Test item	JP	EP	USP
Version	XVII (2016)	10	
Molecular formula	C20H24O2	C20H24O2	C20H24O2
CAS number	57-63-6	57-63-6	57-63-6
Molecular weight	296.40	296.4	296.40
Substance name	19-Nor-17α-pregna-1,3,6(10)-trien-20-yne-3,17-diol	19-Nor-17α-pregna-1,3,6(10)-trien-20-yne-3,17-diol	19-Nor-17α-pregna-1,3,6(10)-trien-20-yne-3,17-diol
Content	Not less than 98.0 %	97.5 - 102.0 % (dried substance)	97.0 % - 102.0 %
Appearance(形状)	White to pale yellow crystals or crystalline powder. It is odorless.	white or slightly yellowish-white, crystalline powder	
Solubility(溶解度)	It is freely solubel in pridine and in tetrahydrofulan, soluble in ethanol (95) and in diethyl ether, and practically insoluvle in water. Dissolves in sodium hydoroxide TS.	practically insoluvle in water, freely soluble in ethanol (96 %). It dissolves in dilute alkaline solutions. It shows polymorphism.	
Identification (1)	<spec> 1) A purplish red color develops with a yellow-green fluorescence. 2) The color of the solution changes to red-purple. <test method> 1) Dissolve 2 mg of Ethinylestradiol in 1 mL of a mixture of ethanol (95) and sulfuric acid (1:1). 2) Add carefully 2 mL of water to this solution.		
Identification (2)	<spec> The precipitate melts between 200 °C and 202 °C. <test method> Transfer 0.02 g of Ethinylestradiol to a glass-stoppered test tube, dissolve in 10 mL of a solution of potassium hydroxide (1 in 20), add 0.1 g of benzoyl chloride, and shake. Collect the resulting precipitate, recrystallize from methanol, and dry in a desiccator (in vacuum, phosphorus (V) oxide).		
Identification (IR)	Not specified.	<spec> must comply <test method> Comparison: ethynilestradiol CRS If different, dissolve the substance in methonal R, evaporate to dryness and record IR using the residues.	197K
Identification (TLC)		<spec> the prinsipal spot obtained with the test solution is simlar in position, colour, flourescence and size to the principal spot in the chromatogram obtained with the reference solution. <test method> Solvent mixture: Methanol R, methylene chloride R (10:90 V/V) test solution: 25mg/25mL solvent mixture ref solution; 25 mg CRS/25mL solvent mixture plate; TLC silicagel Movile phase: ethanol (96%), toluene (10:90 V/V) Application: 5 microL Development: over 2.3 of plate Detection: heat at 100 degree for 10 min, spray with alcoholic solution of sulfuric acid, heat again. Examine in day light and in UV at 365 nm.	
Identification (UV)			<spec> absorptivities, calculated on dried basis, not differ by more than 3.0 % 197 U Sample solution: 50 microg/mL in alcohol Wevelength: 281nm
Optical rotation	<spec> [α]20, D: -26 to - 31 ° <test method> as per JP <2.49> after drying, 0.1 g, pyridine, 25mL, 100 mm		<spec> 50 mg/mL, using sonication if necessary, in colorless pridine from freshly opened container. - 28.0 to -29.5° <test method> Prepare a solution with 0.050 g/mL (0.0475-0.0525 g/mL) test substance by dissolving at (20 ± 0.5) °C in pyridine in a volumetric flask. Light source Sodium lamp, 589.3 nm Temperature 20 ± 0.5 °C Light path 1 dm
Melting point/Melting range	<spec> melting point. 180 to 186 °C or 142 to 146 °C		<spec> melting range. 180 to 186 °C or 142 to 146 °C
Clarity of solution			clear and free rom undisolved solid (100mg in 5 mL of alcohol)
Purity	<spec>		
Estrone	The solution has no more color than the following control solution. <test method> Dissolve 5 mg of Ethinylestradiol in 0.5 mL of ethanol (95), and add 0.05 g of 1,3-dinitrobenzene. Add 0.5 mL of freshly prepared dilute potassium hydroxide-ethanol TS, allow to stand in a dark place for 1 hour, and add 10 mL of ethanol (95). Control solution: Proceed in the same manner as mentioned above, omitting Ethinylestradiol.		
Related substances (HPLC)	Not specified.	<spec> Impurity B : max 0.5% Impurity H: max 0.2% Impurity I: max 0.2% Impurity K: max 0.2% Impurity C: max 0.15 %	

		Impurity F: max 0.15 % Any unspecified impurity max. 0.10 % Total impurities max. 0.8 % disregard limit: 0.05% <test method> Test procedure solvent mixture:water,acetonitrile (40:60 V/V) test solution 50.0mg/30mL acetonitrile and dilute to 50.0 mL with water reference solution a dilute 1.0mL test soluion with 100.0mL solvent mixture, dilute 1.0mL of the solution/10.0 mL with solvent mixture Reference solution B (blank/solvent mixture) 2mg estrone (impurity C) in 10.0 ml with the solvent mixrue. 1.0 mL to dissolve the contents of EE for systemsuitability CRS Reference solution c 50.0 mg EE in 30ml of acetonitril and dilute to 10.0 ml with water. Test conditions Volume injected 30 µL Detector UV-Detector at 220 nm Column: length 0.25 m, internal diameter 4.6 mm Stationary phase End-capped butylsilyl silica gel, 5 µm Flow rate 1.5 mL/minute Mobile phase Gradient A: Acetonitril, Water (30:70 V/V) B: Water, acetonitrile (25:75 V/V) Time [minutes]: A% (v/v)/B%(v/v) 0-35 :100/0 35-65: 100→0/0→100 Column temperature 30 °C Substance: Correction factor Impurity I: 0.4 Impurity B: 0.7 Relative retention to EE (35min): Impruity F=0.2, Impurity H=0.5, Impruity I=0.8, Impurity B=0.88, Imruity C=0.92, Impurity K= 1.3 System suitability test Resolution: min 1.2 between impurity I/impurity B in solution b	
Loss on drying	<spec> Not more than 0.5%. <test method> 0.5g, in vacum, phosphours (V) oxide, 5 hours	<spec> max. 1.0 % <test method> 0.500g, 105°C, 3 h	NMT 1.0 %, 105°C, 3 h
Sulfated ash/residue on ignition	<spec> Not more than 0.1% <test method> 0.5 g		
Assay (potentiometric titration)	<test method> Weigh accurately about 0.2 g of Ethinylestradiol, previously dried, and dissolve in 40 mL of tetrahydrofuran. Add 10 mL of a solution of silver nitrate (1 in 20), and titrate with 0.1 mol/L sodium hydroxide VS. Each mL of 0.1 mol/L sodium hydroxide VS = 29.64 mg of C20H24O2		
Assay (HPLC)		<test method> Same as Related substances (HPLC). referemce solution c	mobile phase: acetonitrile and water (1:1) internal standard solution: 0.5mg/mL of ethylparaben in mobile phase standard solution: 0.2 mg/mL of USP EE RS in mobile phase. 10 mg USP EE RS to a 50 ml volumentric flask, add 10 ml of moble phase and 5. 0 mL internal standard solution. Dilue with mobile phase to volume. Sample stock solution: 1.0 mg/mL of EE in mbile phase Sample solution 0/2 mg/mL of EE. LC system UV 280 nm Column: 4.6 mm x 15 cm; packing L1 Flow rate: 1mL/min Inje. 25 micro L System suitability: sample: standard solution Relative retention times for ethylparaben and EE: 0.6 and 1.0 Suitability requirements: Reolution: NLT 4.5 btwn ethyl paraben and EE peaks RSD: NMT 2.0 % Formula:
Containers	Tight containers		tight, non metallic
Strage	light resistant	protected from light	light-resistant