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INDUSTRY STANDARD OF THE PEOPLE'S REPUBLIC OF CHINA

SN/T 3398-2012

Detection for quarantine viruses on soybean by multiple-real time RT-PCR

大豆检疫性病毒多重实时荧光RT-PCR检测方法

(English Translation)

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Foreword

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

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

This standard was drafted. People's Republic of China, Shenzhen CIQ, Shenzhen Inspection and Quarantine Science Research Institute.

1 Scope

This standard specifies the tobacco ringspot virus, Arabis mosaic virus, tomato ringspot virus, southern bean mosaic virus and bean pod mottle disease Toxic fluorescent multiplex real-time RT-PCR detection method.

This standard applies to soybean and its processed products detected in these five viruses.

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.

SN/T 2122 entry and exit quarantine of plants and plant products sampled

3 Terms and Definitions

The following terms and definitions apply to this document.

Reverse transcription - polymerase chain reaction reversetranscriptionpolymerasechainreaction; RT-PGR Reverse transcription - polymerase chain reaction, referred to as RT-PCR, under appropriate reaction conditions to the proposed pumping RNA sample as a template, reverse Transcriptase under suitable

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reaction conditions, RNA was reverse transcribed into cDNA, and then to cDNA as a template for PCR amplification.

Fluorescent multiplex real-time RT-PCR multiple-realtimeRT-PCR with the increase of the fluorescent signal based on RT-PCR products and the number of simultaneous amplification principle, the RT-PCR amplification process, even at the same time Continued monitoring of the reaction system in a variety of different viruses labeled amplified fragment changes the fluorescence signal, according to the average value of the fluorescence signal and the mean baseline Standard deviation, mean fluorescence value obtained i.e. greater than the threshold value, the fluorescence signal was collected to enhance the number of PCR cycles when the predetermined threshold value, i.e., the Ct value.

Ct value determination based on a variety of virus samples with the fluorescent signal whether multiple quarantine viruses.

Principle 4

Using multiplex real-time RT-PCR method to detect fluorescence of tobacco ringspot virus, Arabis mosaic virus, tomato ringspot virus, Southern bean flowers Mosaic virus and bean pod mottle virus (see Appendix A).

5 Equipment and appliances

Ovens, autoclaves, water bath, low temperature refrigerators, water meter, high-speed refrigerated centrifuge, real-time PCR instrument, micropipette (0.1μL ~ 2.5μL, 2μL ~ 20μL, 20μL ~ 200μL, 100μL ~ 1000μL), Vortex, clean benches, electrophoresis Instrument, horizontal electrophoresis tank gel imager, the various tip (1000μL, 200μL, 10μL), centrifuge tubes (1.5mL, 0.6mL, 0.2mL).

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