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**NATIONAL STANDARD OF THE
PEOPLE'S REPUBLIC OF CHINA**

GB/T 46279-2025

Replacing GB/T 8381.11-2005

**Determination of amprolium hydrochloride, ethopabate and
sulfaquinoxaline in feeds**

饲料中盐酸氨丙啉、乙氧酰胺苯甲酯和磺胺喹噁啉的测定

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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 replaces GB/T 8381.11-2005 "Determination of Aminopropionate Hydrochloride in Feed by High Performance Liquid Chromatography".

Compared with GB/T 8381.11-2005, apart from structural adjustments and editorial changes, the main technical changes are as follows.

- The scope of application and limit of detection have been changed, and a limit of quantitation has been added (see Chapter 1, Chapter 1 of the 2005 edition);
- The principles of high-performance liquid chromatography have been modified (see Chapter 4, Chapter 3 of the 2005 edition);

- The experimental procedures for high-performance liquid chromatography have been modified (see 4.5, Chapter 7 of the 2005 edition);
- The data processing for high performance liquid chromatography has been modified (see 4.6, Chapter 8 of the 2005 edition);
- A new chapter on "Liquid Chromatography-Tandem Mass Spectrometry" has been added (see Chapter 5).

Please note that some content in this document may involve patents. The issuing organization of this document assumes no responsibility for identifying patents.

This document was proposed and is under the jurisdiction of the National Technical Committee on Standardization of Feed Industry (SAC/TC76).

This document was drafted by: Institute of Agricultural Quality Standards and Testing Technology, Chinese Academy of Agricultural Sciences; Shaanxi Provincial Animal Husbandry Technology Extension Station.

Shaanxi Zhonghe Agricultural Inspection and Testing Co., Ltd., and Shaanxi Provincial Agricultural Inspection and Testing Center.

The main drafters of this document are: Song Rong, Li Hong, Wei Shulin, Yan Hua, Xiao Hongnian, Yang Kun, Chao Juanjuan, Luo Yaxuan, Li Libei, Wang Suqin, and Zhang Hui.

The release history of this document and the document it replaces is as follows.

- First published in 2005 as GB/T 8381.11-2005;
- This is the first revision.

Aminopropionate hydrochloride, ethoxymethyl benzoate and Determination of sulfaquinoline

1 Scope

This document describes high-performance liquid chromatography (HPLC) and liquid chromatography-tandem chromatography (LC-tandem chromatography) for the detection of aminopropionate hydrochloride, ethoxybenzone, and sulfaquinoxaline in feed. Mass spectrometry method.

This document applies to ammonium hydrochloride in compound feed, concentrated feed, concentrate supplements, additive premixes, and mixed feed additives.

Determination of propionyl, ethoxybenzoate and sulfaquinoline.

In this document, the limit of detection (LOD) for amprolium hydrochloride using high-performance liquid chromatography (HPLC) is 10 mg/kg, and the limit of quantitation (LOQ) is 20 mg/kg; the detection limit for ethoxybenzoate is...

The limit of expiratory volume (LOV) was 0.2 mg/kg, and the limit of quantitation (LOQ) was 0.4 mg/kg; the LAV of sulfaquine was 6 mg/kg, and the LOQ was 12 mg/kg. Liquid chromatography (LC) The detection limit for both spectrophotometers and tandem mass spectrometers was 0.02 mg/kg, and the quantitation limit was 0.05 mg/kg.

2 Normative references

GB/T 6682

GB/T 20195

3 Terms and Definitions

This document does not contain any terms or definitions that need to be defined.

4.1 Principle

The analyte in the sample was extracted with methanol solution, defatted with n-hexane, purified by hydrophilic-lipophilic equilibrium solid-phase extraction column, and then separated by high performance liquid chromatography.

The sample was measured using a UV detector in series with a fluorescence detector, and quantified using the external standard method.

4.2 Reagents or Materials

Unless otherwise specified, use only analytical grade reagents.

4.2.1 Water. GB/T 6682, Grade I.

4.2.2 Methanol. chromatographic grade. 4.2.3 n-Hexane.

4.2.4 Extraction solution. a mixture of methanol (4.2.2) and water, $V(\text{methanol}) : V(\text{water}) = 4 : 1$.

4.2.5 Phosphate buffer. Weigh 0.2g potassium dihydrogen phosphate and 13.68g dipotassium hydrogen phosphate trihydrate, dissolve in water and dilute to a final volume. Mix 1000mL thoroughly.

4.2.6 Eluent. Transfer 10 mL of methanol (4.2.2), dilute with water and bring the volume to 100 mL, and mix well.

4.2.7 Sodium heptanesulfonate solution (0.005 mol/L). Weigh 1 g of sodium heptanesulfonate into 800 mL of water, stir to dissolve, and add 6 mL of trichloroethylene (TCM) sodium sulfate solution.

Ethylamine and 24 mL of glacial acetic acid were diluted with water to a final volume of 1000 mL and mixed thoroughly.

4.2.8 Reconstituted solution. a mixture of sodium heptanesulfonate solution (4.2.7) and methanol (4.2.2), $V(\text{sodium heptanesulfonate solution})/V(\text{methanol}) = 1/1$.

4.2.9 Standard stock solution (0.5 mg/mL). Accurately weigh amprolium hydrochloride (CAS No.. 137-88-2, purity not less than 98.0%), ethoxysulfate...

Methylparaben (CAS No.. 59-06-3, purity not less than 98.0%) and sulfaquinoline (CAS No.. 59-40-5, purity not less than 98.0%).

50 mg of each solution (accurate to 0.01 mg) was placed in separate 100 mL volumetric flasks, dissolved and diluted to volume with methanol (4.2.2), and mixed well. The solution was then stored at -18°C.

Stored at the following conditions, the shelf life of the standard stock solutions of amprolium hydrochloride and ethoxybenzoate is 9 months. The standard stock solution of sulfaquinoline is...

The stock solution has a shelf life of 6 months.

4.2.10 Standard intermediate solution (100 µg/mL). Accurately transfer 10 mL of each of the standard stock solutions (4.2.9) into separate 50 mL volumetric flasks.

Dilute with methanol (4.2.2) and bring to volume, then mix well. Store below -18°C; shelf life is 3 months. Ethoxybenzoate Standard Intermediate The solution was diluted 20 times with methanol (4.2.2) (5 µg/mL) before use.

4.2.11 Mixed Standard Series Solutions. Accurately transfer an appropriate amount of the intermediate standard solution (4.2.10) and prepare ampicillin hydrochloride using the reconstitution solution (4.2.8).

The mass concentrations of porphyrin were 1 µg/mL, 2 µg/mL, 5 µg/mL, 10 µg/mL, 20 µg/mL and 50 µg/mL, and the mass of ethoxyamide benzyl ester was...

The concentrations of sulfaquinoline were 0.05 µg/mL, 0.1 µg/mL, 0.25 µg/mL, 0.5 µg/mL, 1.0 µg/mL, and 2.5 µg/mL.

A series of mixed standard solutions with concentrations of 0.6 µg/mL, 1.2 µg/mL, 3 µg/mL, 6 µg/mL, 12 µg/mL, and 30 µg/mL. (Prepared immediately before use) Prepared on the spot.

4.2.12 Solid phase extraction column. hydrophilic-lipophilic balanced column, 60mg/3mL, or equivalent.

4.2.13 Microporous filter membrane. 0.45µm, organic system.

4.3 Instruments and Equipment

4.3.1 Liquid Chromatograph. Equipped with an ultraviolet detector (or diode array detector) and a fluorescence detector, with the fluorescence detector connected in series with the