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NATIONAL STANDARD OF THE PEOPLE'S REPUBLIC OF CHINA

GB 31656.5-2021

National food safety standard - Determination of methaqualone residue in fishery products by liquid chromatography-tandem mass spectrometric method

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Foreword

This document complies with the provisions of GB/T 1:1-2020 "Standardization Work Guidelines Part 1: Structure and Drafting Rules of Standardization Documents"

Drafting:

This document is published for the first time:

National food safety standards

Determination of methaqualone residues in aquatic products by liquid chromatography-tandem mass spectrometry

1 Scope

This document specifies the sample preparation and liquid chromatography-tandem mass spectrometry determination methods for the detection of methaqualone residues in aquatic products:

This document is applicable to the detection of methaqualone residues in edible tissues of fish and shrimp:

2 Normative reference documents

The contents of the following documents constitute essential provisions of this document through normative citations in the text: Among them, the cited documents with dates are:

Only the version corresponding to the date applies to this document; for undated referenced documents, the latest version (including all amendments) applies to this document:

document:

GB/T 6682 Specifications and test methods for water used in analytical laboratories

GB/T 30891-2014 Sampling specifications for aquatic products

3 Terms and definitions

There are no terms or definitions that need to be defined in this document:

4 Principles

The remaining methaqualone in the sample was extracted with n-hexane, purified with a silica gel column, measured by liquid chromatography-tandem mass spectrometry, and quantified by the internal standard method:

5 Reagents and materials

The reagents used below are of analytical grade unless otherwise noted, and the water is first-grade water that complies with GB/T 6682:

5:1 Reagents

5:1:1 Acetone (CH3COCH3): chromatographically pure:

5:1:2 Methanol (CH3OH): chromatographically pure:

5:1:3 Formic acid (HCOOH): chromatographically pure:

5:1:4 n-hexane (C6Hu): chromatographically pure:

5:1:5 Diethyl ether (C2H5OC2H5): chromatographically pure:

5:2 Solution preparation

5:2:1 20% ether-n-hexane eluent: Take 20 mL of ether and 80 mL of n-hexane, and mix well:

5:2:2 50% diethyl ether-n-hexane mixed solution: Take 50 mL of diethyl ether and 50 mL of n-hexane, and mix well:

5:2:3 50% methanol solution: Take 50mL of methanol, add water to 100mL, and mix well:

5:2:4 0:1% formic acid solution: Take 1mL of formic acid, add water to 1000mL, and mix well:

5:3 Reference materials

5:4 Preparation of standard solution

5:4:1:200ng/mL methaqualone standard intermediate solution: Precisely measure an appropriate amount of methaqualone standard solution, dilute it with methanol to prepare a concentration of

200ng/mL standard intermediate solution: Store below -18°C in the dark: Valid for 3 months:

5:4:2 Internal standard working solution: Precisely measure an appropriate amount of methaqualone-D7 standard solution, dilute it with methanol to prepare an internal standard working solution with a concentration of:200ng/mL: Store it below -18°C in the dark and have a validity period of 3 months:

5:5 Materials

5:5:1 Solid phase extraction column: silica gel column, 500mg/3mL, or equivalent performance:

5:5:2 Aqueous polysulfone ether needle filter: 0:22μm:

6 Instruments and equipment

6:1 Liquid chromatography-tandem mass spectrometer: equipped with electrospray ion source:

6:2 Balance: sensitivity is 0:01g and sensitivity is 0:00001g:

6:3 Homogenizer:

6:4 Centrifuge: 6000r/min:

6:5 Nitrogen blower:

6:6 Ultrasonic cleaner:

6:7 Vortex mixer:

6:8 Stoppered glass centrifuge tube: 10mL:

7 Measurement steps

7:1 Preparation of specimens

Prepare samples according to the requirements of Appendix B in GB/T 30891-2014:

a) Take the homogenized test sample as the test material;

b) Take the homogenized blank sample as the blank sample;

c) Take the homogenized blank sample, add an appropriate concentration of methaqualone standard intermediate solution, and add the sample as a blank:

7:2 Storage of samples

Store below -18 degrees C, shelf life is 3 months:

8 Measurement steps

8:1 Extraction

Take 5g of the sample (accurate to +/-0:05g), put it into a 50mL centrifuge tube, add 50μL of internal standard working solution and 3mL of water, vortex and mix for 30s, add

Take 15 mL of n-hexane, extract with oscillation for 3 min, ultrasonic for 5 min, centrifuge at 4500 r/min for 7 min, take the n-hexane layer into a 25 mL volumetric flask; add 8 mL of n-hexane to the residue, repeat the extraction once, combine the n-hexane liquids, and dilute with n-hexane to the mark: , mix well and set aside:

8:2 Purification

Take the solid phase extraction column and activate it with 5 mL of acetone and 5 mL of n-hexane: Precisely measure 10 mL of the reserve solution and pass it through the column at a speed of about 1 per second:

drop, rinse with 5 mL of 20% ether-n-hexane eluent, discard the effluent, and drain: Elute with 8 mL of 50% ether-n-hexane mixed solution:

Collect the eluate and blow dry with nitrogen at 40°C: Add 1 mL of 50% methanol solution to dissolve the residue, and filter it with an aqueous needle filter until the injection is small:

bottle for liquid chromatography-tandem mass spectrometry determination:

8:3 Preparation of standard working curve

Precisely measure appropriate amounts of methaqualone standard intermediate solution and internal standard working solution respectively, and dilute them with 50% methanol solution to prepare the isotope-containing internal standard concentration:

A series of standard working solutions with methaqualone concentrations of 0:4ng/mL, 1:0ng/mL, 5:0ng/mL, 25:0ng/mL and 100:0ng/mL for liquid chromatography-tandem Mass spectrometry: Draw a standard curve using the ratio of the characteristic ion mass chromatographic peak area of methaqualone and the characteristic ion mass chromatographic peak area of the isotope internal standard as the ordinate and the corresponding concentration as the abscissa, and find the regression equation and correlation coefficient:

8:4 Determination

8:4:1 Liquid chromatography conditions

a) Chromatographic column: C18 column (100mmx2:1mm, 5μm) or equivalent;