NATIONAL STANDARD OF THE

PEOPLE'S REPUBLIC OF CHINA

GB 5009.280-2020

National food safety standard - Determination of 4-Hexylesorinol Residues in Foods

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1 Scope

This Standard specifies the method for the determination of 4-hexylresorcinol residues in shrimp and crabs.

This Standard is applicable to the determination of 4-hexylresorcinol residues in shrimp and crabs.

2 Principle

The sample was extracted by ethyl acetate, removed fat by acetonitrile saturated n-hexane, separated by high performance liquid chromatography, detected by fluorescence detector, and quantified by external standard method.

3 Reagents and Materials

Unless otherwise specified, the reagents used in this method are analytically pure; and the water is the Class-I water specified in GB/T 6682.

3.1.5 Anhydrous sodium sulfate (Na2SO4): Place anhydrous sodium sulfate in a muffle

furnace at 400°C for 4h; then place it in a desiccator for later-use.

3.2.1 Acetonitrile saturated n-hexane solution: Add a certain amount of acetonitrile to

n-hexane; shake and mix; after the mixture is clearly separated into layers, the upper

5.1 Preparation of the specimen

Take about 500g of the edible part of the representative sample; pulverize and mix by a tissue grinder, and then put it into a clean container as a specimen; seal it and make a mark. The specimen is stored at -18°C.

5.2 Extraction of the specimen

Take 2g (accurate to 0.01g) of the sample in a 50mL centrifuge tube; add 20mL of ethyl acetate and 3g of anhydrous sodium sulfate; homogenize for 30s; then centrifuge at 4000r/min for 3min; and transfer all the supernatant to a 100mL concentrate bottle.

Add 20mL ethyl acetate to the residue again; homogenize for 30s; then centrifuge at 4000r/min for 3min; combine the supernatant with the supernatant obtained for the first time in the same concentrate bottle; and evaporate to near dryness in a 35°C water bath.

5.3 Purification of the specimen

Accurately add 2mL of methanol-acetonitrile-water mixed solution to the residue for reconstitution; sonicate for 10s; vortex for 30s. After the residue is dissolved, add 2mL of acetonitrile saturated n-hexane solution; vortex for 30s; transfer to a 15mL centrifuge tube and centrifuge at 4000r/min for 3min. After discarding the n-hexane layer, add 2 mL of acetonitrile saturated n-hexane solution to the centrifuge tube again; vortex for 30s; centrifuge at 4000r/min for 3min; discard the upper n-hexane layer; take the lower layer and filter through a 0.45um organic microporous filter; the filtrate is reserved for using on the machine.

5.4 Blank test

Except that no sample is added, the determination procedures are carried out in accordance with 5.2 and 5.3.

5.5.1 Reference condition of the instrument

a) Chromatographic column: C18 column, 250mmx4.6mm (i.d.), particle size 5um;

1000 - Unit conversion factor;

m - The weighing amount of the specimen, in g.

The calculation result retains 3 significant digits.

7 Precision The absolute difference between two independent determination results obtained under repeatability conditions shall not exceed 10% of the arithmetic mean.

8 Others When the weight of shrimp and crab is 2.00g, the limit of detection 0.0200mg/kg, and the quantification limit is 0.0500mg/kg.