ICS 67.120
CCS X22

NATIONAL STANDARD OF THE PEOPLE'S REPUBLIC OF CHINA

GB/T 22338-2008

Determination of multi-residues of chloramphenicols in animal-original food

动物源性食品中氯霉素类药物残留量测定

Issued on: September 1, 2008 **Implemented on: December 1, 2008**

Issued by: General Administration of Quality Supervision, Inspection and Quarantine of the People's Republic of China;
Standardization Administration of China

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Foreword

Annex A, Annex B, Annex C, Annex D and Annex E of this Standard are all informative. This document was proposed by General Administration of Quality Supervision, Inspection and Quarantine of the People's Republic of China. This document shall be under the jurisdiction of the National Certification and Accreditation Administration Commission. The drafting organizations of this document. China Academy of Inspection and Quarantine, Shandong Entry-Exit Inspection and Quarantine Bureau of the People's Republic of China. Main drafters of this document. Qiu Yueming, Lin Liming, Li Peng, Zhang Hongwei, Zhao Haixiang, Dong Yiyang, Cai Huixia, Xie Mengxia, Wang Liping, Kong Ying. Determination of multi-residues of chloramphenicol in animal-original food

1 Scope

GB/T 22338-2008 is the Chinese national standard on determination of multi-residues of chloramphenicols in animal-original food, in the field of food technology. The /T suffix marks it as a recommended standard: it is not compulsory by itself, but it becomes binding as soon as a contract, a tender or a customer specification calls it up - which in practice is how most foreign buyers meet it. It was issued on 1 September 2008 by the General Administration of Quality Supervision, Inspection and Quarantine of the People's Republic of China; Standardization Administration of China, and has been in force since 1 December

2008. Classification: ICS 67.120, CCS X22. This page is published from the official record of the standard held by the Chinese standards administration: the identification, the dates, the classification and the issuing body are taken from there. The clause text, the tables and the numeric limits are in the document itself, which is delivered complete in English translation.

This Standard specifies the methods of gas chromatography-mass spectrometry and liquid chromatography-mass spectrometry/mass spectrometry to determine the multi- residues of chloramphenicol in animal-original food. This Standard is applicable to the qualitative confirmation and quantitative determination of chloramphenicol, florfenicol and thiamphenicol residues in aquatic products, livestock and poultry products and livestock and poultry by-products.

2 Gas chromatography-mass spectrometry

2.1 Principle The sample is extracted by ethyl acetate. It is distributed and purified by 4% sodium chloride solution and n-hexane solution. After purification by Florisil column, take toluene as the reaction medium. Use N,O bis(trimethylsilyl)trifluoroacetamide-trimethylchlorosilane (BSTFA+TMCS, 99+1) to silanize at 70°C. Use gas chromatography/negative chemical ionization source mass spectrometry to determine. Use internal standard working curve method for quantification.

2.2 Reagents and materials Unless otherwise stated, it shall only use the confirmed analytically-pure reagents and secondary deionized water or water of equivalent purity in the analysis.

2.2.1 Methanol. Chromatographically pure.

2.2.2 Toluene. Pesticide residue level.

2.2.3 N-hexane. Pesticide residue level.

2.2.4 Ethyl acetate.

2.2.6 Sodium chloride.

2.2.13 Derivatization reagent. N, O bis (trimethylsilyl) trifluoroacetamide-trimethylchlorosilane (BSTFA+TMCS, 99+1).

2.2.14 Solid phase extraction column. Florisil column (6.0mL, 1.0g).

2.3 Instruments and equipment

2.3.1 Gas chromatography/mass spectrometer. equipped with chemical ionization source (CI).

2.3.2 Tissue masher.

2.3.3 Solid phase extraction device.

2.3.4 Oscillator.

2.3.5 Rotary evaporator.

2.3.6 Vortex mixer.

2.3.7 Centrifuge.

2.3.8 Incubator.

2.4 Determination steps

2.4.1 Extraction Weigh 10g (accurate to 0.01g) of the crushed tissue sample into a 50mL stoppered centrifuge tube. Add 1.0mL of internal standard solution (2.2.11) and 30mL of ethyl acetate. Shake for 30min. Centrifuge at 4000r/min for 2min. Transfer the supernatant to a round bottom flask. Use 30mL of ethyl acetate to extract the residue one more time. Combine the extracts. Conduct rotary evaporation at 35 °C to 1mL ~ 2mL, for purification.

2.4.4.1 Gas chromatography-mass spectrometry conditions

2.4.4.4 Quantitation Use m/z466 (m-CAP and CAP), 339 (FF) and 409 (TAP) as quantitative ions. The response values of chloramphenicol derivatives in the sample solution shall be within the linear range determined by the instrument. Under the above chromatographic conditions (2.4.4.1), the reference retention time of m-CAP, CAP, FF, TAP standard substance derivatives is about 11.4min, 11.8min, 12.6min, 13.6min. For the total ion chromatogram and mass spectrum of chloramphenicol derivatives, see Figure A.1, Figure A.2 in Annex A.

2.4.5 Parallel experiment According to the above steps, carry out parallel test determination on the same specimen.

2.4.6 Blank experiment Carry out according to the above-mentioned determination steps except that no specimen is added.

2.5 Result calculation The result is calculated according to formula (1).

3 Liquid chromatography-mass spectrometry/mass spectrometry

3.1 Principle For residues of chloramphenicol, thiamphenicol and florfenicol in different animal- original food, use acetonitrile, ethyl acetate-ether or ethyl acetate to respectively extract. The extract is purified with a solid phase extraction cartridge.

3.2 Reagents and materials Unless otherwise stated, it shall only use the confirmed analytically-pure reagents and secondary deionized water or water of equivalent purity in the analysis.

3.2.1 Methanol. liquid chromatography level.

3.2.8 Sodium acetate.

3.2.9 Ammonium acetate. 3.2.10 beta-glucuronidase. about 40,000 activity units.

3.2.11 Acetonitrile saturated n-hexane. Take 200mL of n-hexane (3.2.5) into a 250mL separatory funnel. Add a small amount of acetonitrile (3.2.2). Shake vigorously. After standing for stratification, discard the lower acetonitrile layer.

3.2.12 Acetone-n-hexane (1+9). Mix acetone (3.2.3) and n-hexane (3.2.5) well in a volume ratio of 1.9.

3.2.19 Standard stock solution. Accurately and respectively weigh an appropriate amount of chloramphenicol, thiamphenicol and florfenicol standard substances (3.2.17) (accurate to 0.1mg). Use acetonitrile to prepare 500μg/mL standard stock solution (it can be used for 6 months when stored in the dark at 4°C).

3.2.25 LC-Si solid phase extraction cartridge or equivalent. 200mg, 3mL.

3.2.26 EN solid phase extraction cartridge or equivalent. 200mg, 3mL.

3.2.27 Disposable syringe filter. equipped with 0.45μm microporous membrane.

3.3 Instruments and equipment

3.3.1 Liquid chromatography-tandem mass spectrometer. equipped with electrospray ionization source.

3.3.2 High-speed tissue masher.

3.3.3 Homogenizer.

3.3.4 Rotary evaporator.

3.3.5 Analytical balance.

3.3.6 Pipette gun. 200μL, 1mL.

3.3.7 Heart-shaped bottle. 100mL, brown.

3.3.8 Separation funnel. 200mL.

3.3.9 Teflon centrifuge tube. 50mL.

3.3.10 Centrifuge.

3.4.2 Specimen storage The specimen shall be stored at -20°C.

3.5.1 Extraction

3.5.3.4 Quantitative determination Chloramphenicol is quantified by the internal standard method. Thiamphenicol and florfenicol are quantified by the external standard method.