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NATIONAL STANDARD OF THE
PEOPLE'S REPUBLIC OF CHINA

GB 5009.263-2016

**National Standard of Food Safety -
Determination of Aspartame and Alitame in Foods**

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National Standard of Food Safety -

Determination of Aspartame and Alitame in Foods

1 Scope

This Standard specifies the determination of aspartame and alitame in foods.

This Standard is applicable to the determination of aspartame and alitame in foods.

2 Principle

According to the characteristics that the aspartame and alitame are soluble in water, while the methanol and ethanol are not soluble in the fat-soluble solvents, use aqueous methanol solution to extract the samples of vegetables and their products, fruits and their products, edible fungi and algae, cereals and their products, baked products, puffed products and jellies under ultrasonic vibration conditions; use water to extract the samples of concentrated juices, carbonated beverages, solid beverages, table sauces and other candies except gum base candies; after the ethanol precipitates the protein, use the aqueous ethanol solution to extract the samples of dairy products, milk-containing beverages and frozen drinks; for gum base candies, use n-hexane to dissolve gum base, then use water to extract; use water to extract the fat emulsified products, cocoa products, chocolate and its products, nuts and seeds, aquatic products, and egg products, then use n-hexane to remove the fat compositions. Separate the extracting solutions on the liquid chromatography C₁₈ reversed-phase column; detect at a wavelength of 200nm; qualitative by retention time of the chromatography peak; and quantitative by the external standard method.

3 Reagents and Materials

Unless otherwise specified, all used reagents shall be analytically pure; while the water shall be Class-I water for laboratory as specified in GB/T 6682.

3.1 Reagents

3.1.1 Methanol (CH₃OH): chromatographically pure.

3.1.2 Ethanol (CH₃CH₂OH): guarantee reagent.

20min; transfer the extracting solution into 25mL volumetric flask; add 10mL of water again into the beaker, use ultrasonic vibrator to extract for 10min; transfer the extracting solution into the same 25mL volumetric flask for later-use. Use water to make constant volume against the above liquid in the volumetric; mix evenly; centrifuge for 5min at 4000r/min; filter the supernatant through 0.45μm aqueous filter membrane for the chromatographic analysis.

5.1.2 Dairy products, dairy drinks and frozen drinks

For the liquid-dairy products containing the solid fruit pulp, it is necessary to use a food processor for homogenization; for the solid dairy products such as cheese, it is necessary to use a food processor for homogenization according to the mass ratio of 1:4 between specimen and water.

Take about 5g (accurate to 0.001g) of homogenate specimen of liquid-dairy products, milk-containing beverages, frozen drinks, solid dairy products into 50mL centrifuge tube; add 10mL of ethanol; cover the lid; for the milk-containing beverage and frozen drinks specimens, firstly, gently invert the centrifuge tube 5 times (not shaking); for dairy products, firstly perform vortex mixing against the centrifuge tube for 10s, then stand for 1min, centrifuge 5min at 4000r/min; filter the supernatant into a 25mL volumetric flask; use 8mL of ethanol-water (2+1) to wash the precipitation; after centrifuging, the supernatant is transferred into the same 25mL volumetric flask; use the ethanol-water (2+1) to make constant volume; filter through a 0.45μm organic filter membrane for the chromatographic analysis.

5.1.3 Jelly

For the suckable and transparent jellies, stir well with a glass rod; for the jellies containing the fruit pulp, it is necessary to use a food processor for homogenization.

Take about 5g (accurate to 0.001g) of uniformly-prepared jelly specimen into a 50mL colorimetric tube; add 25mL of 80% methanol aqueous solution; heat on the 70°C water bath for 10min; take out the colorimetric tube; transfer the extracting solution into a 50mL volumetric flask as it is hot; then use 15mL of 80% methanol aqueous solution to wash the colorimetric tube for twice; each time shake it for about 10s; transfer into the same 50mL volumetric flask; cool off to the room temperature; use 80% methanol aqueous solution to make constant volume to the scale; mix evenly; centrifuge for 5min at 4000r/min; filter the supernatant through a 0.45μm organic filter membrane for chromatographic analysis.

5.1.4 Vegetables and their products, fruits and their products, edible fungi and algae

The specimen of fruits and their products shall first be removed if they have fruit core.

For the dry and hard specimen, use a food processor to homogenize the specimen

scale; after shaking evenly; filter through a 0.45 μ m aqueous filter membrane for the chromatographic analysis.

Fat emulsified products, cocoa products, chocolate and its products, nuts and seeds, aquatic products, egg products: use a food processor to homogenize according to the mass ratio 1:4 between the specimen and water; take about 5g (accurate to 0.001g) of homogenate specimen into a 25mL centrifuge tube; add 10mL of water to extract for 20min through ultrasonic vibration; stand for 1min; centrifuge for 5min at 4000r/min; transfer the supernatant into a 100mL separator funnel; add 8mL of water into the centrifuge tube to extract for 10min through ultrasonic vibration; after standing and centrifuging, transfer the supernatant into the separator funnel; add 15mL of n-hexane into the separator funnel; shake for 30s; stand and layer for 5min; place the lower aqueous phase into a 25mL volumetric flask; use water to make constant volume to the scale; after shaking evenly, filter through a 0.45 μ m aqueous filter membrane for the chromatographic analysis.

5.2 Apparatus reference conditions

- a) Chromatographic column: C₁₈, column length of 250mm, inner diameter of 4.6mm, particle size of 5 μ m.
- b) Column temperature: 30°C.
- c) Mobile phase: methanol-water (40+60) or acetonitrile-water (20+80).
- d) Flow rate: 0.8mL/min.
- e) Sample-injection volume: 20 μ L.
- f) Detector: diode array detector or UV detector.
- g) Detection wavelength: 200nm.

5.3 Drawing of standard curve

Measure the corresponding peak area (peak height) of the standard series of working fluid under the above chromatographic conditions; take the concentration of standard working solution as the abscissa, while the peak area (peak height) as the ordinate; then draw the standard curve. The standard chromatograms are shown in Appendix A.

5.4 Measurement of specimen solution

Under the same liquid chromatographic conditions, inject the specimen solution into the liquid chromatograph; qualitative with retention time; quantitative with comparison between the specimen peak height or peak area and the standard one.

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