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

GB/T 44342-2024

**Detection and identification of *Didymella glomerata* (Corda)
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苏铁叶枯病菌检疫鉴定方法

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Foreword

This document is in accordance with the provisions of GB/T 1.1-2020 "Guidelines for standardization work Part

1. Structure and drafting rules for standardization documents" Drafting is required. Please note that some of the contents of this document may involve patents. The issuing organization of this document does not assume the responsibility for identifying patents. This document was proposed and coordinated by the National Technical Committee on Plant Quarantine Standardization (SAC/TC 271). This document was drafted by: China Institute of Inspection and Quarantine, Institute of Microbiology, Chinese Academy of Sciences, Shenzhen Fairy Lake Botanical Garden, Ningbo Hai Guan Technology Center, Fujian Tropical Crops Science Research Institute, and Tibet Autonomous Region Plateau Biology Research Institute. The main drafters of this document are. Liu Fang, Huang Ying, Cheng Yinghui, Cai Lei, Xu Han, Duan Weijun, Cai Jiangqiao, Luo Jinshui, Wang Cong, Yan Jin, Phub Dorje. Quarantine and identification methods for cycad leaf blight pathogen

1 Scope

GB/T 44342-2024 is the Chinese phytosanitary method for detecting and identifying *Didymella glomerata*, the fungus that causes cycad leaf blight. Standards of this kind exist because a quarantine decision has to be defensible: a consignment is held or destroyed on the basis of an identification, and the identification has to be reproducible by another laboratory from the same material. The standard gives the basic information about the pathogen, its hosts, distribution, symptoms and biology, then the principle of the method, the reagents, materials and culture media required, the sampling and the preparation of the specimens, the examination of symptoms, the isolation and culture of the fungus, the morphological identification of the colony, the pycnidia and the conidia with their measurements, the molecular identification by PCR and sequencing with the primers and conditions specified, the criteria by which a positive identification is concluded, and the

retention of specimens and records. It took effect on 23 August 2024, and for a plant quarantine laboratory or an exporter of cycads and ornamentals to China it is the governing method.

This document describes the isolation, culture, morphological identification, and molecular biological characterization of *Didymella glomerata*. method. This document applies to the screening and identification of cycad leaf blight pathogens in plants and their products.

2 Normative references

This document has no normative references.

3 Terms and definitions

There are no terms or definitions that require definition in this document.

4 Basic information of Cycad leaf blight pathogen

Scientific name. *Didymella glomerata* (Corda) Qian Chen Synonyms. *Coniothyrium glomeratum* Corda, Icon. Fungorum (Prague) 4.39.1840. *Aposphaeria glomerata* (Corda) Sacc., Syll.Fung. (Abellini) 3.175.1884. *Phoma glomerata* (Corda) Wollenw. Chinese name. Subspora tuanji Chinese synonym. Grape stem blight pathogen Classification. Fungi, Ascomycota, Dothideomycetes, Pleosporales, Didymellaceae, *Didymella*. The morphological characteristics of the pathogen of Cycad leaf blight are similar to those of other species in the genus *Subspora*, and it is difficult to distinguish them based on morphological characteristics alone. Including *D.anserina*, *D.aurea*, *D.musae*, etc. Additional information on the cycad leaf blight pathogen is provided in Appendix A.

5 Principles of the method

Based on the disease symptoms, morphological characteristics of isolated bacteria (see Appendix A), and real-time fluorescent polymerase chain reaction (PCR) detection method Or beta-tubulin tub2 sequence characteristics are used to detect and identify cycad leaf blight pathogens.

6 Reagents, Materials, and Media

6.1 Reagents and Materials Unless otherwise specified, all experimental reagents were of analytical grade or biochemical reagents. Reagents and materials required for isolation and culture. potatoes, oatmeal, distilled water, glucose, agar powder, sodium hypochlorite, 75% ethanol, and sterile water.