Egualen Sodium Granules

Dissolution <*6.10*> Weigh accurately an amount of Egualen Sodium Granules, equivalent to about 5 mg of egualen sodium ($C_{15}H_{17}NaO_3S$·1/3 H2O) according to the labeled amount, and perform the test at 75 revolutions per minute according to the Paddle method, using 900 mL of 2nd fluid for dissolution test as the dissolution medium. Start the test, withdraw not less than 20 mL of the medium at the specified minute after starting the test, and filter through a membrane filter with a pore size not exceeding 0.45 μm. Discard the first 10 mL of the filtrate, and use the subsequent filtrate as the sample solution. Separately, weigh accurately about 22 mg of Egualen Sodium RS (separately, determine the water <*2.48*> with 0.5 g by direct titration in volumetric titration), and dissolve in 2nd fluid for dissolution test to make exactly 100 mL. Pipet 5 mL of this solution, add 2nd fluid for dissolution test to make exactly 200 mL, and use this solution as the standard solution. Determine the absorbances, A_T and A_S, at 284 nm of the sample solution and standard solution as directed under Ultraviolet-visible Sectrophotometry <*2.24*>.

The requirements are met if Egualen Sodium Granules conform to the dissolution requirements.

Dissolution rate (%) with respect to the labeled amount of egualen sodium ($C_{15}H_{17}NaO_3S$·1/3 H_2O)

$= M_S/M_T \times A_T/A_S \times 1/C \times 45/2 \times 1.020$

M_S: Amount (mg) of Egualen Sodium RS, calculated on the anhydrous basis

M_T: Amount (g) of sample

C: Labeled amount (mg) of egualen sodium ($C_{15}H_{17}NaO_3$ S·1/3 H_2O) in 1 g

Dissolution Requirements

Labeled amount	Specified minute	Dissolution rate
25 mg/g	15 minutes	Not less than 85%

Egualen Sodium RS $C_{15}H_{17}NaO_3$ S·1/3 H_2O: 306.35 Sodium 3-ethyl-7-isopropyl-1-azulenesulfonate 1/3 hydrate. It meets the following requirements. Purify according to the following method if needed.

Purification method−Dissolve 10 g of egualen sodium in 30 mL of ethanol (99.5) by warming, and filter while warm. After cooling, collect the separated crystals, and wash with two 2-mL portions of ethanol (99.5). Recrystalize in ethanol (99.5), and wash the crystals so obtained with two 5-mL

portions of ethanol (99.5). Dry the crystals so obtained at 80ºC for 2 hours, and allow to cool in a desiccator with silica gel.

Description−Egualen Sodium RS occurs as blue, crystals or crystalline powder.

Identification (1) Determine the absorption spectrum of a solution of Egualen Sodium RS (1 in 4000) as directed under Ultraviolet-visible Spectrophotometry <*2.24*>: it exhibits a maximum between 580 nm and 584 nm. Also, determine the absorption spectrum of a solution of Egualen Sodium RS (1 in 200000) as directed under Ultraviolet-visible Spectrophotometry <*2.24*>: it exhibits maxima between 237 nm and 241 nm, between 283 nm and 287 nm, and between 293 nm and 297 nm.

(2) Determine the infrared absorption spectrum of Egualen Sodium RS, previously dried, as directed in the potassium bromide disk method under Infrared Spectrophotometry <*2.25*>: it exhibits absorption at the wave numbers of about 2950 cm^{-1}, 1576 cm^{-1}, 1385 cm^{-1}, 1179 cm^{-1} and 1047 cm^{-1}.

(3) Determine the ^{1}H spectrum of a solution of Egualen Sodium RS in deuterated methanol for nuclear magnetic resonance spectroscopy (1 in 50), using tetramethylsilane for nuclear magnetic resonance spectroscopy as an internal reference compound, as directed under Nuclear Magnetic Resonance Spectroscopy <*2.21*>: it exhibits a multiple signal A at around δ 1.4 ppm, a broad multiple signal B at around δ 3.0 ppm, a triplet signal C at around δ 7.2 ppm, a double signal D at around δ 7.7 ppm, a single signal E at around δ 8.0 ppm, a double signal F at around δ 8.3 ppm, and a single or a slightly splitting double signal G at around δ 9.2 ppm. The ratio of integrated intensity of each signal, A:B:C:D:E:F:G, is about 9:3:1:1:1:1:1.

Purity (1) 1-ethyl-5-isopropylazulene and 1,3-diethyl-5-isopropylazulene−Dissolve 20 mg of Egualen Sodium RS in methanol to make exactly 100 mL, and use this solution as the sample solution. Separately, dissolve 10 mg each of 1-ethyl-5-isopropylazulene and 1,3-diethyl-5-isopropylazulene in methanol to make them exactly 100 mL. Pipet 1 mL each of these solutions, add methanol to make them exactly 50 mL. Pipet 1 mL each of these solutions, add methanol to make them exactly 50 mL, and use these solutions as the standard solution (1) and standard solution (2). Perform the test with exactly 20 μL each of the sample solution, standard solution (1) and standard solution (2) as directed under Liquid Chromatography <*2.01*> according to the following conditions, and determine each peak area by the automatic integration method: the peak area of 1-ethyl-5-isopropylazulene obtained from the sample solution is not larger than that from the standard solution (1), and the peak area of 1,3-diethyl-5-isopropylazulene obtained from the sample solution is not larger than that from the standard solution (2).

Operating conditions

Detector: An ultraviolet absorption photometer (wavelength: 285 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 20ºC.

Mobile phase: Dissolve 2.7 g of potassium dihydrogen phosphate in water to make 1000 mL, and adjust the pH to 6.0 with a solution prepared by dissolving 7.2 g of disodium hydrogen phosphate dodecahydrate in water to make 1000 mL. To 100 mL of this solution add 400 mL of acetonitrile.

Flow rate: Adjust the flow rate so that the retention time of 1-ethyl-5-isopropylazulene is about 9 minutes.

Time span of measurement: About 2 times as long as the retention time of 1-ethyl-5-isopropylazulene.

System suitability

System performance: Dissolve 10 mg each of 1-ethyl-5-isopropylazulene and 1,3-diethyl-5-isopropylazulene in methanol to make 100 mL. To 1 mL each of these solutions add methanol to make 50 mL. To 1 mL of this solution add methanol to make 50 mL, and use this solution as the solution for system suitability test. When the procedure is run with 20 μL of the solution for system suitability test under the above operating conditions, 1-ethyl-5-isopropylazulene and 1,3-diethyl-5-isopropylazulene are eluted in this order with the resolution between these peaks being not less than 3.

System repeatability: When the test is repeated 6 times with 20 μL of the solution for system suitability test under the above operating conditions, the relative standard deviations of the peak areas of 1-ethyl-5-isopropylazulene and 1,3-diethyl-5-isopropylazulene are not more than 2.0%, respectively.

(2) Related substances−Dissolve 20 mg of Egualen Sodium RS in 100 mL of the mobile phase, and use this solution as the sample solution. Pipet 1 mL of this solution, and add the mobile phase to make exactly 100 mL. Pipet 5 mL of this solution, add the mobile phase to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 20 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions. Determine each peak area of both solutions by the automatic integration method: the total area of the peaks, having the relative retention time of not less than 0.25 with respect to egualen, obtained from the sample solution is not larger than the peak area of egualen from the standard solution, and the total area of the peaks having the relative retention time of not more than 0.25 with respect to egualen, obtained from the sample solution is not larger than the peak area of egualen from the standard solution.

Operating conditions

Detector: An ultraviolet absorption photometer (wavelength: 285 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 20ºC.

Mobile phase: Dissolve 2.7 g of potassium dihydrogen phosphate in water to make 1000 mL, and

adjust the pH to 6.0 with a solution prepared by dissolving 7.2 g of disodium hydrogen phosphate dodecahydrate in water to make 1000 mL. To 700 mL of this solution add 300 mL of acetonitrile.

Flow rate: Adjust the flow rate so that the retention time of egualen is about 12 minutes.

Time span of measurement: About 2 times as long as the retention time of egualen.

System suitability

Test for required detectability: Pipet 5 mL of the standard solution, and add the mobile phase to make exactly 25 mL. Confirm that the peak area of egualen obtained from 20 μL of this solution is equivalent to 15 to 25% of that from 20 μL of the standard solution.

System performance: To 10 mg of methyl parahydroxybenzoate add 5 mL of the sample solution, and add the mobile phase to make 25 mL. When the procedure is run with 20 μL of this solution under the above operating conditions, methyl parahydroxybenzoate and egualen are eluted in this order with the resolution between these peaks being not less than 2.0.

System repeatability: When the test is repeated 6 times with 20 μL of the standard solution under the above operating conditions, the relative standard deviation of the peak area of egualen is not more than 5%.

Water <*2.48*>: 1.8 − 2.2% (0.5 g, volumetric titration, direct titration).

Content: not less than 99.0% of egualen sodium ($C_{15}H_{17}NaO_3S$: 300.35), calculated on the anhydrous basis. Assay−Weigh accurately about 0.3 g of Egualen Sodium RS, dissolve in 30 mL of water, apply to a chromatographic column, 15 mm in diameter, previously prepared with 2 g of strongly acidic ion exchange resin for column chromatography (type H), and allow to flow at a rate of 5 mL per minute. Wash the column with 50 mL of water, combine the washing and the former effluent solution, and titrate <*2.50*> with 0.05 mol/L sodium hydroxide VS (potentiometric titration). Perform a blank determination in the same manner, and make any necessary correction.

Each mL of 0.05 mol/L sodium hydroxide VS
= 15.02 mg of $C_{15}H_{17}NaO_3S$

1-Ethyl-5-Isopropylazulene $C_{15}H_{18}$ A clear and blue liquid.

Identification (1) Determine the absorption spectrum of a solution of 1-Ethyl-5-Isopropylazulene in methanol (1 in 4000) as directed under Ultraviolet-visible Spectrophotometry <*2.24*>: it exhibits a maximum between 613 nm and 617 nm. Also, determine the absorption spectrum of a solution of 1-Ethyl-5-Isopropylazulene in methanol (1 in 400000) as directed under Ultraviolet-visible Spectrophotometry <*2.24*>: it exhibits a maximum between 279 nm and 283 nm.

(2) Determine the infrared absorption spectrum of 1-Ethyl-5-Isopropylazulene as directed in the liquid film method under Infrared Spectrophotometry <*2.25*>: it exhibits absorption at the wave numbers of about 2950 cm^{-1} and 1572 cm^{-1}.

Related substances−Dissolve 10 mg of 1-Ethyl-5-Isopropylazulene in 100 mL of methanol, and use this solution as the sample solution. Pipet 1 mL of this solution, add methanol to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 20 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions. Determine each peak area of both solutions by the automatic integration method: the total area of the peaks other than 1-ethyl-5-isopropylazulene obtained from the sample solution is not larger than the peak area of 1-ethyl-5-isopropylazulene from the standard solution.

Operating conditions

Detector: An ultraviolet absorption photometer (wavelength: 285 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 20ºC.

Mobile phase: Dissolve 2.7 g of potassium dihydrogen phosphate in water to make 1000 mL, and adjust the pH to 6.0 with a solution prepared by dissolving 7.2 g of disodium hydrogen phosphate dodecahydrate in water to make 1000 mL. To 100 mL of this solution add 400 mL of acetonitrile.

Flow rate: Adjust the flow rate so that the retention time of 1-ethyl-5-isopropylazulene is about 9 minutes.

Time span of measurement: About 2 times as long as the retention time of 1-ethyl-5-isopropylazulene.

System suitability

Test for required detectability: Pipet 1 mL of the standard solution, and add methanol to make exactly 20 mL. Confirm that the peak area of 1-ethyl-5-isopropylazulene obtained from 20 μL of this solution is equivalent to 3.5 to 6.5% of that from 20 μL of the standard solution.

System performance: Dissolve 10 mg each of 1-ethyl-5-isopropylazulene and 1,3-diethyl-5-isopropylazulene in methanol to make 100 mL. To 1 mL each of these solutions add methanol to make 50 mL. To 1 mL of this solution add methanol to make 50 mL, and use this solution as the solution for system suitability test. When the procedure is run with 20 μL of the solution for system suitability test under the above operating conditions, 1-ethyl-5-isopropylazulene and 1,3-diethyl-5-isopropylazulene are eluted in this order with the resolution between these peaks being not less than 3.

System repeatability: When the test is repeated 6 times with 20 μL of the solution for system suitability test under the above operating conditions, the relative standard deviations of the peak areas of 1-ethyl-5-isopropylazulene and 1,3-diethyl-5-isopropylazulene are not more than 10%, respectively.

1,3-Diethyl-5-Isopropylazulene $C_{17}H_{22}$ A clear and blue liquid.

Identification (1) Determine the absorption spectrum of a solution of 1,3-Diethyl-5-Isopropylazulene in methanol (1 in 4000) as directed under Ultraviolet-visible Spectrophotometry <*2.24*>: it exhibits a maximum between 640 nm and 644 nm. Also, determine the absorption spectrum of a solution of 1,3-Diethyl-5-Isopropylazulene in methanol (1 in 400000) as directed under Ultraviolet-visible Spectrophotometry <*2.24*>: it exhibits a maximum between 282 nm and 286 nm.

(2) Determine the infrared absorption spectrum of 1,3-Diethyl-5-Isopropylazulene as directed in the liquid film method under Infrared Spectrophotometry <*2.25*>: it exhibits absorption at the wave numbers of about 2950 cm^{-1} and 1572 cm^{-1}.

Related substances−Dissolve 10 mg of 1,3-Diethyl-5-Isopropylazulene in 100 mL of methanol, and use this solution as the sample solution. Pipet 1 mL of this solution, add methanol to make exactly 100 mL, and use this solution as the standard solution. Perform the test with exactly 20 μL each of the sample solution and standard solution as directed under Liquid Chromatography <*2.01*> according to the following conditions. Determine each peak area of both solutions by the automatic integration method: the total area of the peaks other than 1,3-diethyl-5-isopropylazulene obtained from the sample solution is not larger than the peak area of 1,3-diethyl-5-isopropylazulene from the standard solution.

Operating conditions

Detector: An ultraviolet absorption photometer (wavelength: 285 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 15 cm in length, packed with octadecylsilanized silica gel for liquid chromatography (5 μm in particle diameter).

Column temperature: A constant temperature of about 20ºC.

Mobile phase: Dissolve 2.7 g of potassium dihydrogen phosphate in water to make 1000 mL, and adjust the pH to 6.0 with a solution prepared by dissolving 7.2 g of disodium hydrogen phosphate dodecahydrate in water to make 1000 mL. To 100 mL of this solution add 400 mL of acetonitrile.

Flow rate: Adjust the flow rate so that the retention time of 1,3-diethyl-5-isopropylazulene is about 15 minutes.

Time span of measurement: About 2 times as long as the retention time of 1,3-diethyl-5-isopropylazulene.

System suitability

Test for required detectability: Pipet 1 mL of the standard solution, and add methanol to make exactly 20 mL. Confirm that the peak area of 1,3-diethyl-5-isopropylazulene obtained from 20 μL of this solution is equivalent to 3.5 to 6.5% of that from 20 μL of the standard solution.

System performance: Dissolve 10 mg each of 1-ethyl-5-isopropylazulene and 1,3-diethyl-5-isopropylazulene in methanol to make 100 mL. To 1 mL each of these solutions add methanol to

make 50 mL. To 1 mL of this solution add methanol to make 50 mL, and use this solution as the solution for system suitability test. When the procedure is run with 20 μL of the solution for system suitability test under the above operating conditions, 1-ethyl-5-isopropylazulene and 1,3-diethyl-5-isopropylazulene are eluted in this order with the resolution between these peaks being not less than 3.

System repeatability: When the test is repeated 6 times with 20 μL of the solution for system suitability test under the above operating conditions, the relative standard deviations of the peak areas of 1-ethyl-5-isopropylazulene and 1,3-diethyl-5-isopropylazulene are not more than 10%, respectively.