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NATIONAL STANDARD OF THE
PEOPLE'S REPUBLIC OF CHINA

GB 22255-2014

**National food safety standard - Determination of sucralose
in foods**

食品安全国家标准 食品中三氯蔗糖（蔗糖素）的测定

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National food safety standard - Determination of sucralose in foods

1 Scope

This standard specifies the method for the determination of sucralose in food.

This standard applies to the determination of sucralose in food.

2 Principle

The sucralose in the specimen is extracted by methanol aqueous solution, to remove protein and fat; then purified and enriched by solid phase extraction column; separated by high performance liquid chromatography and reversed phase C₁₈ chromatographic column; detected by evaporative light scattering detector or differential detector; qualified according to the retention time and quantified according to peak area.

3 Reagents and materials

Note: Unless otherwise specified, the reagents used in this method are of analytical grade; the water is the grade-1 water specified in GB/T 6682.

3.1 Reagents

3.1.1 Methanol (CH₃OH).

3.1.2 Acetonitrile (CH₃CN): Chromatographically pure.

3.1.3 n-Hexane (C₆H₁₄).

3.1.4 Zinc acetate [Zn(CH₃COO)₂·2H₂O].

3.1.5 Potassium ferrocyanide [K₄Fe(CN)₆·3H₂O].

3.1.6 Neutral alumina (100 mesh ~ 200 mesh).

3.2 Preparation of reagents

3.2.1 Zinc acetate solution (219 g/L): Weigh 21.9 g of zinc acetate. Add 3 mL of acetic acid. Add water to dissolve it to 100 mL.

3.2.2 Potassium ferrocyanide solution (106 g/L): Weigh 10.6 g of potassium ferrocyanide. Add water to dissolve to 100 mL.

3.2.3 Methanol aqueous solution (75 + 25): Measure 75 mL of methanol. Add 25 mL of water. Mix well.

3.2.4 Acetonitrile aqueous solution (11 + 89): Measure 11 mL of acetonitrile. Add 89 mL of water. Mix well.

3.3 Standards

Sucralose standard ($C_{12}H_{19}C_{13}O_8$): CAS No. 56038-13-2, purity $\geq 99\%$.

3.4 Preparation of standard solution

3.4.1 Standard stock solution of sucralose (10.0 mg/mL): Weigh 0.25 g (accurate to 0.0001 g) of sucralose standard product, in a 25 mL volumetric flask. Use water to dilute to the mark. Mix well. The concentration of sucralose is 10.0 mg/mL. The stock solution is stored in a refrigerator at 4 °C. The shelf life is 6 months.

3.4.2 Standard intermediate solution of sucralose (1.00 mg/mL): Pipette 5.00 mL of sucralose standard stock solution, into a 50 mL volumetric flask. Use water to dilute it to the mark. Mix well. The concentration of sucralose is 1.00 mg/mL. It is stored in a refrigerator at 4 °C. The shelf life is 3 months.

3.4.3 Standard working solution of sucralose: Pipette 0.200 mL, 0.500 mL, 1.00 mL, 2.00 mL, 4.00 mL of sucralose intermediate solution, into a 10 mL volumetric flask. Use water to dilute to the mark. The concentration of sucralose working solution is 0.0200 mg/mL, 0.0500 mg/mL, 0.100 mg/mL, 0.200 mg/mL, 0.400 mg/mL, respectively.

3.5 Materials

Solid-phase extraction cartridges (200 mg, type N-vinylpyrrolidone and divinylbenzene hydrophilic-lipophilic balanced packing) are activated, by 4 mL of methanol and 4 mL of water sequentially, before use.

4 Instruments and equipment

4.1 High performance liquid chromatography: Equipped with a differential detector or an evaporative light scattering detector.

4.2 Balance: Sensitivity is 0.1 mg and 1 mg.

4.3 Vortex mixer.

4.4 Centrifuge: speed ≥ 3000 r/min.

Weigh 2 g of the mixed specimen (accurate to 0.001 g). Put it in a 50 mL centrifuge tube. Add 1.0 g of neutral alumina. Add 3 mL of water. Oscillate it on a vortex mixer for 3 min. Then add 15 mL of methanol. The following steps are same as from 5.1.1.1 "Continue to oscillate for 30 s. Ultrasonically extract for 20 min. Centrifuge at 3000 r/min for 10 min", to 5.1.1.3 "The filtrate is the prepared specimen solution, which is set aside".

5.1.3 Alcohol-containing specimens (fermented wine, blended wine)

Weigh 5 g of the mixed specimen (accurate to 0.001 g). Place it in a 50 mL evaporating dish. Evaporate to dryness on a boiling water bath. Use 1.00 mL of acetonitrile aqueous solution (11 + 89), to dissolve the residue. Make the solution pass through a 0.45 µm filter membrane. The filtrate is the prepared specimen solution, which is set aside.

5.1.4 Beverages

Weigh 5 g of the mixed specimen (accurate to 0.001 g). Place it in a 15 mL centrifuge tube. Add 5 mL of water. Oscillate on a vortex mixer for 30 s. Centrifuge at 3000 r/min for 10 min. The following steps are processed according to 5.1.1.3.

5.1.5 Flavored fermented milk and milk tea

Weigh 1 g ~ 5 g of the mixed specimen (accurate to 0.001 g). Place it in a 50 mL centrifuge tube. Add 5 mL of water. Oscillate on a vortex mixer for 3 min. Then add 15 mL of methanol, 0.50 mL of zinc acetate solution, 0.50 mL of potassium ferrocyanide solution. The following steps are same as from 5.1.1.1 "Continue to oscillate for 30 s. Ultrasonically extract for 20 min. Centrifuge at 3000 r/min for 10 min", to 5.1.1.3 "The filtrate is the prepared specimen solution, which is set aside".

The pretreatment of different specimens requires a specimen blank test at the same time.

5.2 Instrument reference conditions

5.2.1 Chromatographic column: C₁₈ column (4.6 mm × 150 mm, 5 µm) or equivalent performance.

5.2.2 Mobile phase: water + acetonitrile = 89 + 11.

Note: When the detection sample matrix is complex AND the strongly retained substances affect the subsequent detection, an elution procedure can be adopted (applicable to evaporative light scattering detectors), as shown in Appendix A.

5.2.3 Flow rate: 1.0 mL/min.

5.2.4 Column temperature: 35 °C.

5.2.5 Differential detector conditions:

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