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INDUSTRY STANDARD OF THE PEOPLE'S REPUBLIC OF CHINA

SN/T 3174-2012

Detection and identification of Leptosphaeria lindquistii Frezzi

向日葵黑茎病菌检疫鉴定方法

(English Translation)

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Foreword

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

Please note that some of the content of this document may involve patents. Distribution of this document Institutions do not assume the responsibility to identify these patents.

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

This standard was drafted. Tianjin People's Republic of China Entry-Exit Inspection and Quarantine Bureau, Chinese Academy of Inspection and Quarantine.

1 Scope

This standard specifies the sunflower black stem germs (LeptosphaerialindquistiiFrezzi) (see Appendix A) Isolation, pathogenicity Determination of the experimental step sequence alignment and identification methods.

This standard applies to sunflower seeds Sunflower black stem quarantine and identification of bacteria and germs sunflower in black stem Sunflower Field Testing.

2 Sunflower black stem germs basic information

Name. LeptosphaerialindquistiiFrezzi Asexual generation scientific name. PhomamacdonaldiiBoerma.

English name. SunflowerPhomablackstem Taxonomic position. Sunflower black stem bacteria fungi industry (Fungi), Ascomycota (Ascomycota), chamber Proteobacteria (Leculoascomycetes), Alternaria fungus cavity mesh (Pleosporales), Alternaria fungus chamber Branch (Pleosporaceae), the ball chamber genus (Lepto-sphaeria); anamorph

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belongs mitosis fungal spores (Sphaeropsidales), Coelomycetes (Coelomycetes), Sphaeropsidales (Spha-eropsidales), Phoma (Phoma).

3 PRINCIPLE OF THE METHOD

Pathogen isolation and culture traits, bacteria morphology, host of the pathogenicity of symptoms (see Appendix B) and bacteria ITS1 /ITS4 and ActF1/R1 sequence alignment as a result of quarantine and identification based on sunflower black stem bacteria.

4.1 Equipment

Sterile Petri dish (diameter 90mm ~ 100mm), flask, forceps, surgical scissors, scalpels, inoculation needles, disposable syringes (1mL), cotton yarn, cotton, alcohol lamp, plastic pestle, adjustable micro pipette.

Stereo microscope, biological microscope, clean benches, incubators (24 °C ± 1 °C), electronic balance (inductance 0.01g), autoclaving Pot, conventional refrigerator (-20 °C), PCR amplification device, electrophoresis, gel imager, high-speed refrigerated centrifuge, heated water bath.

4.2 Reagents and culture medium

Ethylene diamine tetra-acetic acid (EDTA), sodium dodecyl sulfate (SDS), Tris-HCl, iso-amyl alcohol, chloroform, ethanol, potassium chloride, Magnesium, proteinase K, primers ITS1/ITS4 and ActF1/R1, Taq polymerase and other molecular biology reagents. Or plant tissue and fungal groups Extraction Kit genomic DNA. Potato, dextrose, agar, penicillin, streptomycin, lactic acid, 1.0% sodium hypochlorite (NaClO).

Acidic medium potato APDA (pH4.5 ~ 5.0), APDA selective medium (see Appendix C).

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