

Identification Saponify 5 g of Peanut Oil by boiling with 2.5 mL of sodium hydroxide solution (3 in 10) and 12.5 mL of ethanol (95). Evaporate the ethanol, dissolve the residue in 50 mL of hot water, and add dilute hydrochloric acid in excess until the free fatty acids separate as an oily layer. Cool the mixture, remove the separated fatty acids, and dissolve them in 75 mL of diethyl ether. To the diethyl ether solution add a solution of 4 g of lead (II) acetate trihydrate in 40 mL of ethanol (95), and allow the mixture to stand for 18 hours. Filter the supernatant liquid, transfer the precipitate to the filter with the aid of diethyl ether, and filter by suction. Place the precipitate in a beaker, heat it with 40 mL of dilute hydrochloric acid and 20 mL of water until the oily layer is entirely clear, cool, and decant the water layer. Boil the fatty acids with 50 mL of diluted hydrochloric acid (1 in 100). When the solution prepared by dissolving 0.1 g of the fatty acids in 10 mL of ethanol (95) is not darkened by the addition of 2 drops of sodium sulfide TS, allow the fatty acids to solidify, and press them between dry filter papers to exclude moisture. Dissolve the solid fatty acid in 25 mL of diluted ethanol (9 in 10) with the aid of gentle heat, and then cool to 15°C to crystallize the fatty acids. Recrystallize them from diluted ethanol (9 in 10) and dry in a desiccator (phosphorus (V) oxide, in vacuum) for 4 hours: the melting point of the dried crystals is between 73°C and 76°C.

Acid value Not more than 0.2.

Saponification value 188 – 196

Unsaponifiable matters Not more than 1.5%.

Iodine value 84 – 103

Containers and storage Containers—Tight containers.

Peony Root

Paeoniae Radix

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Peony Root is the root of *Paeonia lactiflora* Pallas (*Paeoniaceae*).

It contains not less than 2.0% of peoniflorin, calculated on the dried basis.

Description Cylindrical root, 7–20 cm in length, 1–2.5 cm in diameter; externally brown to light grayish brown, with distinct longitudinal wrinkles, with warty scars of lateral roots, and with laterally elongated lenticels; fractured surface dense in texture, light grayish brown, and with light brown radial lines in xylem. Odor, characteristic; taste, slightly sweet at first, followed by an astringency and a slight bitterness.

Identification (1) Shake 0.5 g of pulverized Peony Root with 30 mL of ethanol (95) for 15 minutes, and filter. Shake 3 mL of the filtrate with 1 drop of iron (III) chloride TS: a blue-purple to blue-green color is produced, and it changes to dark blue-purple to dark green.

(2) To 2 g of pulverized Peony Root add 10 mL of methanol, warm on a water bath for 5 minutes, cool, filter, and use the filtrate as the sample solution. Separately, dis-

solve 1 mg of paeoniflorin for thin-layer chromatography in 1 mL of methanol, and use this solution as the standard solution. Perform the test with these solutions as directed under the Thin-layer Chromatography. Spot 10 μ L each of the sample solution and the standard solution on a plate of silica gel for thin-layer chromatography. Develop the plate with a mixture of acetone, ethyl acetate and acetic acid (100) (10:10:1) to a distance of about 10 cm, and air-dry the plate. Spray evenly 4-methoxybenzaldehyde-sulfuric acid TS upon the plate, and heat at 105°C for 5 minutes: one spot among the spots from the sample solution and the purple-red spot from the standard solution show the same color tone and the same Rf value.

Loss on drying Not more than 14.0% (6 hours).

Total ash Not more than 6.5%.

Acid-insoluble ash Not more than 0.5%.

Assay Weigh accurately about 0.5 g of pulverized Peony Root, add 50 mL of diluted methanol (1 in 2), heat under a reflux condenser on a water bath for 30 minutes, cool, and filter. To the residue add 50 mL of diluted methanol (1 in 2), and proceed in the same manner. Combine the filtrates, add diluted methanol (1 in 2) to make exactly 100 mL, and use this solution as the sample solution. Separately, weigh accurately about 0.01 g of Paeoniflorin Reference Standard, dissolve in diluted methanol (1 in 2) to make exactly 100 mL, and use this solution as the standard solution. Pipet 20 μ L each of the sample solution and the standard solution, and perform the test as directed under the Liquid Chromatography according to the following conditions. Determine the peak areas, A_T and A_S , of paeoniflorin in each solution.

$$\begin{aligned} & \text{Amount (mg) of paeoniflorin (C}_{23}\text{H}_{28}\text{O}_{11}) \\ &= \text{amount (mg) of Paeoniflorin Reference Standard,} \\ & \quad \text{calculated on the anhydrous basis} \\ & \quad \times \frac{A_T}{A_S} \end{aligned}$$

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 230 nm).

Column: A stainless steel column 4 to 6 mm in inside diameter and 15 to 25 cm in length, packed with octadecylsilanized silica gel (5 to 10 μ m in diameter).

Column temperature: a constant temperature of about 20°C.

Mobile phase: A mixture of water and acetonitrile (4:1).

Flow rate: Adjust the flow rate so that the retention time of paeoniflorin is about 10 minutes.

Selection of column: Dissolve 1 mg each of Paeoniflorin Reference Standard and *p*-hydroxyacetophenone in diluted methanol (1 in 2) to make 10 mL. Perform the test with 20 μ L of this solution under the above operating conditions. Use a column giving elution of paeoniflorin and *p*-hydroxyacetophenone in this order with the resolution between these peaks being not less than 3.

System repeatability: Repeat the test five times with the standard solution under the above operating conditions: the relative standard deviation of the peak areas of paeoniflorin is not more than 1.5%.