelength UV light and under white light.

System suitability

Samples: *Standard solution A* and *Standard solution B*

Suitability requirements

Under long wavelength UV light: *Standard solution A* exhibits a dark brown band due to paeoniflorin in the lower-third of the chromatogram. *Standard solution B* exhibits a band corresponding in R_f and color to the band due to paeoniflorin in *Standard solution A*, a blue band due to albiflorin below the paeoniflorin band, and a dark band due to gallic acid above the paeoniflorin band. In the upper half of the chromatogram, *Standard solution B* exhibits bands in the following order with increasing R_f : a pinkish-violet band, a light dark band, a narrow violet band, a pinkish-violet band, and a broad purple band.

Under white light: *Standard solution A* exhibits a brown band due to paeoniflorin in the lower third of the chromatogram. *Standard solution B* exhibits a band corresponding in R_f and color to the band due to paeoniflorin in *Standard solution A*, a brown band due to albiflorin below the paeoniflorin band, and a brown band due to gallic acid above the paeoniflorin band. In the upper half of the chromatogram, *Standard solution B* exhibits brown bands in the following order with increasing R_f : three well-separated bands, a purple brown band, and a reddish brown band.

Acceptance criteria

Under long wavelength UV light: The *Sample solution* exhibits the following bands corresponding to similar bands in *Standard solution B*. In the lower third of the chromatogram, the *Sample solution* exhibits a dark brown band corresponding in R_f and color to the band due to paeoniflorin in *Standard solution A*; a blue band due to albiflorin below the paeoniflorin band (a distinction from the bark of *Paeonia × suffruticosa*, the unpeeled root of *Paeonia lactiflora*, and the root of *Paeonia anomala* subsp. *veitchii*); and a dark band due to gallic acid.

above the paeoniflorin band. In the upper half of the chromatogram, the *Sample solution* exhibits several bands in the following order with increasing R_F : a pinkish-violet band, a light dark band, a narrow violet band, a pinkish-violet band, and a broad purple band.

Under white light: The *Sample solution* exhibits the following bands corresponding to similar bands in *Standard solution B*. In the lower third of the chromatogram, the *Sample solution* exhibits a brown band corresponding in R_F and color to the band due to paeoniflorin in *Standard solution A*; a brown band due to albiflorin below the paeoniflorin band (a distinction from the bark of *Paeonia* × *suffruticosa*, the unpeeled root of *Paeonia lactiflora*, and the root of *Paeonia anomala* subsp. *veitchii*); and a brown band due to gallic acid above the paeoniflorin band. In the upper half of the chromatogram, the *Sample solution* exhibits several brown bands in the following order with increasing R_F : three well-separated bands, a purple brown band, and a reddish brown band.

• **B. HPLC**

Analysis: Proceed as directed in the test for *Content of Benzoyl Monoterpenoid Glycosides*.

Acceptance criteria: The *Sample solution* exhibits the most-intense peak at the retention time corresponding to paeoniflorin in *Standard solution A* and *Standard solution B*. The *Sample solution* also exhibits peaks due to albiflorin (a distinction from the bark of *Paeonia* × *suffruticosa* and the root of *Paeonia anomala* subsp. *veitchii*) and benzoylpaeoniflorin at the retention times corresponding to the same constituents in *Standard solution B*. No significant peak corresponding to paeonol appears after the benzoylpaeoniflorin peak (a distinction from the bark of *Paeonia* × *suffruticosa* and the unpeeled root of *Paeonia lactiflora*). No peak corresponding to paeoniflorin sulfonate is observed at a relative retention time of about 0.45 with respect to paeoniflorin (a distinction from sulfur-fumigated White Peony Root). The content ratios of albiflorin and benzoylpaeoniflorin relative to paeoniflorin are within the ranges in [Table 2](#).

COMPOSITION

• **CONTENT OF BENZOYL MONOTERPENOID GLYCOSIDES**

Solution A: 0.10% [phosphoric acid](#) in [water](#)

Solution B: [Acetonitrile](#)

Mobile phase: See [Table 1](#).

Table 1

Time (min)	Solution A (%)	Solution B (%)
0	87	13
4	86	14
7	84	16
15	79	21
25	36	64
25.1	5	95
30	5	95
30.1	87	13
35	87	13

Solvent: [Methanol](#) and [water](#) (1:1)

Standard solution A: 0.12 mg/mL of [USP Paeoniflorin RS](#) in *Solvent*

Standard solution B: 2 mg/mL of [USP Paeonia lactiflora Peeled Root Dry Extract RS](#) in *Solvent*. Sonicate for 10 min, centrifuge, and pass through a suitable membrane filter of 0.45-µm or finer pore size.

Sample solution: Transfer about 40 mg of finely powdered White Peony Root, accurately weighed, into a suitable flask. Add 10.0 mL of *Solvent*, and close tightly. Weigh the filled flask accurately, and sonicate for 30 min. Cool to room temperature, and adjust to the initial weight by adding *Solvent*, if needed. Before injection, pass through a suitable membrane filter of 0.45-µm or finer pore size, and discard the first portion of the filtrate.

Chromatographic system

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