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INDUSTRY STANDARD OF THE PEOPLE'S REPUBLIC OF CHINA

SN/T 3584-2013

Quarantine protocol for hemorrhagic disease of grass carp

草鱼出血病检疫技术规范

(English Translation)

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Preview: page 1 of 3 shown, 9 in the full document. Complete English PDF, instant download:
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Table of Contents

Foreword

1 Scope

2 Normative references

3 Abbreviations

4 Disease Overview

Foreword

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

The standard reference to the Asia-Pacific Aquaculture Network Center (NACA) disease diagnostic card "Diseasecard" in the "grass carp hemorrhage (Hemorrhagicdiseaseofgrasscarp) ". Using diagnostic card recommended cell culture isolation, RT-PCR method, specific details of the ELISA method, and it increases the direct observation of viral nucleic acids by electrophoresis band approach.

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

This standard was drafted. Chinese Academy of Inspection and Quarantine, Beijing People's Republic of China Exit Inspection and Quarantine, the People's Republic of China Exit Inspection and Quarantine and the State Hunan.

1 Scope

This standard specifies the method for detection of grass carp hemorrhage laboratory.

This standard applies to grass carp hemorrhage identified pathogen, for epidemiological investigation, diagnosis, and monitoring of quarantine.

2 Normative references

The following documents for the application of this document is essential. For dated references, only the dated version suitable for use herein Member. For undated references, the latest edition (including any amendments) applies to this document.

Laboratory use specifications and test methods GB/T 6682 Analysis

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3 Abbreviations

The following abbreviations apply to this document.

CO. grass carp ovary cell line (Grasscarpovarycelline) CIK. grass carp kidney cell line (Grasscarpkidneycelline) CPE. cytopathic effect (Cytopathiceffect) EB. ethidium bromide (Ethidiumbromide) DEPC. diethyl pyrocarbonate (Diethypyrocarbonate) PCR. polymerase chain reaction (Polymerasechainreaction) ELISA. enzyme-linked immunosorbent assay (Enzyme-linkedimmunosorbentassay) PAGE. polyacrylamide gel electrophoresis (polyacrylamidegelelctrophoresis) HEPES. N-2-Hydroxyethylpiperazine-N-2-ethanesulphonicacid TMB. tetramethylbenzidine (3,3 ', 5,5'-Tetramethylbenzidine) OPD. o-phenylenediamine (O-PhenylenediamineDihydrochloride) SDS. sodium dodecyl sulfate (Sodiumdodecylsulfate)

4 Disease Overview

Grass Carp Hemorrhage (Hemorrhagicdiseaseofgrasscarp) (see Appendix A) mainly infects grass carp and black carp in those days, death Rate of up to 80%. Other freshwater carps, such as silver carp, bighead carp, carp after infection no clinical symptoms, but can carry the virus to at the spread. The disease is higher than the water temperature above 20 °C popular, 25 °C ~ 28 °C popular peak. Common clinical symptoms of eye protrusion, body Color black, mouth, gills and fins of the base cover bleeding. Ripped skin, muscles visible punctate or massive bleeding. Necropsy abdominal cavity, the intestine visible charge Blood, liver, spleen and blood loss due to congestion or white.

Pathogen grass carp reovirus (Grasscarpreovirus, GCRV), aquatic reovirus genus (Aquareovirus) into member. Virus spherical particles 70nm in diameter, and has a double coat, no capsule, containing fragments of double-stranded RNA 11. According fragment length Divided into large size (L1, L2, L3, size 4.0kb ~ 3.7kb), the (M4, M5, M6, size 2.5kb ~ 2.0kb), small (S7, S8, S9, S10, S11, the size of the 1.6kb ~ 1.0kb) three groups. Antigen, DNA electrophoresis pattern of cells sensitive to the presence in different regions

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