



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

**中华人民共和国国家标准**

GB/T 5413.20-1997  
Replace GB/T 5413-85

---

**Milk powder and formula foods for infant and young  
children-Determination of choline**

**婴幼儿配方食品和乳粉 胆碱的测定**

**Issued on May 28, 1997**

**Implemented on Sept 01, 1998**

---

**Issued by CSBTS (State Bureau of Technical Supervision)**

## Contents

<b>Foreword</b> .....	1
<b>1. Scope</b> .....	1
<b>2. Principle</b> .....	1
<b>3. Reagents</b> .....	1
<b>4. Instruments and devices</b> .....	2
<b>5. Analyzing steps</b> .....	2
<b>5.1</b> Preparation of test sample.....	2
<b>5.2</b> Hydrolysis.....	2
<b>5.3</b> Filtration.....	2
<b>5.4</b> Enzyme reaction.....	2
<b>5.5</b> Colorimetric determination.....	3
<b>5.6</b> Making standard curve.....	3
<b>6. Result calculation and presentation</b> .....	4
<b>6.1</b> Calculation of net light absorption value.....	4
<b>6.2</b> Calculation of choline content.....	5

## **Foreword**

This Standard refers to the methods from *Swiss Nestle*.

This Standard will replace GB 5413.85 since implementation date.

This Standard is proposed by China Light Industry Association.

This Standard is under jurisdiction of *National Technical Committee on Dairy Machinery of Standardization Administration of China*.

Chief draft unit of this Standard are Nestle (China) Investment Services Co., Ltd and National Quality Supervision and Inspection Centre of Dairy Products

Participated draft unit of this Standard are Food Hygiene Supervision and Inspection Institute of MOH, Zhejiang Light Industry Research Institute, Harbin Hiyoko Dairy Co., Ltd.

Chief drafter of this Standard: Zhang Fucai.

# Milk powder and formula foods for infant and young children-Determination of choline

## 1. Scope

This Standard specifies the determination of choline.

This Standard is applicable to milk powder and formula foods for infant and young children-determination of choline.

The detection limit for the method in this standard is 2mg/100g.

## 2. Principle

Choline in the test samples is turned into free choline, and then it is oxidized with enzyme and reacts with developing agent to generate color matter, whose shade is positively proportional to choline content in a certain range.

## 3. Reagents

Unless otherwise provided, in this method, all reagents are analytically pure and the water is third level water as provided in GB/T6682.

### 3.1 0.05mol/L Tris buffer solution (pH 8.0): Phenol containing mass fraction 0.05%

Dissolve 6.057g trihydroxymethyl aminomethane and 0.5g phenol in a 1000ml volumetric flask, which is filled with about 500ml distilled water, and dilute them with distilled water to volume.

The solution may be regulated with 6mol/L hydrochloric acid to pH 8.0.

### 3.2 Color developing agent for enzyme reaction

In a 50ml volumetric flask, dissolve 7.81ml choline oxidase (12.8U/mg), 1.31mg peroxydase (190U/mg), 7.5mg 4- amino-antipyrine and 0.02mg phosphatidase and dilute them with Tris buffer solution (4.1) to volume.

### 3.3 Standard solution: 250 $\mu$ g/ml hydroxide containing choline

Dissolve 523mg choline bitartrate ( $C_5H_{14}NO \cdot C_4H_5O_6$ ) in a 100ml volumetric flask and dilute it with distilled water to volume. Suck 10ml of this solution with a volumetric pipette into a 100ml volumetric flask and dilute it with distilled water to volume.

### 3.4 Sodium hydroxide solution: Mass fraction is 60%.

### 3.5 Hydrochloric acid: c (HCL) is 3mol/L.



北京文心雕语翻译有限公司  
Beijing Lancarver Translation Inc.

---

---

---

## 完整版本请在线下单

或咨询：

TEL: 400-678-1309

QQ: 19315219

Email: [info@lancarver.com](mailto:info@lancarver.com)

<http://www.lancarver.com>

---

---

## 线下付款方式：

### 1. 对公账户：

单位名称：北京文心雕语翻译有限公司

开户行：中国工商银行北京清河镇支行

账 号：0200 1486 0900 0006 131

---

---

### 2. 支付宝账户：[info@lancarver.com](mailto:info@lancarver.com)

---

---

注：付款成功后，请预留电邮，完整版本将在一个工作日内通过电子 PDF 或 Word 形式发送至您的预留邮箱，如需索取发票，下单成功后的三个工作日内安排开具并寄出，预祝合作愉快！

---



银联特约商户