ICS 11.220
CCS B 41

NATIONAL STANDARD OF THE PEOPLE'S REPUBLIC OF CHINA

GB/T 18652-2025

Replacing GB/T 18652-2002

Methods for detection of pathogenic Aeromonas hydrophila

致病性嗜水气单胞菌检验方法

Issued on: August 29, 2025 **Implemented on: March 1, 2026**

Issued by: State Administration for Market Regulation;
Standardization Administration of the PRC

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Foreword

This document complies with the provisions of GB/T 1.1-2020 "Standardization Work Guidelines Part 1: Structure and Drafting Rules of Standardization Documents". Drafting.

This document replaces GB/T 18652-2002 "Test Methods for Pathogenic Aeromonas Hydrophila". Compared with GB/T 18652-2002, except for...

Aside from structural adjustments and editorial changes, the main technical changes are as follows.

a) The scope has been changed (see Chapter 1, Chapter 1 of the 2002 edition);

b) A new chapter on "Abbreviations" has been added (see Chapter 4);

c) A new chapter on "Instruments and Equipment" has been added (see Chapter 5);

d) A new chapter, "Reagents and Materials," has been added (see Chapter 6);

e) Added "Sample Collection, Transportation, and Preservation" (see Chapter 8);

f) A PCR identification assay method has been added (see 10.2);

g) The dotted enzyme-linked immunosorbent assay (ELISA) has been removed (see 2.3.2 in the 2002 edition);

h) A blood plate test has been added (see 11.2);

i) A comprehensive judgment has been added (see Chapter 12).

Please note that some content in this document may involve patents. The issuing organization of this document assumes no responsibility for identifying patents.

This document was proposed by the Ministry of Agriculture and Rural Affairs of the People's Republic of China.

This document is under the jurisdiction of the National Technical Committee on Animal Health Standardization (SAC/TC181).

This document was drafted by Nanjing Agricultural University and the China Academy of Inspection and Quarantine Sciences.

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The release history of this document and the document it replaces is as follows.

- First published in 2002 as GB/T 18652-2002;

- This is the first revision.

Introduction

The pathogen can infect fish, amphibians, reptiles, birds, mammals, and other animals, causing acute septicemia or localized infections such as skin ulcers.

In aquaculture, pathogenic Aeromonas hydrophila is a major cause of bacterial septicemia in freshwater fish. Human infection with pathogenic Aeromonas hydrophila...

The main manifestation of the bacteria is acute gastroenteritis.

The pathogenicity of Aeromonas hydrophila is closely related to the virulence factors it produces, among which extracellular products such as toxins and proteases are pathogenic.

It plays an important role in the process. The toxins produced by *Aeromonas hydrophila* mainly include aerolysins, hemolysins, cytotoxic enterotoxins, and cell excitoxins.

Enterotoxins, which are perforating toxins, are very similar in structure and function, possess the same biological activities, including hemolysis.

Enterotoxicity and cytotoxicity. Aeromonas hydrophila also secretes various proteases that

degrade casein, elastin, etc., causing direct damage to tissues.

Injury is also a very important virulence factor. Therefore, all Aeromonas hydrophila that are hemolytic or protease-positive are pathogenic.

Detecting the hemolytic activity and/or protease activity of strains can identify the pathogenicity of Aeromonas hydrophila.

1 Scope

This document describes the methods for isolating, purifying, physicochemically identifying, and PCR identifying Aeromonas hydrophila, as well as the detection of pathogenic Aeromonas hydrophila.

Methods for identifying skim milk plate and blood plate tests.

This document applies to the testing of Aeromonas hydrophila and pathogenic Aeromonas hydrophila.

2 Normative references

GB/T 18088

3 Terms and Definitions

This document does not contain any terms or definitions that need to be defined.

4 Abbreviations

The following abbreviations apply to this document. gyrB. DNA gyrase B subunit TAE. Tris-aceticacid-EDTA buffer

5 Instruments and Equipment

5.1 Constant temperature incubator. 28°C±1°C.

5.2 Dry constant temperature metal bath.

5.3 High-speed low-temperature centrifuge. rotation speed not less than 12000 r/min.

5.4 Level 2 biosafety cabinet or clean bench.

5.5 PCR amplification instrument. 5.6 Gel imaging system.

5.7 Micropipette. Capacity range 1μL~10μL, 20μL~200μL.

5.8 Nucleic acid electrophoresis apparatus. 5.9 Autoclave.

5.10 Refrigerator (4°C, -20°C or below).