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NATIONAL STANDARD OF THE
PEOPLE'S REPUBLIC OF CHINA

GB 1886.307-2020

National food safety standard - Food additive -

Potassium copper chlorophyllin

食品安全国家标准 食品添加剂 叶绿素铜钾盐

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National food safety standard - Food additive - Potassium copper chlorophyllin

1 Scope

This Standard is applicable to the food additive potassium copper chlorophyllin that uses silkworm excrement or grass, alfalfa, nettle, spinach and other plants as raw materials to extract chlorophyll or that is processed by saponification, copper substitution and other steps by directly using chlorophyll as raw material. The solvent used is acetone, dichloromethane, methanol, ethanol, isopropanol, n-hexane and (or) petroleum ether (boiling range is 90°C ~120°C).

2 Molecular formula and relative molecular mass

2.1 Molecular formula

Copper dipotassium chlorophyllin ($C_{34}H_{30}O_5N_4CuK_2$)

Copper tripotassium chlorophyllin ($C_{34}H_{31}O_6N_4CuK_3$)

2.2 Relative molecular mass

Copper dipotassium chlorophyllin: 716.38 (according to 2016 international relative atomic mass)

Copper tripotassium chlorophyllin: 772.48 (according to 2016 international relative atomic mass)

3 Technical requirements

3.1 Sensory requirements

Sensory requirements shall comply with the provisions of Table 1.

Table 1 -- Sensory requirements

Annex A

Inspection methods

A.1 General

Unless otherwise specified, the reagents and water used in this Standard shall refer to the analytically-pure reagents AND grade 3 water specified in GB/T 6682. The standard solutions, standard solutions for impurity determination, preparations and products used in the test are prepared in accordance with GB/T 601, GB/T 602, and GB/T 603 when other requirements are not specified. The solution used in the test refers to aqueous solution.

A.2 Identification test

A.2.1 Solubility

Soluble in water. Almost insoluble in low molecular weight alcohols, ketones and ether. Insoluble in chlorinated alkanes, hydrocarbons and fixed oils.

A.2.2 Maximum absorption wavelength

Take the sample solution in the determination of potassium copper chlorophyllin content in A.3.3.1. There are maximum absorption peaks in the two wavelength ranges of $405\text{nm} \pm 3\text{nm}$ and $630\text{nm} \pm 3\text{nm}$.

A.2.3 Copper potassium ion test

Take 1g of sample. Place in a crucible that has been burnt to constant weight at $800^{\circ}\text{C} \pm 25^{\circ}\text{C}$. Slowly heat until the sample is completely carbonized. Cool the carbonized sample. Use 0.5mL~1mL of sulfuric acid to wet the residue. Continue heating until the sulfuric acid vapor is exhausted. Burn the residue to constant weight in a high temperature furnace of $800^{\circ}\text{C} \pm 25^{\circ}\text{C}$. Add 10mL of hydrochloric acid solution (1+3) to the residue. Heat on a water bath to dissolve. Replenish water to 10mL after filtration, as sample solution. Perform the following tests:

- a) Take 5mL of sample solution. Add 10% ammonia solution. Show blue.
- b) Take 5mL of sample solution. Add 0.5mL of 0.1% sodium diethyldithiocarbamate solution. Generate reddish brown precipitate.
- c) Take sample solution for flame test. Show purple.

A.3 Determination of potassium copper chlorophyllin content (on a dry basis)

565 - Extinction value of a solution with a concentration of 1% in a 1cm cuvette (100mL of solvent contains 1g of potassium copper chlorophyllin);

100 - Concentration conversion factor;

m - Mass of sample, in grams (g);

w₀ - Loss on drying of sample, %.

The test results are based on the arithmetic mean of the parallel determination results. The absolute difference between two independent determination results obtained under repeatability conditions is not more than 1.0% of the arithmetic mean.

NOTE: Measure the loss on drying according to the direct drying method in GB 5009.3. The drying temperature and time are 105°C and 2h respectively.

A.4 Determination of total copper (Cu)

A.4.1 Reagents and materials

Same with GB 5009.13.

A.4.2 Instruments and equipment

Same with GB 5009.13.

A.4.3 Sample processing

A.4.3.1 Microwave digestion

Weigh 0.2g~0.5g of sample, to the nearest of 0.0002g. In the microwave digestion tank, add 5mL of nitric acid. Digest the sample according to the microwave digestion steps. Refer to B.1 for digestion conditions. Take out the digestion tank after cooling. Drive acid to about 1mL at 120°C~140°C. After the digestion tank is left cold, transfer the digestion solution to a 25mL volumetric flask. Use a small amount of grade 2 water to rinse the digestion tank 2~3 times. Combine the washing liquid in the volumetric flask. Use grade 2 water to set volume to the scale. Mix well for future use. Conduct the blank test at the same time.

A.4.3.2 Dry ashing

Weigh 0.1g of sample, to the nearest of 0.0002g. In a crucible, heat with a small fire. Carbonize to smokeless. Transfer to a 550°C muffle furnace. Conduct ashing 3h~4h. Take out after cooling. For samples with incomplete ashing, add a few drops of nitric acid. Heat with a small fire. Steam dry carefully. Then transfer to the 550°C muffle furnace. Continue ashing 1h~2h till the sample is

20µL and inject into the headspace bottle. Cover it. Heat at 60°C for 10min and shake violently. Mix well.

A.7.5.4 Preparation of sample solution

Weigh 0.2g of sample, to the nearest of 0.0001g. Place in a headspace bottle. Add 5.0mL of water and 1.0mL of internal standard solution. Pipette 20µL of N-copper methylpyrrolidine. Inject into the headspace bottle. Cover it. Heat at 60°C for 10min and shake violently. Mix well.

A.7.5.5 Determination

Under the reference operating conditions in A.7.3 and A.7.4, respectively conduct chromatographic analysis of blank solution, standard solution and sample solution after headspace processing.

A.7.6 Result calculation

A.7.6.1 Calibration factor f_i

The calibration factor f_i is calculated according to formula (A.2):

$$f_i = \frac{m_i \times 50}{m_s \times (A_f - A_g)} \dots\dots\dots (A.2)$$

Where,

m_i - Mass of the component to be tested in the standard solution, in milligrams (mg);

50 - Mass conversion factor;

m_s - Mass of internal standard substance in standard solution, in milligrams (mg);

A_f - Ratio of the peak area of the component to be tested to the peak area of the internal standard substance in the standard solution chromatogram;

A_g - Ratio of the peak area of the component to be tested to the peak area of the internal standard substance in the blank solution chromatogram.

A.7.6.2 Content of components to be tested

The content of the components to be tested (dichloromethane, methanol, isopropanol, n-hexane, n-heptane) in the sample, w , in milligrams per gram (mg/kg), is calculated according to formula (A.3):

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