



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

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**National food safety standard - Food Microbiology Testing -
Quality Requirements for Medium and Reagents**

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1 Scope

This standard specifies the quality requirements for culture media and reagents used in food microbiology testing.

This standard applies to the quality control of culture media and reagents for food microbiology testing.

2.1 Quality control

Technical operations and activities undertaken to meet quality requirements.

2.2 Lots of media and reagents

The complete traceability unit of culture media and reagents, which refers to a certain number of products produced in a specific production cycle and meeting production quality requirements. These products have the same batch number.

2.3 Performance of media and reagents

The reaction of culture media and reagents to the test strain under certain conditions.

2.4 Culture medium

Substances in liquid, semi-solid or solid form, containing natural or synthetic ingredients, used to ensure the reproduction, identification or maintenance of vitality of microorganisms.

2.5 Liquid culture medium

A culture medium consisting of an aqueous solution containing one or more components (e.g. buffered peptone water).

2.6 Solid media

A culture media, which is formed by adding certain amount of solidified material (such as agar) to the liquid culture medium, after boiling or moist heat sterilization, cooled to room temperature and solidified.

Solid culture medium poured into a petri dish is generally called a "plate"; solid culture medium poured into a test tube and placed at an angle is often called a "slant", when the culture medium solidifies.

2.7 Semi-solid medium

A culture media, which is formed by adding a small amount of solidified matter (such as agar) to the liquid culture medium, after boiling or moist heat sterilization, semi-solidified after cooling.

2.8 Transport media

A culture medium, that protects and maintains microbial activity after sampling and before laboratory processing, AND does not allow obvious proliferation or reduction of microorganisms.

The transport medium usually does not contain substances that allow microorganisms to proliferate, but the medium shall protect the strains (e.g., buffered glycerol-sodium chloride solution transport medium).

3.1.1 Documents provided by the manufacturer

The manufacturer shall provide the following information (electronic text can be provided).

3.2.1 General requirements

Culture media and reagents shall be stored and used, in strict accordance with the storage conditions, expiration date, usage methods provided by the supplier.

3.2.2 Commercial dehydrated synthetic culture medium and its additives

Acceptance shall include the following.

3.2.3 Commercial ready-to-use media and reagents

It shall be stored and used, in strict accordance with the storage conditions, validity period, usage methods provided by the supplier.

3.2.4 Laboratory-made media

Store it under conditions that ensure that its ingredients will not change, that is, avoid light and dry storage. If necessary, store it in a refrigerator at 2 °C ~ 8 °C. It is usually recommended that the storage period of plates shall not exceed 4 weeks; the storage period of bottled and test tube media shall not exceed 6 months.

If it needs to add unstable additives to the culture medium, they shall be prepared and used immediately. The solid culture media containing active chemicals or unstable components shall also be prepared and used immediately, which shall not be melted twice.

3.3.1 General requirements

The correct preparation of culture media is one of the most basic steps in microbiological testing. When using dehydrated culture media and other ingredients, especially those containing toxic and harmful substances (such as bile salts or other selective agents), good laboratory practices and instructions for use provided by manufacturers shall be followed. Improper preparation of culture medium can lead to quality problems in the culture medium (see Appendix B).

When using commercial dehydrated synthetic medium to prepare culture medium, the preparation shall be strictly in accordance with the instructions provided by the manufacturer, such as mass (volume), pH, preparation date, sterilization conditions, operating procedures, etc.

3.3.2 Water

The conductivity of the test water shall not exceed 25 $\mu\text{S}/\text{cm}$ (equivalent to resistivity $\geq 0.04 \text{ Mohm}/\text{cm}$) at 25 °C, unless otherwise specified.

The microbial contamination of test water shall not exceed 103 CFU/mL. Microbial contamination levels shall be checked regularly according to GB 4789.2.

3.4.1 Melting of agar medium

Melt the culture medium by placing it in a boiling water bath or using an equivalent method (such as laminar steam in a pressure cooker). Media after high-pressure treatment shall be reheated to a minimum. The melted culture medium is placed in a constant temperature device, at 47 °C ~ 50 °C, for cooling and insulation (the temperature of the constant temperature device can be appropriately increased, according to the actual culture medium solidification temperature) until use. The time for the culture medium to reach 47 °C ~ 50 °C is related to the variety, volume, quantity of the culture medium. The melted culture medium shall be used as soon as possible and shall not be left for more than 4 hours. Unused culture medium cannot be re-solidified for next use.

4.1 General requirements

In order to successfully preserve and use strains, different preservation methods shall be used for different strains. It can choose to use freeze-drying preservation, ultra-low temperature (not higher than -70 °C) freezing preservation, liquid nitrogen preservation or other effective preservation methods.

4.2 Commercially sourced test strains

For original packaged test strains obtained from a standard strain preservation center or other validly certified commercial agency, recovery and use shall be carried out in accordance with the instructions provided by the manufacturer.

4.3 Standard stock strains prepared in the laboratory

When storing and using the standard reserve strain, which is used for performance testing (see Figure C.1 in Appendix C), care shall be taken to avoid cross-contamination and reduce strain mutations or typical characteristic changes. It shall prepare multiple copies of the standard reserve strain; it shall be stored at ultra-low temperature (not higher than -70 °C) or in freeze-dried form. Storage times at higher temperatures shall be shortened.

4.4 Reserve strains

Reserve strains are usually prepared from standard reserve strains that are freeze-dried or cryopreserved (see Figure C.2).

4.5 Working strains

Working strains are prepared from standard reserve strains or reserve strains.

Working strains cannot be used to make standard strains, standard reserve strains, or reserve strains.