



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

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**National food safety standard - Determination of T-2 toxin in
foods**

(English Translation)

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Table of Contents

Foreword...	4
1 Scope...	5
2 Principle...	5
3 Reagents and materials...	5
4 Instruments and apparatuses...	6
5 Analysis steps...	7
6 Precision...	10
7 Precision...	10
8 Principle...	10
9 Reagents and materials...	10
10 Instruments and apparatuses...	12
11 Analysis steps...	12
12 Description of the analysis result...	13
13 Precision...	14
14 Others...	14
15 Principles and basis...	14
16 Reagents and materials...	15
17 Instruments and apparatuses...	16
18 Analysis steps...	17
19 Description of the analysis result...	18
20 Precision...	19
21 Others...	20
Appendix A Liquid chromatogram of T-2 toxin standard substance...	21

1 Scope

This Standard specifies methods for the determination of T-2 toxin in foods.

Method 1 of this Standard is applicable to the determination of T-2 toxin content in food and food products, alcohol, soy sauce, vinegar, sauce and sauce products.

Method 2 and Method 3 are applicable to the determination of T-2 toxin in food and food products.

Method 1 - Immunoaffinity chromatography purification liquid chromatography

2 Principle

Use extract to extract T-2 toxin in the sample; after it is purified and derived by the immunoaffinity column, use high-performance liquid chromatography fluorescence detector to measure, and use external standard method to quantify.

3 Reagents and materials

Unless otherwise specified, all the reagents in this method are analytical reagents, the water is grade-1 water that is specified by GB/T 6682.

3.1 Reagents

3.1.1

Methanol (CH₃OH)

Chromatographic pure.

3.1.2

Acetonitrile (CH₃CN)

Chromatographic pure.

3.1.3

Toluene (C₆H₅CH₃)

Chromatographic pure.

3.1.4 4-dimethylaminopyridine.

3.1.5 1-anthrolynitrile, 1-AN.

3.2 Preparation of reagents

4.7 Air pressure pump.

4.8 Test sieve: aperture of 1.0 mm.

4.9 Balance: sensitivity of 0.000 1 g and 0.01 g.

4.10 Ultrasonic generator: power > 180 W.

4.11 Glass syringe: 10 mL.

5.1.1 Food and food products

Grind the sample; for hard food or the like, use a high-speed pulverizer to grind well, and use a test sieve to filter. Weigh 25.0 g (accurate to 0.1g) of sieved sample in a volumetric flask; use extract to fix-volume to 100 mL; transfer it to a homogenizing cup;

use a homogenizer to stir and extract at high speed for 2 min; use a quantitative filter paper to filter it. Pipette 10.0 mL of the filtrate into 40 mL of water; dilute and mix; use a glass-fiber filter paper to filter it until the filtrate is clarified; reserve the filtrate for later use.

5.1.2 Soy sauce, vinegar, sauce and sauce products

Weigh 25.0 g (accurate to 0.1 g) of the mixed sample; use methanol to fix-volume to 50.0 mL; ultrasonically extract it for 10 min; use quantitative filter paper to filter. Pipette 10.0 mL of the filtrate into 40 mL of water; dilute and mix; use a glass-fiber filter paper to filter it until the filtrate is clarified; reserve the filtrate for later use.

5.1.3 Alcohol

Take 20.0 g (accurate to 0.1 g) of degassed alcohol samples (alcohols which contain carbon dioxide, before using, shall be placed in a refrigerator at 4°C for 30 min, and filtered or ultrasonically degassed) or other alcohol samples which do not contain carbon dioxide in a 50 mL volumetric flask; use methanol to fix-volume to the scale;

shake well; use quantitative filter paper to filter. Pipette 10.0 mL of the filtrate into 40 mL of water; dilute; use a glass-fiber filter paper to filter it until the filtrate is clarified;

reserve the filtrate for later use.

5.2 Purification and elution

Connect the immunoaffinity column to a glass syringe; accurately transfer and inject 10 mL of filtrate which is extracted in 5.1 into the glass syringe. Connect the air pressure pump to the glass syringe; adjust the pressure, so that the solution slowly

Where:

y -- the peak area ratio of the target substance;

a -- the slope of the regression curve;

x -- the concentration of the target substance;

b -- the intercept of the regression curve.

The response values of the test substance in both the standard working solution and the sample solution shall be within the linear response range of the instrument; if the sample content exceeds the standard curve range, it needs be diluted before measurement.

5.5 Blank test

Do not weigh the sample; do a blank test according to the steps of 5.1, 5.2 and 5.3. It shall be confirmed that it does not contain any substance that interferes with the to-be-test component.

5.6 Result calculation

Calculate the content of T-2 toxin in the sample according to Formula (2):

Where:

X -- the content of T-2 toxin in the sample, in micrograms per kilogram ($\mu\text{g/kg}$);

ρ -- the concentration of T-2 toxin in the sample solution, in nanograms per milliliter (ng/mL);

V -- the constant volume after derivatization, in milliliters (mL);

1 000 -- unit conversion coefficient;

m -- the weighing sample, in grams (g);

f -- dilution ratio.

Note: The calculation result needs to be deducted from the blank value, and retain two significant figures.

9.1.5 Ethyl acetate ($\text{C}_4\text{H}_8\text{O}_2$).

9.1.6 Dimethylformamide ($\text{C}_3\text{H}_7\text{NO}$).

9.1.7 Tetramethylbenzidine (TMB).

9.1.8 Tween-20 ($\text{C}_{58}\text{H}_{114}\text{O}_{26}$).

9.1.9 30% hydrogen peroxide (30% H_2O_2).

9.1.10 Sodium carbonate (Na_2CO_3).

9.1.11 Sodium bicarbonate (NaHCO_3).

9.1.12 Monopotassium phosphate (KH_2PO_4).

9.1.13 Disodium hydrogen phosphate ($\text{Na}_2\text{HPO}_4 \cdot 12\text{H}_2\text{O}$).

9.1.14 Sodium chloride (NaCl)

9.1.15 Potassium chloride (KCl).

9.1.16 Citric acid ($\text{C}_6\text{H}_8\text{O}_7 \cdot \text{H}_2\text{O}$).

9.1.17 Antibody: a specific monoclonal antibody against T-2 toxin that is produced by