

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 280 nm).

Column: A stainless steel column about 4 mm in inside diameter and about 15 cm in length, packed with octadecyl-silicized silica gel for liquid chromatography (5 μ m in particle diameter).

Column temperature: A constant temperature of about 25°C.

Mobile phase: A mixture of water and methanol (51:49).

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

Selection of column: Proceed with 10 μ L of the standard solution under the above operating conditions, and calculate the resolution. Use a column giving elution of estriol and the internal standard in this order with the resolution between these peaks being not less than 8.

Containers and storage Containers—Tight containers.

Estriol Injection (Aqueous Suspension)

エストリオール水性懸濁注射液

Estriol Injection (Aqueous Suspension) is an aqueous suspension for injection. It contains not less than 90% and not more than 110% of the labeled amount of estriol ($C_{18}H_{24}O_3$: 288.38).

Method of preparation Prepare as directed under Injections, with Estriol.

Description Shake Estriol Injection (Aqueous Suspension): a white turbidity is produced.

Identification (1) Shake well, take a volume of Estriol Injection (Aqueous Suspension), equivalent to 2 mg of Estriol according to the labeled amount, add ethanol (95) to make 20 mL, and use this solution as the sample solution. Proceed with the sample solution as directed in the Identification (1) under Estriol.

(2) Determine the absorption spectrum of the sample solution obtained in (1) as directed under the Ultraviolet-visible Spectrophotometry: it exhibits a maximum between 279 nm and 283 nm.

Assay Shake well, pipet a volume of Estriol Injection (Aqueous Suspension), equivalent to about 5 mg of estriol ($C_{18}H_{24}O_3$), and dissolve in methanol to make exactly 20 mL. Pipet 4 mL of this solution, add exactly 5 mL of the internal standard solution, then add methanol to make 50 mL, and use this solution as the sample solution. Separately, weigh accurately about 0.025 g of Estriol Reference Standard, previously dried at 105°C for 3 hours, and dissolve in methanol to make exactly 100 mL. Pipet 4 mL of this solution, add exactly 5 mL of the internal standard solution, then add methanol to make 50 mL, and use this solution as the standard solution. Proceed as directed in the Assay under Estriol.

$$\begin{aligned} & \text{Amount (mg) of estriol (C}_{18}\text{H}_{24}\text{O}_3\text{)} \\ & = \text{amount (mg) of Estriol Reference Standard} \end{aligned}$$

$$\times \frac{Q_T}{Q_S} \times \frac{1}{5}$$

Internal standard solution—A solution of methyl benzoate for estriol limit test in ethanol (95) (1 in 5000).

Containers and storage Containers—Hermetic containers.

Estriol Tablets

エストリオール錠

Estriol Tablets contain not less than 90% and not more than 110% of the labeled amount of estriol ($C_{18}H_{24}O_3$: 288.38).

Method of preparation Prepare as directed under Tablets, with Estriol.

Identification (1) Weigh a portion of powdered Estriol Tablets, equivalent to 2 mg of Estriol according to the labeled amount, add ethanol (95) to make 20 mL, shake for 10 minutes, centrifuge, and use the supernatant liquid as the sample solution. Proceed with the sample solution as directed in the Identification (1) under Estriol.

(2) Determine the absorption spectrum of the sample solution obtained in (1) as directed under the Ultraviolet-visible Spectrophotometry: it exhibits a maximum between 279 nm and 283 nm.

Content uniformity To one tablet of Estriol Tablets add exactly 5 mL of water, disperse the fine particles with ultrasonic wave, add exactly 15 mL of methanol, and shake for 15 minutes. Centrifuge this solution for 10 minutes, pipet a definite amount of the supernatant liquid, and add methanol to make exactly a definite amount of solution so that each mL of the solution contains about 5 μ g of estriol ($C_{18}H_{24}O_3$). Pipet 5 mL of this solution, add exactly 1 mL of the internal standard solution, and use this solution as the sample solution. Proceed with 20 μ L of the sample solution as directed in the Assay under Estriol. Use a solution of methyl benzoate in methanol (1 in 40,000) as the internal standard solution. Calculate the mean value from each ratio of peak areas of 10 samples: the samples conform to the requirements if the deviation (%) of the mean value and each ratio of peak areas is within 15%. If the deviation (%) exceeds 15%, and 1 sample shows deviation within 25%, repeat the test with 20 samples. Calculate the deviation (%) of the mean value from each ratio of peak areas of the 30 samples used in the 2 tests and each ratio of peak areas: the samples conform to the requirements if the deviation exceeds 15%, not more than 1 sample shows deviation within 25%, and no sample shows deviation exceeding 25%.

Dissolution test Perform the test with 1 tablet of Estriol Tablets at 50 revolutions per minute according to Method 2 under the Dissolution Test, using 900 mL of water as the test solution. Take 20 mL or more of the dissolved solution 30 minutes after starting the test, and filter through a membrane filter with pore size of not more than 0.8 μ m. Discard the first 10 mL of the filtrate, pipet the subsequent V mL, add water to make exactly V' mL so that each mL contains about 0.1 μ g of estriol ($C_{18}H_{24}O_3$) according to the labeled