

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

SN/T 1146.2-2017

Replacing SN/T 1146.2-2009

Molecular detection of Tobacco ringspot virus

烟草环斑病毒分子生物学检测方法

(English Translation)

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Foreword

SN/T 1146 is divided into 3 parts.

- Part 1. Quarantine and identification methods for quarantine tobacco ring spot virus;
- Part 2. Molecular biological detection methods for tobacco ring spot virus;
- Part 3. Quarantine and identification methods for tobacco ring spot virus.

This part is the second part of SN/T 1146.

This part is drafted in accordance with the rules given in GB/T 1.1-2009.

This part replaces SN/T 1146.2-2009 "Tobacco ring spot virus molecular biology detection method".

The main technical differences between this part and SN/T 1146.2-2009 are as follows.

- Appendix C nucleic acid extraction increased magnetic beads adsorption method and commercial kit method;
- PCR primers have been adjusted;
- Increased RT-LAMP detection method;
- Increased preparation method of buffer;
- Deleted the basic information of Appendix A tobacco ring spot virus.

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

This section drafted by. Shanghai Entry-Exit Inspection and Quarantine Bureau of the

People's Republic of China, Beijing Entry-Exit Inspection and Quarantine Bureau, China Academy of Inspection and Quarantine.

1 Scope

This part of SN/T 1146 specifies the basic principles and methods for molecular biology testing of tobacco ring spot virus.

This section applies to the detection of tobacco ring spot virus carried in asexual propagation materials, tissue culture seedlings, bulbs and seedlings.

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 SN/T 1146 Tobacco ring spot virus quarantine and identification method

3 Tobacco ring spot virus basic information

Chinese name. Tobacco ring spot virus English name. Tobaccoringpotvirus Abbreviation. TRSV Taxonomic status. Comoviridae, nepovirus.

Route of transmission. The virus can be transmitted by seed, grafting, mechanical inoculation and mediators. *S. cerevisiae* (*Xiphinema americanum*); soybean, melon, lettuce, cowpea, amaranth, potato and geranium can be planted, soybean seed rate Up to 40% to 100%.

Genome. The viral genome contains two ssRNAs. RNA-1 is 7.514 kb in length and RNA-2 is 3.929 kb in length, coat protein gene 1.548kb.

4 Principle of the method

The serological and molecular biological characteristics of tobacco ring spot virus are the main basis for quarantine identification of the virus. Using RT-PCR, IC-RT-the detection of the virus was carried out by molecular biological methods such as PCR, real-time fluorescent RT-PCR and RT-LAMP.

5.1 Instrumentation

PCR instrument, real-time PCR instrument, ultra-clean bench, electronic balance (1/10000g), electrophoresis, tank, gel imaging system System, water bath, high speed

refrigerated centrifuge, refrigerator, -80 ° C ultra-low temperature refrigerator, autoclave, ice machine, microwave oven, vortex shaker, pH Count.

5.2 Appliances

Adjustable micropipettes (2 μ L, 10 μ L, 100 μ L, 200 μ L, 1000 μ L, 5000 μ L) and corresponding RNase-free tips, no