tion under the above operating conditions. Use a column giving elution of sennoside A and naringin in this order with the resolution between these peaks being not less than 3.

System repeatability: Repeat the test 6 times with the standard solution under the above operating conditions: the relative standard deviation of the peak areas of sennoside A is not more than 1.5%.

Powdered Rhubarb

Rhei Rhizoma Pulveratum

ダイオウ末

Powdered Rhubarb is the powder of Rhubarb.

It contains not less than 0.25% of sennoside A, calculated on the basis of dried materials.

Description Powdered Rhubarb occurs as a brown powder. It has a characteristic odor and a slightly astringent and bitter taste; is gritty between the teeth and colors the saliva yellow on chewing.

Under a microscope, Powdered Rhubarb reveals starch grains, dark brown substances or druses of calcium oxalate, fragments of parenchyma cells containing them, and reticulate vessels. The starch grains are spherical, simple, or 2- to 4-compound grains. Simple grain, 3 - 18 μm in diameter, rarely 30 μm; crystal druses of calcium oxalate, 30 - 60 μm in diameter, sometimes exceeding 100 μm.

Identification To 2.0 g of Powdered Rhubarb add 40 mL of a mixture of tetrahydrofuran and water (7:3), shake for 30 minutes, and centrifuge. Transfer the supernatant liquid to a separator, add 13 g of sodium chloride, and shake for 30 minutes. Separate the water layer with undissolved sodium chloride, and adjust the pH to 1.5 with 1 mol/L hydrochloric acid TS. Transfer this solution to another separator, add 30 mL of tetrahydrofuran, shake for 10 minutes, separate the tetrahydrofuran layer, and use this solution as the sample solution. Separately, dissolve 1 mg of sennoside A for thin-layer chromatography in 4 mL of a mixture of tetrahydrofuran and water (7:3), and use this solution as the standard solution. Perform the test with these solutions as directed under the Thin-layer Chromatography. Spot 40 μL each of the sample solution and the standard solution on a plate of silica gel for thin-layer chromatography at 10 mm along the initial line. Develop the plate with a mixture of 1-propanol, ethyl acetate, water and acetic acid (100) (40:40:30:1) to a distance of about 15 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 365 nm): one of the spots from the sample solution and a red fluorescent spot from the standard solution show the same in color tone and *Rf* value.

Purity Raponticin—To 0.5 g of Powdered Rhubarb add 10 mL of ethanol (95), heat on a water bath under a reflux condenser for 10 minutes, and filter. Perform the test as directed under the Thin-layer Chromatography, using the filtrate as the sample solution. Spot 10 μL of the sample solution on a plate of silica gel for thin-layer chromatography. Develop the chromatogram with a mixture of isopropyl ether, methanol and 1-butanol (26:7:7) to a distance of about 10 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 365 nm): no spot with blue-purple fluorescence is observed at the *Rf* value of between 0.3 and 0.6, though a bluish white fluorescence may appear.

Loss on drying Not more than 13.0% (6 hours).

Total ash Not more than 13.0%.

Acid-insoluble ash Not more than 2.0%.

Extract content Dilute ethanol-soluble extract: not less than 30.0%.

Component determination Weigh accurately about 0.5 g of Powdered Rhubarb, add exactly 50 mL of a solution of sodium hydrogen carbonate (1 in 1000), shake for 30 minutes, filter, and use the filtrate as the sample solution. Separately, weigh accurately about 0.01 g of sennoside A for component determination, previously dried in a desiccator (in vacuum at a pressure not exceeding 0.67 kPa, phosphorus (V) oxide) for not less than 12 hours, dissolve in a solution of sodium hydrogen carbonate (1 in 1000) to make exactly 50 mL. Pipet 5 mL of this solution, add a solution of sodium hydrogen carbonate (1 in 1000) to make exactly 20 mL, and use this solution as the standard solution. Pipet 10 μL 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 sennoside A in each solution.

$$\text{Amount (mg) of sennoside A} = \text{amount (mg) of sennoside A for component determination} \times \frac{A_T}{A_S} \times 0.25$$

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 340 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 for liquid chromatography (5 to 10 μm in particle diameter).

Column temperature: A constant temperature of about 40°C.

Mobile phase: A mixture of diluted acetic acid (100) (1 in 80) and acetonitrile (4:1).

Flow rate: Adjust the flow rate so that the retention time of sennoside A is about 15 minutes.

Selection of column: Dissolve 1 mg each of sennoside A for component determination and naringin for thin-layer chromatography in a solution of sodium hydrogen carbonate (1 in 1000) to make 10 mL. Proceed with 20 μL of this solution under the above operating conditions, and calculate the resolution. Use a column giving elution of sennoside A and naringin in this order with the resolution between these peaks being not less than 3.

System repeatability: When the test is repeated 6 times with the standard solution under the above operating conditions, the relative standard deviation of the peak areas of sennoside A is not more than 1.5%.