

component determination and 4-hydroxy-3-methoxybenzyl nonylic acid amide in methanol to make 50 mL. When the procedure is run with 20 μ L of this solution under the above operating conditions, 4-hydroxy-3-methoxybenzyl nonylic acid amide and capsaicin are eluted in this order with the resolution between these peaks being not less than 1.5.

System repeatability: When the test is repeated 6 times with 20 μ L of the standard solution under the above operating conditions, the relative standard deviation of the peak areas of capsaicin is not more than 1.5%.

Powdered Capsicum

Capsici Fructus Pulveratus

トウガラシ末

Powdered Capsicum is the powder of Capsicum.

Powdered Capsicum contains not less than 0.10% of total capsaicins (capsaicin and dihydrocapsaicin), calculated on the basis of dried material.

Description Powdered Capsicum occurs as a yellow-red powder. It has a slight, characteristic odor and a hot, acrid taste.

Under a microscope, Powdered Capsicum reveals fragments of parenchyma containing oil droplets and yellow-red chromoplasts; fragments of outer pericarp with thick cuticle; fragments of stone cells from inner surface of pericarp, with wavy curved side walls; fragments of thin vessels; fragments of seed coat with thick wall, and fragments of parenchyma consisting of small cells of endosperm containing fixed oil and aleuron grains.

Identification To 2.0 g of Powdered Capsicum add 5 mL of ethanol (95), warm on a water bath for 5 minutes, cool, centrifuge, and use the supernatant liquid as the sample solution. Separately, dissolve 1 mg of capsaicin for thin-layer chromatography in 1 mL of ethanol (95), 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 diethyl ether and methanol (19:1) to a distance of about 12 cm, and air-dry the plate. Spray evenly 2,6-dibromo-N-chloro-1,4-benzoquinone monoimine TS on the plate, and allow to stand in ammonia gas: a spot from the sample solution and blue spot from the standard solution show the same in color tone and *R_f* value.

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

Total ash Not more than 8.0%.

Acid-insoluble ash Not more than 1.2%.

Extract content Ether-soluble extract: not less than 9.0%.

Component determination Weigh accurately about 0.5 g of medium powder of Powdered Capsicum in a glass-stoppered centrifuge tube, add 30 mL of methanol, shake for 15 minutes, centrifuge, and separate the supernatant liquid. To the residue add 10 mL of methanol, shake for 5 minutes, centrifuge, and separate the supernatant liquid. Repeat this

procedure again, combine the extracts, add methanol to make exactly 50 mL, and use this solution as the sample solution. Separately, weigh accurately about 0.01 g of capsaicin for component determination, previously dried in a desiccator (in vacuum, phosphorus (v) oxide, 40°C) for 5 hours, and dissolve in methanol to make exactly 50 mL. Pipet 2 mL of this solution, add methanol to make exactly 25 mL, and use this solution as the standard solution. Perform the test with exactly 20 μ L each of the sample solution and the standard solution as directed under the Liquid Chromatography according to the following conditions, and determine the peak areas, A_{TC} and A_{TD} , of capsaicin and dihydrocapsaicin (the relative retention time to capsaicin is about 1.3) in the sample solution, and the peak area, A_S , of capsaicin in the standard solution.

$$\begin{aligned} &\text{Amount (mg) of total capsaicins} \\ &= \text{amount (mg) of capsaicin for component} \\ &\quad \text{determination} \\ &\quad \times \frac{A_{TC} + A_{TD}}{A_S} \times 0.08 \end{aligned}$$

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength : 281 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 25 cm in length, packed with phenylated silica gel for liquid chromatography (5 μ m in particle diameter).

Column temperature: A constant temperature of about 30°C.

Mobile phase: A mixture of diluted phosphoric acid (1 in 1000) and acetonitrile (3:2).

Flow rate: Adjust the flow rate so that the retention time of capsaicin is about 20 minutes.

System suitability—

System performance: Dissolve 1 mg each of capsaicin for component determination and 4-hydroxy-3-methoxybenzyl nonylic acid amide in methanol to make 50 mL. When the procedure is run with 20 μ L of this solution under the above operating conditions, 4-hydroxy-3-methoxybenzyl nonylic acid amide and capsaicin are eluted in this order with the resolution between these peaks being not less than 1.5.

System repeatability: When the test is repeated 6 times with 20 μ L of the standard solution under the above operating conditions, the relative standard deviation of the peak areas of capsaicin is not more than 1.5%.

Capsicum Tincture

トウガラシチンキ

Capsicum Tincture contains not less than 0.010 w/v% of total capsaicins (capsaicin and dihydrocapsaicin).

Method of preparation

Capsicum, in medium cutting	100 g
Ethanol	a sufficient quantity
To make 1000 mL	

Prepare as directed under Tinctures, with the above ingredients.

Description Capsicum Tincture is a yellow-red liquid. It has a burning, pungent taste.

Specific gravity d_{20}^{20} : about 0.82

Identification Proceed as directed in the Identification under Capsicum, using Capsicum Tincture as the sample solution. Spot 20 μ L each of the sample solution and the standard solution.

Alcohol number Not less than 9.7 (Method 2).

Component determination Pipet 2 mL of Capsicum Tincture, add methanol to make exactly 20 mL, and use this solution as the sample solution. Separately, weigh accurately about 0.01 g of capsaicin for component determination, previously dried in a desiccator (in vacuum, phosphorus (v) oxide, 40°C) for 5 hours, dissolve in methanol to make exactly 50 mL. Pipet 2 mL of this solution, add methanol to make exactly 25 mL, and use this solution as the standard solution. Perform the test with exactly 20 μ L each of the sample solution and the standard solution as directed under the Liquid Chromatography according to the following conditions, and determine the peak areas, A_{TC} and A_{TD} , of capsaicin and dihydrocapsaicin (the relative retention time to capsaicin is about 1.3) in the sample solution, and the peak area, A_S , of capsaicin in the standard solution.

$$\begin{aligned} & \text{Amount (mg) of total capsaicins} \\ &= \text{amount (mg) of capsaicin for component} \\ & \quad \text{determination} \\ & \quad \times \frac{A_{TC} + A_{TD}}{A_S} \times 0.032 \end{aligned}$$

Operating conditions—

Detector: An ultraviolet absorption photometer (wavelength: 281 nm).

Column: A stainless steel column 4.6 mm in inside diameter and 25 cm in length, packed with phenylated silica gel for liquid chromatography (5 μ m in particle diameter).

Column temperature: A constant temperature of about 30°C.

Mobile phase: A mixture of diluted phosphoric acid (1 in 1000) and acetonitrile (3:2).

Flow rate: Adjust the flow rate so that the retention time of capsaicin is about 20 minutes.

System suitability—

System performance: Dissolve 1 mg each of capsaicin for component determination and 4-hydroxy-3-methoxybenzyl nonylic acid amide in methanol to make 50 mL. When the procedure is run with 20 μ L of this solution under the above operating conditions, 4-hydroxy-3-methoxybenzyl nonylic acid amide and capsaicin are eluted in this order with the resolution between these peaks being not less than 1.5.

System repeatability: When the test is repeated 6 times with 20 μ L of the standard solution under the above operating conditions, the relative standard deviation of the peak areas of capsaicin is not more than 1.5%.

Containers and storage Containers—Tight containers.

Storage—Light-resistant.

Capsicum and Salicylic Acid Spirit

トウガラシ・サリチル酸精

Method of preparation

Capsicum Tincture	40 mL
Salicylic Acid	50 g
Liquefied Phenol	20 mL
Castor Oil	100 mL
Aromatic substance	a suitable quantity
Ethanol	a sufficient quantity
To make 1000 mL	

Prepare as directed under Medicated Spirits, with the above ingredients.

Description Capsicum and Salicylic Acid Spirit is a light brown-yellow liquid.

Specific gravity d_{20}^{20} : about 0.84

Identification (1) Shake 10 mL of Capsicum and Salicylic Acid Spirit with 15 mL of sodium hydrogen carbonate TS and 10 mL of diethyl ether, and separate the water layer. To 1 mL of the solution add hydrochloric acid-potassium chloride buffer solution, pH 2.0, to make 200 mL, and to 5 mL of this solution add 5 mL of a solution of iron (III) nitrate enneahydrate (1 in 200): a red-purple color is produced (salicylic acid).

(2) To 0.5 mL of Capsicum and Salicylic Acid Spirit add 20 mL of water and 5 mL of dilute hydrochloric acid, extract with 20 mL of diethyl ether, wash the diethyl ether extract with two 5-mL portions of sodium hydrogen carbonate TS, and then extract with 20 mL of dilute sodium hydroxide TS. To 1 mL of the extract add 1 mL of sodium nitrite TS and 1 mL of dilute hydrochloric acid, shake, and allow to stand for 10 minutes. Add 3 mL of sodium hydroxide TS: a yellow color is produced (phenol).

(3) To 0.2 mL of Capsicum and Salicylic Acid Spirit add 5 mL of dilute hydrochloric acid, extract with 5 mL of chloroform, and use the extract as the sample solution. Dissolve 0.01 g of salicylic acid and 0.02 g of phenol in 5 mL and 25 mL of chloroform, respectively, and use both solutions as standard solution (1) and standard solution (2). Perform the test with the sample solution and standard solution (1) and standard solution (2) as directed under Thin-layer Chromatography. Spot 5 μ L each of these solutions on a plate of silica gel with fluorescent indicator for thin-layer chromatography. Develop the plate with a mixture of chloroform, acetone and acetic acid (100) (45:5:1) to a distance of about 10 cm, and air-dry the plate. Examine under ultraviolet light (main wavelength: 254 nm): two spots from the sample solution exhibit the same R_f values as those from standard solution (1) and standard solution (2). Spray evenly iron (III) chloride TS upon the plate: the spot from standard solution (1) and the corresponding spot from the sample solution reveal a purple color.

Alcohol number Not less than 8.1 (Method 2). Prepare the sample solution as follows: Pipet 5 mL of Capsicum and Salicylic Acid Spirit at $15 \pm 2^\circ\text{C}$ into a glass-stoppered, conical flask containing exactly 45 mL of water while shaking vigorously, allow to stand, and filter the lower layer. Discard the first 15 mL of the filtrate. Pipet 25 mL of the subsequent