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SN

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

SN/T 1699.1-2006

Replacing SN/T 1699.2-2006

Quarantine protocol for porcine epidemic diarrhea

猪流行性腹泻微量血清中和试验操作规程

(English Translation)

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Foreword

This standard was drafted in accordance with the rules given in GB/T 1.1-2009.

This standard replaces SN/T 1699.1-2006 "Microseline Neutralization Test Procedures for Porcine Epidemic Diarrhea", SN/T 1699.2-2006 "Directive immunofluorescence test protocol for swine epidemic diarrhea virus", SN/T 1699.3-2006 "Indirect spot enzyme coupling for pig epidemic diarrhea" Operational Procedures for Epidemic Adsorption Tests.

Main technical changes compared with SN/T 1699.1-2006, SN/T 1699.2-2006, SN/T 1699.3-2006 as follows.

- Increased the method for isolation and identification of porcine epidemic diarrhea virus;
- Added reverse transcription-polymerase chain reaction method;
- Added real-time fluorescence reverse transcription polymerase chain reaction method;
- Increased the enzyme-linked immunosorbent assay method, replacing the indirect dot enzyme-linked immunosorbent assay protocol;
- Revised the direct immunofluorescence test protocol, adopting NY/T 544-2015 "Diagnostic Techniques for Porcine Epidemic Diarrhea" 6.2 "Direct immunofluorescence method" and detailed description of its operation steps;
- Revised the micro-sera neutralization test protocol, recommended cell lines, virus proliferation and toxic rate determination, control settings, etc.

The content has been revised and supplemented.

This standard is proposed and managed by the National Certification and Accreditation Administration.

This standard was drafted. Guangdong Entry-Exit Inspection and Quarantine Bureau of the People's Republic of China, South China Agricultural University, Shenzhen, People's Republic of China Entry Inspection and Quarantine Bureau.

1 Scope

This standard specifies the isolation and identification of porcine epidemic diarrhea, reverse transcription polymerase chain reaction, real-time fluorescent reverse transcription polymerase chain Laboratory quarantine specifications for reactions, direct immunofluorescence assays, enzyme-linked immunosorbent assays, and microserum neutralization assays.

This standard is applicable to the quarantine, epidemic monitoring and diagnosis of epidemic diarrhea in pigs entering and leaving the country.

2 Normative references

The following documents are indispensable for the application of this document. For dated references, only the dated version applies to this article.

Pieces. For undated references, the latest edition (including all amendments) applies to this document.

GB/T 6682 Analytical laboratory water specifications and test methods

3 Abbreviations

The following abbreviations apply to this document.

PED. Porcine epidemic diarrhea (porcineepidemicdiarrhea) PEDV. Porcine epidemic diarrhea virus (porcineepidemicdiarrheavirus) PBS. phosphate buffer (phosphatebuffersaline) Vero. African Green Monkey Kidney Cells DMEM. an improved cell culture fluid (dulbecco'smodifiedeaglemedium) CPE. cytopathic effect RT-PCR. reverse transcription-polymerase chain reaction (reversetranscription-polymerase chain reaction) ddH₂O. double distilled water (doubledistilledwater) DEPC. diethylpyrocarbonate BSA. bovine serum albumin (bovineserumalbumin) Ct. number of cycles experienced when the fluorescence signal reaches the set threshold FITC. fluorescein isothiocyanate ELISA. enzyme-linked immunosorbent assay (enzyme-linked immunosorbent assay) TMB. 3,3',5,5'-tetramethylbenzidine (3,3',5,5'-Tetramethylbenzidine) OD. optical density (optical density) TCID₅₀. half tissue culture infection dose (tissuecultureinfecteddose)

4 Overview of porcine epidemic diarrhea

Porcine epidemic diarrhea (PED) is a porcine epidemic diarrhea virus (PEDV) from Coronaviridae, Coronavirus an acute highly exposed swine intestinal infectious disease with clinical manifestations of watery diarrhea, vomiting, dehydration, exercise stiffness and high death in suckling piglets Characteristics such as death rate. Pigs of all ages can be infected, with lactating piglets at 1 week of age being the most susceptible. PEDV mainly

through infected pigs the spread of feces or other viral contaminants, under natural conditions, PEDV enters the pig's body through the oral and nasal routes. The prevalence of PED