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

GB 31622-2014

**National Food Safety Standard -- Food Additives -- Myrica rubra
red**

食品安全国家标准 食品添加剂 杨梅红

(English Translation)

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1 Scope

This standard applies to Yang Mei (*MynicarubraSied.etZucc*) ripe fruit as a raw material, an ethanol solution by extraction, food industry the resin adsorption purification, and then concentrated, dried to obtain a food additive Yangmei Hong.

2 Formula, Structure and molecular weight

Formula 2.1

Cyanidin -3-O- glucoside. C₂₁H₂₁O₁₁

2.3 Molecular Weight

Cyanidin-3-glucoside. 449.38 (2007 international relative atomic mass press)

3.1 Sensory requirements

Shall comply with the requirements in Table 1.

Table 1 Sensory requirements

Project requires test methods Color purple to red and black Status powder Proper amount of sample is placed in a clean, dry porcelain dish, from the Under natural light, the color observed and the state

3.2 Physical and Chemical Indicators

Shall be in accordance with Table 2.

Table 2. Physical and chemical indicators

Item Index Test Method Color value E1 m (525 ± 5) nm ≥ 40 Appendix A A.4 pH (10g/L

solution) 3.0 ~ 4.5 GB/T 9724 Cyanidin -3-O- glucoside (w) /% ≥ 5 A.4 Loss on drying (w) /% ≤ 18 GB 5009.3 a direct drying method Residue on ignition (w) /% ≤ 6 GB 5009.4 Total arsenic (As)/(mg/kg) ≤ 2 GB 5009.11 Lead (Pb)/(mg/kg) ≤ 3 GB 5009.12 a drying temperature and time were $105^{\circ}\text{C} \pm 2^{\circ}\text{C}$ and 4h.

Appendix A

Testing method

A.1 General Provisions Unless otherwise specified in this standard, the reagents were of analytical grade, standard titration solution used, the impurity determined by standard solution, and made preparations Product, should GB/T 601, GB/T 602, GB/T 603 provisions prepared test water shall comply with GB/T 6682's. The test when the solution does not indicate what formulated with a solvent, refer to the aqueous solution.

A.2 Identification Test A.2.1 maximum absorption peak Sample taken 0.1g, with ethanol - hydrochloric acid solution is dissolved (A.3.1.2) and diluted to 100mL, $525\text{nm} \pm 5\text{nm}$ in the scope of the sample liquid Within a maximum absorption peak.

A.2.2 color reaction Sample taken 0.1g, was dissolved in 50mL of water, the solution was purple red ~, the solution was added sodium hydroxide solution (4.3g sodium hydroxide it was dissolved in 100mL of water), or blue color of the solution turned dark green.

A.3 Color Value Determination A.3.1 Reagents and materials A.3.1.1 ethanol.

A.3.1.2 ethanol - hydrochloric acid solution. Take 4mL hydrochloric acid solution (14), placed in 1000mL volumetric flask with 40% ethanol solution was diluted to volume, shake.

A.3.2 Instruments and Equipment Spectrophotometer.

A.3.3 Analysis step

Sample weighed 0.1g, accurate to 0.0001g, with ethanol - hydrochloric acid to dissolve and dilute to 100mL, shake, and then draw 10mL, Transferred to a 100mL volumetric flask, add ethanol - hydrochloric acid solution to volume, shake. This sample was placed taken 1cm cuvette, to B Alcohol - hydrochloric acid solution as a blank, the absorbance was measured with a spectrophotometer at the wavelength of maximum absorption of $525\text{nm} \pm 5\text{nm}$ (control absorbance should Made between 0.2 and 0.8, the sample concentration should be adjusted or re-measuring the absorbance).

A.3.4 calculation results Color value at the concentration of 1% test sample, a 1cm cuvette, the absorbance measured at the maximum absorption wavelength at $525\text{nm} \pm 5\text{nm}$ of E1 m (525 ± 5) nm meter, according to formula (A.1) is calculated.

$E1\text{ m } (525 \pm 5)\text{ nm} =$

Where.

A --- absorbance of the sample solution measured;

--- C concentration in the sample solution to be tested, expressed in grams per milliliter (g/mL);

100 --- concentration conversion coefficient.

The test result to the arithmetic mean of replicates results. Obtained in two independent determination results under repeatability conditions of absolute difference Value is not more than 10% of the arithmetic mean.

Determination -3-O- glucoside prime A.4 cornflower A.4.1 Reagents and materials A.4.1.1 Acetonitrile. chromatographically pure.

A.4.1.2 hydrochloric acid.

A.4.1.3 formic acid.

A.4.1.4 cyanidin -3-O- glucoside reference substance. purity $\geq 98\%$.

A.4.1.5 methanol. 50%. HPLC grade methanol and an equal volume of water were mixed to homogeneity.

Hydrochloric acid solution A.4.1.6. 1499.

A.4.2 Instruments and Equipment HPLC with UV detector.

A.4.3 Reference chromatographic conditions A.4.3.1 Column. acid type C18 column, 4.6 mm (inner diameter) x 250mm (length), particle size 5 μ m. Or other equivalent column.

A.4.3.2 mobile phase. acetonitrile = water-carboxylic acid (151100).

A.4.3.3 Column temperature. 25 °C.

A.4.3.4 flow rate. 1.0mL/min.

A.4.3.5 injection volume. 20 μ L.

A.4.3.6 Detection wavelength. 515nm.

A.4.4 Analysis step

Preparation of the control solution A.4.4.1 Weighed amount cyanidin -3-O- glucoside reference substance, the nearest 0.0001g, was dissolved with methanol formulated in a concentration of about 0.06mg/mL of cyanidin -3-O- glucoside control solution stored frozen. Prior to assay, this solution was diluted 3-fold with a control solution of hydrochloric acid After backup.

Preparation of sample solution A.4.4.2 Sample weighed 0.1g, accurate to 0.0001g, placed in stoppered Erlenmeyer flask, methanol was added 100mL, sonicated for 10min, with Microporous membrane (0.45 μ m) filtration, frozen. Before and after the measurement, the sample solution was diluted 3-fold with a solution of hydrochloric acid, the standby.

A.4.4.3 system suitability test in reference A.4.3 chromatographic conditions, multiple injections of a control solution was chromatographed separation R should be greater than 1.5, the number of theoretical plates-3-O- cyanidin glucoside calculated to be not less than 2000.

A.4.4.4 Determination in reference A.4.3 chromatographic conditions, respectively, sample solution and control solution for chromatography. Record the chromatogram and a sample solution of According solution chromatogram cyanidin -3-O- glucoside peak area value.

A.4.5 calculation results Cyanidin -3-O- glucoside mass fraction w according to equation (A.2) is calculated.

$A1 \times c \times 3 \times 100$

$A2 \times m \times$

100% (A.2)

Where.

Peak area A1 --- cyanidin -3-O- glucoside chromatogram of the sample solution;

C --- Control solution concentration, in milligrams per milliliter (mg/mL);

3 --- dilution;

100 --- solution volume in milliliters (mL);

Peak area A2 --- control cyanidin -3-O- glucoside solution chromatogram;

--- m sample mass, in milligrams (mg).

The test result to the arithmetic mean of replicates results. Obtained in two independent determination results under repeatability conditions of absolute difference Value is not more than 5% of the arithmetic mean.