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NATIONAL STANDARD OF THE PEOPLE'S REPUBLIC OF CHINA

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National food safety standards-Determination of cloprostinol residues in milk by liquid chromatography-tandem mass spectrometry

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10.3 Precision

foreword

This document is in accordance with the provisions of GB/T 1.1-2020 "Guidelines for Standardization Work Part 1.Structure and Drafting Rules for Standardization Documents"

drafting.

This document is published for the first time.

National Food Safety Standards

Determination of cloprostinol residues in milk by liquid chromatography-tandem mass spectrometry

1 Scope

This document specifies the sample preparation and liquid chromatography-tandem mass spectrometry method for the detection of cloprostinol residues in milk.

This document applies to the detection of cloprostinol residues in milk.

2 Normative references

The content in the following documents constitutes the essential provisions of this document through normative references in the text. Among them, the dated reference documents,

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

document.

GB/T 6682 Analytical laboratory water specifications and test methods

3 Terms and Definitions

This document does not have terms and definitions that need to be defined.

4 principles

The residual cloprostinol in the sample was extracted with acetonitrile, purified by mixed anion-exchange solid-phase extraction column, liquid chromatography-tandem mass spectrometry negative ion mode

determined by formula and quantified by external standard method.

5 Reagents and materials

The reagents used in the following are all analytical reagents unless otherwise specified; the water is the first-class water in accordance with the provisions of GB/T 6682.

5.3 Solution preparation

5.3.11 0.1% formic acid solution. take 1mL of formic acid, dilute it with water to 1000mL, and mix well.

5.3.2 5% ammonia solution. take 50mL of ammonia water, dilute to 1000mL with water, and mix well.

5.3.3 2% formic acid in acetonitrile solution. take 20mL of formic acid, dilute to 1000mL with acetonitrile, and mix well.

5.3.4 0.1mol/L ammonium acetate solution. Weigh 7.70g of ammonium acetate, dissolve in water and dilute to 1000mL, mix well.

5.3.5 5mmol/L ammonium acetate solution. take 50mL of 0.1mol/L ammonium acetate solution, dilute with water to 1000mL, and mix well.

5.3.6 Acetonitrile ammonium acetate solution. Take 30mL of acetonitrile, add 5mmol/L ammonium acetate solution to 100mL, mix well.

5.4 Preparation of standard solution

5.4.1 Standard stock solution. take about 10 mg of cloprostenol sodium reference substance, weigh it accurately, add an appropriate amount of acetonitrile to dissolve and dilute to 10 mL

A volumetric flask was prepared to make a standard stock solution of cloprostinol with a concentration of 1 mg/mL. It was stored below -18 degrees C and the validity period was 3 months.

5.4.2 Standard intermediate solution. Accurately measure 0.1mL of standard stock solution, put it in a 10mL volumetric flask, dilute it to the mark with acetonitrile, and prepare a concentration of

10 μg/mL standard intermediate solution. Store at 2°C~8°C, valid for 1 month.

5.4.3 series standard working solution. Accurately measure an appropriate amount of standard intermediate solution, dilute it with ammonium acetate and acetonitrile to prepare a concentration of 5ng/mL,

A series of standard working solutions of 10ng/mL, 20ng/mL, 50ng/mL, 100ng/mL, and

500ng/mL. Ready to use.

6 Instruments and equipment

6.1 High performance liquid chromatography-tandem mass spectrometer. equipped with electrospray ionization source (ESI).

7.1 Preparation of samples

Take an appropriate amount of fresh or thawed blank or test sample, and homogenize.

a) Take the homogeneous test sample as the test sample;

b) Take a homogeneous blank sample as a blank sample;

c) Take a homogeneous blank sample, add standard working solution of appropriate concentration, and add the sample as a blank.

7.2 Storage of samples

Store below -18°C.

8.1 Extraction

Take 2 g of the test material (accurate to +/-0.05 g) in a 50 mL centrifuge tube, add 5 mL of acetonitrile, 2 g of anhydrous sodium sulfate, vortex and mix, and ultrasonically extract

10 min, centrifuged at 10000 r/min at 4 °C for 10 min, the supernatant was taken into a 10 mL centrifuge tube, the extraction was repeated once, and the supernatant was combined twice.

Blow dry at 40°C with nitrogen, dissolve the residue in 1 mL of acetonitrile, add 3 mL of 0.1% formic acid aqueous solution, vortex and mix well, and set aside.

8.2 Purification

Take a mixed anion-exchange solid-phase extraction column, activate it with 3 mL of acetonitrile, and equilibrate with 3 mL of water. Take the spare liquid to pass through the column, and use 5% ammonia solution to

Rinse with 3mL and drain. Elute with 3mL of 2% formic acid acetonitrile solution, collect the eluent, blow dry with nitrogen at 40°C, and wash the residue with 1mL of acetonitrile acetic acid

Ammonium solution was dissolved, filtered with a microporous membrane, and determined by liquid chromatography-tandem mass spectrometry.