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GB 5009.286-2022

National food safety standard - Determination of Natamycin in foods

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Table of Contents

Foreword 3
1 Scope 4
2 Principle 4
3 Reagents and Materials 4
4 Instruments and Equipment 5
5 Analytical Procedures 5
6 Expression of Analytical Results 7
7 Precision 8
8 Others 8
Appendix A Liquid Chromatogram of Natamycin Reference Substance 9

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1 Scope

This Standard specifies the method of liquid chromatography for the determination of natamycin in foods.

This Standard is applicable to the determination of natamycin in cheese, processed cheese and similar products, marinated meat products, meats (smoked, broiled and grilled), fried meats, western-style ham, meat sausages, fermented meat products, mayonnaise, salad dressings, cakes, fruit and vegetable juice (pulp), fruit juice drinks and fermented wines.

2 Principle

The sample is extracted or diluted with methanol. For the sample containing grease or solid sample, add water to freeze it or centrifuge it to remove the fat or solid particle component in the sample. After filtration, determine the extracting solution through reversed-phase liquid chromatography and ultraviolet detector. Use the external standard method to quantify it.

3 Reagents and Materials

Unless it is otherwise specified, the reagents used in this Method are analytically pure; the water is Grade-1 water specified in GB/T 6682.

3.2 Reference Substance

Natamycin (C33H47NO13, CAS. 7681-93-8). purity ? 98%, or reference substances

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certified by the state and awarded with a reference substance certificate.

3.3.1 Natamycin standard stock solution (250 mg/L). accurately weigh-take 25 mg of

natamycin reference substance (accurate to 0.1 mg); add an appropriate amount of methanol and 1 mL of glacial acetic acid to perform ultrasonic dissolution; use methanol to reach a Weigh-take 5.0 g of sample (accurate to 0.01 g); place it in a 50 mL centrifuge tube; add 15.0 mL of methanol to perform ultrasonic extraction for 20 min. Then, add 5.0 mL of water to mix it up. At 4,000 r/min, centrifuge it for 10 min. In a refrigerator at ?20 ?C, freeze it for 1 h. The supernatant is filtered through 0.22 ?m organic syringe-type filter. Collect about 2 mL of the filtrate and determine it on the instrument.

5.2.2 Pastry samples Weigh-take 5.0 g of sample (accurate to 0.01 g); place it in a 50 mL centrifuge tube; add 15.0 mL of methanol to perform ultrasonic extraction for 20 min. Then, add 5.0 mL of water to mix it up. At 4,000 r/min, centrifuge it for 10 min. The supernatant is filtered through 0.22 ?m organic syringe-type filter. Collect about 2 mL of the filtrate and determine it on the instrument.

5.2.3 Fruit and vegetable juice (pulp), fruit juice drinks Weigh-take 5.0 g of sample (accurate to 0.01 g); place it in a 50 mL centrifuge tube; add 15.0 mL of methanol to perform ultrasonic extraction for 20 min. At 4,000 r/min, centrifuge it for 10 min. The supernatant is quantitatively transferred to a 25 mL volumetric flask. Use water to reach a constant volume and mix it up. Use 0.22 ?m organic syringe-type filter to filter it.

Collect about 2 mL of the filtrate and determine it on the instrument.

5.2.4 Fermented wines Weigh-take an appropriate amount of the wine sample. Use 0.22 ?m syringe-type filter to filter it. Collect about 2 mL of the filtrate and determine it on the instrument.

5.3 Instrument Reference Conditions

5.3.1 Chromatographic column. C18 chromatographic column (4.6 mm ? 250 mm, 5 ?m), or equivalent.

5.3.2 Mobile phase. A. methanol, B. acetic acid + water = 5 + 95 (volume ratio).

5.3.3 Flow rate. 1.0 mL/min.

5.3.4 Detection wavelength. 305 nm.

5.3.5 Column temperature. 30 ?C.

5.3.6 Injection volume. 10 ?L.

5.3.7 The gradient elution procedure is shown in Table 1.

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