

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

HJ 1000-2018

**Water quality - Determination of total bacteria - Plate count
method**

水质 细菌总数的测定 平皿计数法

(English Translation)

Issued on: December 26, 2018

Implemented on: June 1, 2019

Issued by: Ministry of Ecology and Environment

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Foreword

To protect the ecology of the People's Republic of China Environmental Protection Law and the Law of the People's Republic of China on Water Pollution Prevention and Control the environment, the human health, the method for determining the total number of bacteria in the water, and the development of this standard.

This standard specifies the plate counting method for determining the total number of bacteria in surface water, groundwater, domestic sewage and industrial wastewater.

Appendix A of this standard is an informative annex.

This standard is the first release.

This standard is formulated by the Department of Eco-Environmental Monitoring and the Department of Regulations and Standards of the Ministry of Ecology and Environment.

This standard was drafted. Liaoning Provincial Environmental Monitoring Experimental Center.

This standard is verified by. Dalian Environmental Monitoring Center, Dandong Environmental Monitoring Center Station, Jinzhou Environmental Monitoring Center Station, Liaoyang Environmental Monitoring Station, Shenyang Municipal Center for Disease Control and Prevention and Liaoning North Environmental Testing Technology Co., Ltd.

This standard is approved by the Ministry of Ecology and Environment on December 26, 2018.

This standard has been implemented since June 1, 2019.

This standard is explained by the Ministry of Ecology and Environment.

Determination of the total number of bacteria in water

1 Scope of application

This standard specifies the plate counting method for determining the total number of bacteria in water.

This standard applies to the determination of the total number of bacteria in surface water, groundwater, domestic sewage and industrial wastewater.

2 Normative references

This standard refers to the following documents or their terms. For undated references, the valid version applies to this standard.

GB/T 14581 Water quality lake and reservoir sampling technical guidance HJ 494 Water Quality Sampling Technical Guidance HJ/T 91 Surface Water and Wastewater Monitoring Technical Specifications

3 Terms and definitions

The following terms and definitions apply to this standard.

Total bacteria total bacteria the total number of aerobic and facultative anaerobic colonies grown on nutrient agar was 48 h at 36 °C.

Colony forming unit colony-forming units (CFU) the total number of bacterial communities

per unit volume of sample.

4 Principle of the method

The sample was inoculated on a nutrient agar medium and cultured under specific physical conditions (cultured at 36 ° C for 48 h).

The total number of aerobic and facultative anaerobic bacteria is the total number of bacterial colonies in the sample.

5 Interference and elimination

5.1

Active chlorine is oxidizing and can destroy the enzyme activity in microbial cells, leading to cell death, which can be collected in samples (8.1) Add sodium thiosulfate solution (6.5) to eliminate interference.

5.2

Heavy metal ions are cytotoxic and can destroy the enzyme activity in microbial cells, leading to cell death.

Add (VIII) Ethylenediaminetetraacetic acid disodium solution (6.6) to eliminate interference.

6.2 Reagents and materials Analytically pure reagents or biological reagents that meet national standards are used for analysis, unless otherwise stated.

Water or deionized water.

6.1 Nutritional agar medium.

Dissolve the above ingredients or a commercially available product containing the above ingredients in 1000 ml of water, adjust the pH to 7.4 to 7.6, and dispense in a glass container, autoclaved at 121 ° C for 20 min, stored in a cool dark place. Protect from light, dry, if necessary Store in a refrigerator at 5 ° C ± 3 ° C for no more than 1 month. The prepared nutrient agar medium cannot be subjected to multiple melting operations.

It is advisable to use a small amount of work. It should be discarded when the color of the medium changes or the dehydration is obvious.

6.2 Sterile water. Take appropriate amount of experimental water and sterilize it by autoclaving at 121 °C for 20 min.

6.3 Sodium thiosulfate ($\text{Na}_2\text{S}_2\text{O}_3 \cdot 5\text{H}_2\text{O}$).

6.4 Disodium edetate ($\text{C}_{10}\text{H}_{14}\text{N}_2\text{O}_8\text{Na}_2 \cdot 2\text{H}_2\text{O}$).

6.5 Sodium thiosulfate solution. (Na₂S₂O₃) =0.10 g/ml

Weigh 15.7 g of sodium thiosulfate (6.3), dissolve in an appropriate amount of water, and dilute to 100 ml.

6.6 Ethylenediaminetetraacetic acid disodium solution. (C₁₀H₁₄N₂O₈Na₂·2H₂O) =0.15 g/ml

Weigh 15 g of disodium edetate (6.4), dissolve in an appropriate amount of water, and dilute to 100 ml. This solution can be stored for 30 days.

6.7 Glass beads. 3 to 8 mm in diameter.

7 Instruments and equipment

7.1

Sampling bottle. 250 ml wide-mouth glass bottle with screw cap or grinder.

7.2

High pressure steam sterilizer. 115 ° C, 121 ° C adjustable.

7.3

Constant temperature incubator. Allowable temperature deviation of 36 °C ± 1 °C.

7.4

Constant temperature water bath. 47 °C adjustable.

7.5

pH meter. Accurate to 0.1 pH unit.

7.6

Magnifier or colony counter.

7.7

Common instruments and equipment used in general laboratories.

Note: Glassware and sampling equipment should be wrapped according to aseptic operation before testing, and autoclaved at 121 °C for 20 min.

8.1 Sample Collection

The point layout and sampling frequency are carried out in accordance with the relevant provisions of GB/T 14581, HJ/T 494 and HJ/T 91.

When collecting microbial samples, the sampling bottle (7.1) should not be washed with the sample, and the sample should be collected in a sterile sampling bottle. 3 when collecting