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of The People's Republic of China

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Identification of reconstituted milk in pasteurized and UHT milk

巴氏杀菌乳和 UHT 灭菌乳中复原乳的鉴定

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Foreword

Chapter 4 of this Standard is mandatory, the rest are recommended.

Appendix A and Appendix B of this Standard are informative.

This Standard is proposed by Ministry of Agriculture of the People's Republic of China Ministry.

This Standard is under the jurisdiction of the National Standardization Technical Committee of Animal Husbandry.

Drafting organization of this Standard: Livestock Research Institute of Chinese Academy of Agricultural Sciences.

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Identification of reconstituted milk in pasteurized and UHT milk

1 Scope

This Standard specifies the identification and corresponding determination method of reconstituted milk in pasteurized milk and UHT sterilized milk.

This Standard applies to identification of reconstituted milk in pasteurized milk and UHT sterilized milk.

2 Normative references

The provisions contained in following documents, through the reference of this Standard, become provisions of this Standard. For dated references, subsequent amendments (excluding corrections) or revisions do not apply to this Standard, however, parties who enter into agreement based on this Standard are encouraged to study if the latest versions of these documents are usable. For undated references, the latest versions apply to this Standard.

GB/T 5413.1 Milk powder and formula foods for infant and young children – Determination of protein

GB/T 6682-1992 Water for analytical laboratory use – Specification and test methods

ISO 5538:1987 Milk and milk products – Sampling – Inspection by attributes

3 Terms and definitions

The following terms and definitions apply to this Standard.

3.1 raw milk

The regular milk squeezed from healthy cattle, it shall be cooled, and it may be filtered, however, it is not processed by pasteurization, heat treatment lower than pasteurization, net milk and other sterilization.

5.1.3 Instruments and Equipment

5.1.3.1 Common instruments and equipment in testing laboratory.

5.1.3.2 High performance liquid chromatography, with gradient system and UV-detector.

5.1.3.3 Kjeldahl apparatus.

5.1.3.4 C18 extraction column, 500 mg.

5.1.3.5 Silica gel column: Particle size of 5μm, column diameter of 4.6mm, length of 250 mm.

5.1.3.6 Drier: 110 ºC ± 2 ºC.

5.1.3.7 Heat tubes with screw cap or other heat-sealable tubes: Volume of 20mL.

5.1.3.8 syringe: 10mL.

5.1.4 Sampling

TAKE 250mL of representative sample and SEND to laboratory, the sample shall not be destroyed or changed during transit and storage. Sampling shall be executed with reference to ISO 5538:1987.

5.1.5 Analysis steps

5.1.5.1 Preparation of sample hydrolyzate solution

DRAW 2.00 mL of sample, PLACE in a heat-sealed tube (5.1.3.7), ADD 6mL of 10.6 mol/L hydrochloric acid solution (5.1.2.4), MIX well. Slowly ACCESS high-purity nitrogen (5.1.2.2) into the tube for 1 min - 2 min, SEAL the tube, then PLACE it in drier (5.1.3.6), HEAT at 110 ºC for hydrolysis for 23 h - 24 h. After heating for about 1h, gently SHAKE the tube.

After heating, REMOVE the tube from the drier, COOL it and PERFORM dry-filtration, KEEP the filtrate for measurement.

5.1.5.2 Determination of protein content in sample hydrolyzate solution

DRAW 2.00 mL of sample hydrolyzate solution (5.1.5.1), according to GB/T 5413.1 DETERMINE protein content in the sample solution.

MEASURE 50.0 mL of sample into a 100 mL volumetric flask, USE water to dilute to the mark and then MIX well.

5.2.5.2 Purification

DRAW 10.0 mL of test solution (5.2.5.1) in 50 mL conical flask, sequentially ADD 1.75 mL of ferrous hydroxide solution (5.2.2.6), 1.75mL of zinc sulphate solution (5.2.2.5), and 6.5 mL of buffer A (5.2.2.9). After each solution is added, SHAKE sufficiently and evenly. After all the solutions are added, PLACE for 10 min, FILTER it, DISCARD the first 1mL - 2 mL of filtrate, COLLECT the filtrate.

5.2.5.3 Hydrolyzate lactose and lactulose

DRAW 5.00 mL of filtrate into a 10 mL volumetric flask, ADD 50μL of β-D-galactosidase suspension solution (5.2.2.12), MIX well and COVER it. At 40 ºC water bath or drier INCUBATE for at least 10 h.

5.2.5.4 Glucose oxidation

Sequentially ADD 2.0 mL of Buffer C (5.2.2.11), 100μL of glucose oxidase suspension solution (5.2.2.13), 1 drop of octanol (5.2.2.3), 0.5 mL of sodium hydroxide solution (5.2.2.7) of which the concentration is 0.33mol/L, 50μL of hydrogen peroxide (5.2.2.2), and 0.1 mL of catalase suspension solution (5.2.2.14). When each reagent is added, it shall be shaken well. After all solutions are added, INCUBATE for 3h at 40 ºC in water bath or drier. After cooling, DILUTE to 10 mL, FILTER it, DISCARD the first 1mL - 2 mL of filtrate, COLLECT the filtrate.

5.2.5.5 Blank

According to steps 5.2.5.1 - 5.2.5.4 to process blank solution, except that not adding β-D-galactosidase suspension solution (5.2.2.12).

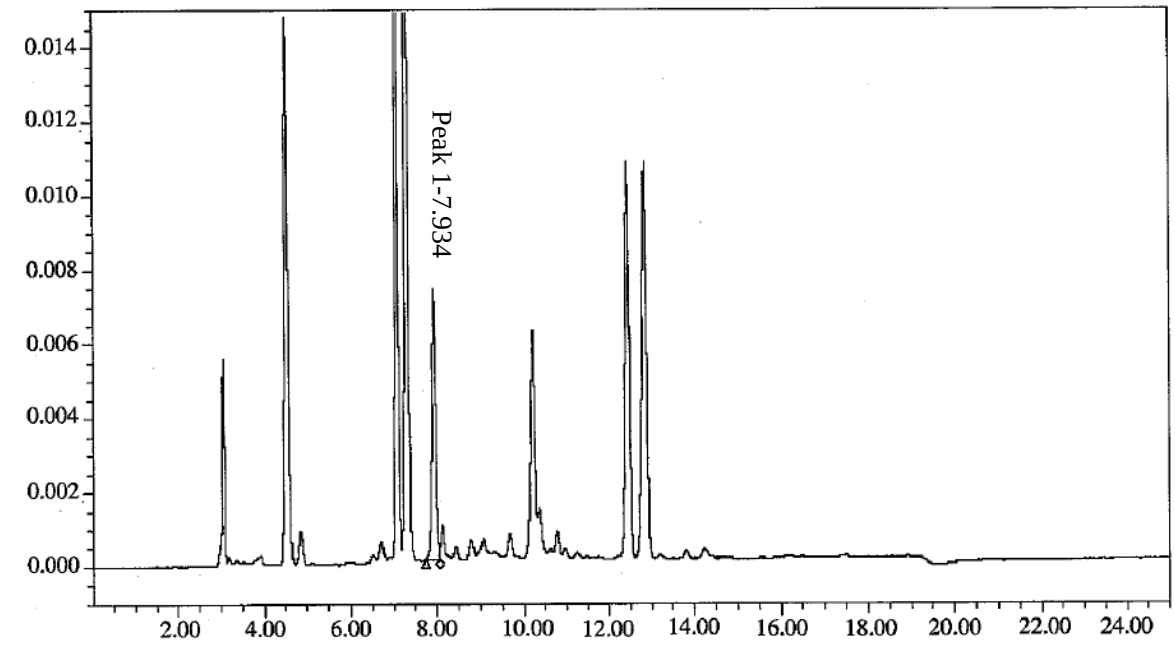
5.2.5.6 Determination

Measurement steps are shown in Table 2.

Appendix B

(Informative)

Separation spectrum of furosine in Pasteurized milk sample



Note: The horizontal axis represents the retention time, in minutes (min); vertical axis represents the absorbance (AU); Peak 1 represents furosine peak.

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