



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

GB 1886.242-2016

**National Food Safety Standard -- Food Additives -- Ammonium
Glycyrrhetate**

食品安全国家标准 食品添加剂 甘草酸铵

(English Translation)

Issued on: August 31, 2016

Implemented on: January 1, 2017

Issued by: National Health and Family Planning Commission

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1 Scope

This standard applies to the raw material licorice (*Glycyrrhiza legume* *Glycyrrhiza uralensis* Fisch., *Glycyrrhiza inflata* *Glycyrrhiza inflata* Bat. *Glycyrrhiza glabra* and *Glycyrrhiza glabra* L.), by Aqueous Extraction, sulfuric acid (or hydrochloric acid) the precipitate was acidified, ethanol extraction, amide, junction Crystal and other food additive prepared by the process ammonium glycyrrhizinate.

2.1 Chemical Name

Licorice ammonium

2.2 Structure

Formula 2.3

C₄₂H₆₁O₁₆NH₄

2.4 Molecular Weight

839.96 (according to 2007 international relative atomic mass)

3.1 Sensory requirements

Sensory requirements shall comply with the requirements in Table 1.

Table 1 Sensory requirements

Project requires test methods Color white, yellowish white or pale yellow State of the fine particles powder Proper amount of sample is placed in a clean, dry white porcelain plate , Under natural light, the color observed and the state

3.2 Physical and Chemical Indicators

Physical and chemical indicators should be consistent with the provisions of Table 2.

Table 2. Physical and chemical indicators

Item Index Test Method Ammonium glycyrrhizinate content (in terms 18beta- ammonium glycyrrhizinate, dry basis), w /% ≥ 65.0 Appendix A A.4 Drying loss, w /% ≤ 7.0 Appendix A A.3 Specific rotation $[\alpha]_{20}^{\circ} D/[(^{\circ}) \cdot mL \cdot dm^{-1} \cdot g^{-1}]$ 45 ~ 53 in Appendix A A.5 Ash, w /% ≤ 0.5 A.6 Appendix A Lead (Pb)/(mg/kg) ≤ 2.0 Appendix A A.7 Total arsenic (As)/(mg/kg) ≤ 2.0 Appendix A A.8 Appendix A Testing method A.1 General Provisions this standard reagents and water, in the absence of other requirements specified, refer to three analytical reagent water and GB/T 6682 provisions. test Test used in the standard solution, the standard solution impurities, products and preparations, in the absence of other requirements specified, are by GB/T 601, GB/T 602, GB/T 603 Preparation predetermined. The test does not indicate when the solution which is formulated with a solvent, refer to the aqueous solution.

A.2 Identification Test A.2.1 Method summary After met alkali salts to produce ammonia, and volatile alkaline using ammonia wetting characteristics above enables the tube alkaline pH test paper color, Kam Do not salt presence.

A.2.2 Reagents and materials Sodium hydroxide solution A.2.2.1. 40g/L.

A.2.2.2 pH test paper.

A.2.3 identification method Take a sample of about 0.2g ammonium glycyrrhizinate in 10mL test tube, adding about 1mL sodium hydroxide solution (A.2.2.1), shaken, which has been rapidly PH paper wetted with water placed in the tube mouth, and generating volatilized ammonia can wet pH paper color is alkaline.

NOTE: The solution was heated, the color reaction is more obvious.

Determination of loss on drying A.3 A.3.1 Instruments and Equipment A.3.1.1 vacuum drying oven.

A.3.1.2 weighing bottle.

A.3.1.3 dryer.

Procedure A.3.2

The weighing bottle placed in a vacuum oven, $78^{\circ}C \pm 0.5^{\circ}C$, 30min dried under 20mm Hg condition, taken out into the dryer the cooled to room temperature, and weighed. 1g sample was weighed to the nearest 0.0001g, stoppered, weighed. Gently shake the horizontal transverse weighing bottle, Uniformly distributed as a sample layer thickness of not more than 5mm. The plated sample weighing bottle placed in a vacuum oven, open the cork, in at $78^{\circ}C \pm 0.5^{\circ}C$, 20mmHg conditions, dried under reduced pressure 6h, taken into the dryer,

stoppered, allowed to cool to room temperature, and weighed.

A.3.3 calculation results Loss on drying of the mass fraction w_1 , calculated according to formula (A.1).

$$m_1 - m_2$$

$$m \times 100\%$$

Where.

--- M1 plus sample weighed bottle under reduced pressure before drying mass in grams (G);

--- M2 plus sample weighed bottle under reduced pressure after drying mass in grams (G);

--- m sample mass values, in units of grams (g).

The results parallel arithmetic mean subject to measurement results (Reserved 1 decimal place). Obtained in two independent measurements under repeatability conditions Absolute difference determination result is not more than 1.0%.

A.4 Determination of Ammonium glycyrrhizinate content (in terms 18beta- ammonium glycyrrhizinate, dry basis) of A.4.1 Method summary Sample acetonitrile - aqueous solution of filtered, determined by liquid chromatography (UV detector), external standard. When the calculation results, the sample Weigh the sample by subtracting the mass of the sample mass of water, the result is a dry mass fraction y_1 ($C_{42}H_{65}NO_{16}$) meter of ammonium glycyrrhizinate.

A.4.2 Reagents and materials A.4.2.1 Acetonitrile. HPLC grade.

A.4.2.2 glacial acetic acid. excellent pure.

A.4.2.3 0.45 μ m membrane (organic phase).

A.4.2.4 ammonium glycyrrhizinate standards. purity $\geq 90\%$ (HPLC or identification).

Note 1: ammonium glycyrrhizinate standards used in the process is a single component 18beta- ammonium glycyrrhizinate standards.

Note 2: Nonaqueous standard (NT) or other non-HPLC method for determination of the identification, which can not be directly used to calculate the value of the content liquid chromatography content.

A.4.2.5 acetonitrile - aqueous solution. mixing a volume of acetonitrile and water.

A.4.2.6 acetonitrile - acetic acid - solution. acetonitrile acetate ice water (38,161), mixed, and passed through 0.45 μ m filter (organic phase) Filtration, thoroughly degassed.

A.4.3 Instruments

A.4.3.1 HPLC equipped with UV detector.

A.4.3.2 balance, a sense of the amount of 0.01mg.

A.4.3.3 vacuum drying oven.

A.4.4 Analysis step

A.4.4.1 Standard Preparation Weigh 12.5mg ammonium glycyrrhizinate standard, accurate to 0.01 mg, placed in a 25mL volumetric flask with acetonitrile - aqueous solution (A.4.2.5) was dissolved Solution, and dilute to the mark. Filtered through a 0.45μm membrane (organic phase). Using now.

Preparation of sample solution A.4.4.2 Weigh the sample 50.0mg ammonium glycyrrhizinate, accurate to 0.01 mg, placed in 100mL volumetric flask with acetonitrile - aqueous solution (A.4.2.5) was dissolved Solution to volume, filtered through a 0.45μm membrane (organic phase), to be tested.

A.4.4.3 Reference chromatographic conditions A.4.4.3.1 Column. C18 column, length 150mm, internal diameter 4.6mm, particle size 5μm. Or equivalent chromatographic column separation effect.

A.4.4.3.2 mobile phase. acetonitrile - acetic acid - solution (A.4.2.6).

A.4.4.3.3 flow rate. 1.0mL/min.

A.4.4.3.4 Column temperature. room temperature.

A.4.4.3.5 Injection volume. 10μL.

A.4.4.4 Determination Injection of sample solution and standard solution are sequentially ammonium glycyrrhizinate, measured at 254nm, external standard. High performance liquid ammonium licorice and the retention time of the chromatogram see Appendix B.

A.4.5 calculation results Ammonium glycyrrhizinate content (in terms 18β- ammonium glycyrrhizinate, dry basis) the mass fraction w_2 , calculated according to equation (A.2).

$V \times c_s \times A$

$m_3 \times (1-w_1) \times A_s \times$

100% (A.2)

Where.

V --- sample constant volume in milliliters (mL);

CS --- Ammonium glycyrrhizinate standard solution concentration in milligrams per milliliter (mg/mL);

A --- sample solution chromatogram peak area of ammonium glycyrrhizinate;

--- M3 sample quality value, in milligrams (mg);