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# NATIONAL STANDARD OF THE PEOPLE'S REPUBLIC OF CHINA

# GB 4789.31-2013

Replacing GB 4789.31-2003

## National Food Safety Standard Food Microbiological Examination -- Determination and testing of Enterobacteriaceae phage of Salmonella, Shigella and Diarrheagenic Escherichia coli

食品微生物学检验 沙门氏菌、志贺氏菌和致泻大肠埃希氏菌的肠杆菌科噬菌体诊断检验

**(English Translation)**

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6 V - - CL vv - Escherichia coli
8 CL - CL - - - - boydii 13a
10.0 G of calcium carbonate
30.0 G of sodium thiosulfate

## Foreword

This standard replaces GB/T 4789.31-2003 "Microbiological examination of food hygiene Salmonella, Shigella and diarrhea caused by Escherichia coli Enterobacteriaceae phage test methods. " this standard compared with GB/T 4789.31-2003, main changes are as follows.

- Modify the procedure;

- Added Appendix B.

National Food Safety Standard Food Microbiology Inspection Salmonella, Shigella and diarrhea caused by E. coli Enterobacteriaceae phage diagnostic tests

## 1 Scope

This standard specifies the application Enterobacteriaceae phage diagnostic method for testing of Salmonella, Shigella and diarrhea caused by Escherichia coli.

This standard applies to all types of foods and foodborne disease events samples Salmonella, Shigella and diarrhea caused by Escherichia coli test.



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This standard applies to the food industry practitioners intestinal Salmonella and Shigella carrier inspection.

## 2 Equipment and Materials

In addition to conventional microbiological laboratory sterilization, training equipment, other equipment and materials as follows.

a) incubator. 36 °C ± 1 °C, 42 °C ± 1 °C, 50 °C ± 1 °C;

b) anaerobic means. 41.5 °C ± 0.5 °C;

c) Microscope. 10 to 100 times;

d) Level. bacteriophage test work surface should be adjusted to a horizontal position;

e) 1 mL sterile disposable syringes (plastic material should be), the needle-free number before using measuring 5 L ~ 10 L per drop (per 100 mL dropwise to 200) are available;

f) Micropipettes and tips (10 L and 5 L);

g) and quantitative pipette tip (100 L and 50 L);

h) sterile pipette. 10 mL, 5 mL, 1 mL;

i) sterile capillary;

j) sterile Petri dish. 90 mm;

k) a sterile cotton swab;

l) with lids sterilized small test tube. 8 mm x 50 mm;

m) loading slide.

## 3 Media and reagents

**3.1**
selenite cystine (SC) enrichment broth. See Appendix A for A.1.

**3.2**
four brilliant green, sodium sulfur (TTB) enrichment broth. See Appendix A A.2.

**3.3**
Shigella (BCT) enrichment broth. See Appendix A A.3.

**3.4**
nutrient broth (NB). See Appendix A in A.4.



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**3.5**
nutrient agar (NA). A A.5 in the Appendix.

**3.6**
semi-solid nutrient agar (NSA). See Appendix A in A.6.

**3.7**
triple sugar iron agar (TSI). See Appendix A A.7.

**3.8**
dextrose agar ammonium. See Appendix A A.8.

**3.9**
diagnosis Enterobacteriaceae phage, 7 and 3. If necessary, the purchase of Escherichia coli phage typing (six kinds) set. Secure After BU open with a sterile capillary aspiration moved within sterilized small test tube, if the amount of each use small, can be divided into 2 or 3 installed in the test tube and stored at 5 °C refrigerator spare.

**3.10**
Salmonella factor serum, Shigella typing serum factor, causing diarrhea Escherichia coli typing serum factor. Category serum factor Depending on need.

## 4.1 Salmonella test procedure

Salmonella test procedure shown in Figure 1.

Figure 1 salmonella testing procedures

## 4.2 Shigella inspection procedures

Shigella inspection procedures shown in Figure 2.

ONPG mannitol, sorbitol, Test sample O-1 cleavage Other not cleaved Non-described as left Various reactions result salmonella Serological test confirmed Non-Salmonella Salmonella Serological test confirmed salmonella Serological test confirmed Non-non-Salmonella Salmonella report Pre-enrichment, enrichment and separation Suspected colonies were picked TSI (slant, bottom gas, H2S) Peptone water (indole) Urea (pH7.2), KCN, lysine H2S, indole -, Urea -, KCN-, Lysine + Phage test H2S, indole, Urea -, KCN-, Lysine Various phage not cleaved Non-described as left the various cracking Results H2S-, indole -, Urea -, KCN-, Lysine/-+, +, - -, -

Figure 2 Shigella inspection procedures

Sh cleavage and exhibits the various cracking mode Sh not cleaved Serological tests consistent with cleavage pattern and lysed 1 RTD Sh Serological tests and split mode does not match or may not be cleaved 1 RTD Sh Biochemical grouping test Ammonium glucose



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test If necessary, a plus sodium acetate, sodium citrate Clinton, Mucus acid, esculin, salicin test Esculin \\ salicin test Shigella Clustering and classification results Glucose ammonium or any other positive results Non-Shigella report Non-Shigella Non-Shigella Phage test Non-referred as left Species Reactivity Results Test sample Suspected colonies were picked TSI, glucose semisolid Enrichment and separation TSI underlying acid production ramp production base, H2S-no gas, no power

### 4.3 Diarrheogenic Escherichia coli testing procedures

Diarrhea caused by Escherichia coli testing procedures shown in Figure 3.

Figure 3 diarrhea caused by Escherichia coli testing procedures

Test sample

And E or Sh, E-4

Phage not cracking

Other phage lysis

Strains isolated from different sources its cleavage pattern have identity TSI, indole, pH7.2 Urea, KCN, Lysine, power TSI bottom +, H2S-, KCN-, urea -Non-described as left Various reactions result Serological tests ETEC Enrichment and separation and E or Sh, E-4 Phage lysis Various phage are not cleaved Oxidase -, G- bacilli EPEC Enterotoxigenic Escherichia coli Non-colon Coli Lysine -, indole, Power - (O124 power/-) Enterotoxin Non-described as left EIEC Various reactions result Non-colon Coli Large non-induced diarrhea Escherichia coli Intestinal invasive Escherichia coli Enteropathogenic Escherichia coli Lactose fermentation or unfermented 3 to 5 colonies Phage test STEC (O157) stx1, stx2, hly Shiga toxin Escherichia coli eae report

### 5.1 Pre-enrichment, enrichment and separation

#### 5.1.1

Salmonella pre-enrichment, enrichment and separation carried out according to GB 4789.4. Salmonella bacteria by measuring sensitivity in Appendix B. in Intestinal colonization of Salmonella in the food industry practitioners test specimens collected should enrichment, should not be pre-enrichment. Samples must not be food poisoning before Enrichment, samples collected at the same time do not enrichment and enrichment step.

Enrichment and separation 5.1.2 Shigella performed according to GB 4789.5. No laboratory anaerobic culture conditions can be used in Appendix A BCT Enrichment broth. Food poisoning patient stool samples at the same time do not enrichment and enrichment step.



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