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NATIONAL STANDARD OF THE PEOPLE'S REPUBLIC OF CHINA

GB 5413.18-2010

National food safety standard Determination of vitamin C in foods for infants and young children, milk and milk products

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Foreword

This Standard replaces GB/T 5413.18-1997 "Milk powder and formula foods for infants and young children - Determination of vitamin C content".

Compared with GB/T 5413.18-1997, the main changes of this Standard are as follows:

- defined the activity unit of enzyme;
- changed the concentration of o-phenylenediamine solution;
- changed the treatment of specimens containing starch;
- added -- the reaction time after adding boric acid-sodium acetate solution;
- added -- the reaction time after adding o-phenylenediamine solution;

This Standard replaces the following previous standards:

- GB 5413-1985, GB/T 5413.18-1997.

National food safety standard Determination of vitamin C in foods for infants and young children, milk and milk products

1 Scope

This Standard specifies the determination method of vitamin C in foods for infants and young children, milk and milk products.

This standard is applicable to the determination of vitamin C in foods for infants and young children, milk and milk products. The determination result of this Standard indicates the total content of reduction-type vitamin C and oxidation-type vitamin C.

2 Normative references

The following referenced documents are indispensable for the application of this document. For dated references, only the edition cited applies. For undated references, the latest edition of the referenced document (including any amendments) applies.

3 Principle

Vitamin C (ascorbic acid) is oxidized to be dehydroascorbic acid in the presence of activated carbon. The dehydroascorbic acid reacts with o-phenylenediamine to generate fluorescent substances. Use fluorescent spectrophotometer to determine its fluorescence intensity. Its fluorescence intensity is proportional to the concentration of ascorbic acid. Use the external standard method to quantify.

4 Reagents and materials

Unless otherwise specified, the reagents used in the method are analytically pure; the water used is Grade 3 water regulated in GB/T 6682.

- **4.1** Amylase: enzyme activity is 1.5U/mg; adjust the dosage in accordance with the activity unit size.
- **4.2** Metaphosphoric acid-acetic acid solution A: weigh 15 g of metaphosphoric

6 Analysis procedures

6.1 Specimen Treatment

- **6.1.1** Starch-containing specimen: WEIGH about 5 g (accurate to 0.0001 g) of well mixed or about 20 g (accurate to 0.0001 g) of liquid specimen (containing about 2 mg of Vitamin C) in a 150 mL erlenmeyer flask. ADD 0.1 g of amylase (4.1). ADD 50 mL of 45° C $\sim 50^{\circ}$ C distilled water into solid specimen. ADD 30 mL of 45° C $\sim 50^{\circ}$ C distilled water into liquid specimen. After well mixing, USE nitrogen to discharge the air in the flask. COVER it with cork. PLACE it in the 45° C±1°C incubator (5.4) for 30 min. TAKE it OUT to cool to room temperature. USE metaphosphoric acid-acetic acid solution B (4.3) to the 100 mL flask for constant volume.
- **6.1.2** Starch-free specimen: WEIGH about 5 g of well mixed solid specimen (accurate to 0.0001 g). USE metaphosphoric acid-acetic acid A (4.2) to dissolve. MAKE constant volume to 100 mL. OR WEIGH about 50 g of well mixed liquid specimen (accurate to 0.0001 g). USE metaphosphoric acid-acetic acid B (4.3) to dissolve. MAKE constant volume to 100 mL.

6.2 Preparation of solution for test

- **6.2.1** TRANSFER the aforementioned specimens (6.1.1, 6.1.2) and Vitamin C standard solution (4.8) to a 250 mL erlenmeyer flask having about 2 g of acidic activated carbon (4.4). Severely VIBRATE it. FILTER it (discard about 5 mL of initial filtration) and it shall be the filtration of specimen and standard solution. Accurately PIPETTE 5.0 mL of the filtration of specimen and standard solution. Respectively PLACE in the 25 mL and 50 mL flasks having 5.0 mL of boric acid-sodium acetate solution (4.6). Place for 30 min. USE distilled water for constant volume. Use it as the blank solution of specimen and standard solution.
- **6.2.2** Within this 30 min, accurately PIPETTE 5.0 mL of the filtration of specimen and standard solution into another 25 mL and 50 mL flasks having 5.0 mL sodium acetate solution (4.5) and about 15 mL of water. USE water to dilute it to scale. Use it as specimen solution and standard solution.
- **6.2.3** Specimen for test: Respectively and accurately PIPETTE 2.0 mL of specimen solution (6.2.2) and specimen's blank solution (6.2.1) into 10.00 mL test tubes. Accurately ADD 5.0 mL of o-phenylenediamine solution (4.7) into each test tube. Well MIX. PLACE for 60 min under dark conditions for test.
- **6.2.4** Standard series of test solution: Respectively PIPETTE 0.5 mL, 1.0 mL, 1.5 mL and 2.0 mL of the aforementioned solution (6.2.2). Respectively PLACE them into 10 mL test tubes. ADD water to 2.0 mL. At the same time, accurately PIPETTE 2.0 mL of blank solution of standard solution (6.2.1) into a 10 mL test

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