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Plant biostimulants - Detection of *Listeria monocytogenes*

Biostimulants des végétaux - Détection de *Listeria* monocytogenes

Pflanzen-Biostimulanzien - Nachweis von *Listeria* monocytogenes

This Technical Specification (CEN/TS) was approved by CEN on 3 January 2022 for provisional application.

The period of validity of this CEN/TS is limited initially to three years. After two years the members of CEN will be requested to submit their comments, particularly on the question whether the CEN/TS can be converted into a European Standard.

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EUROPEAN COMMITTEE FOR STANDARDIZATION COMITÉ EUROPÉEN DE NORMALISATION EUROPÄISCHES KOMITEE FÜR NORMUNG

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Con	Contents	
Europ	pean foreword	3
Intro	duction	4
1	Scope	5
2	Normative references	
3	Terms and definitions	_
4	Principle	
 4.1	General	
4.2	Pre-enrichment in non-selective liquid medium	
4.3	Enrichment in/on selective media	
4.4	Plating out on selective solid media	
4.5	Confirmation	
5	Culture media, reagents, antisera	6
6	Equipment and consumables	6
7	Sampling	7
8	Preparation of test sample	
9	Preparation procedure	7
9.1	Test portion and initial suspension	7
9.2	Non-selective pre-enrichment	
9.3	Selective enrichment	
9.4	Plating out	
9.5	Confirmation of L. monocytogenes	
9.6	Interpretation of morphological and physiological properties and of the bi	iochemical
9.7	reactionsAdditional characterization of isolated strains (optional)	
10	Expression of results	
11	Performance characteristics of the method	13
11.1	Interlaboratory studies	13
11.2	Sensitivity	13
11.3	Specificity	13
11.4	LOD ₅₀	13
12	Test report	13
13	Quality assurance	13
	x A (normative) Diagram of the procedures	
	x B (normative) Composition and preparation of culture media and reagents	
	ography	
~10110	~ ~ ~ ~ ~ ~ ~ · · · · · · · · · · · · ·	

European foreword

This document (CEN/TS 17710:2022) has been prepared by Technical Committee CEN/TC 455 "Plant Biostimulants", the secretariat of which is held by AFNOR.

Attention is drawn to the possibility that some of the elements of this document may be the subject of patent rights. CEN shall not be held responsible for identifying any or all such patent rights.

This document has been prepared under a Standardization Request given to CEN by the European Commission and the European Free Trade Association.

Any feedback and questions on this document should be directed to the users' national standards body. A complete listing of these bodies can be found on the CEN website.

According to the CEN/CENELEC Internal Regulations, the national standards organisations of the following countries are bound to announce this Technical Specification: Austria, Belgium, Bulgaria, Croatia, Cyprus, Czech Republic, Denmark, Estonia, Finland, France, Germany, Greece, Hungary, Iceland, Ireland, Italy, Latvia, Lithuania, Luxembourg, Malta, Netherlands, Norway, Poland, Portugal, Republic of n vaki. North Macedonia, Romania, Serbia, Slovakia, Slovenia, Spain, Sweden, Switzerland, Turkey and the United Kingdom.

Introduction

This document was prepared by the experts of CEN/TC 455 "Plant Biostimulants". The European Committee for Standardization (CEN) was requested by the European Commission (EC) to draft European standards or European standardization deliverables to support the implementation of Regulation (EU) 2019/1009 of 5 June 2019 laying down rules on the making available on the market of EU fertilizing products ("FPR" or "Fertilising Products Regulation").

This standardization request, presented as M/564, also contributes to the Communication on "Innovating for Sustainable Growth: A Bio economy for Europe". The Working Group 5 "Labelling and denominations", was created to develop a work program as part of this request. The technical committee CEN/TC 455 "Plant Biostimulants" was established to carry out the work program that will prepare a series of standards. The interest in biostimulants has increased significantly in Europe as a valuable tool to use in agriculture. Standardization was identified as having an important role in order to promote the use of biostimulants. The work of CEN/TC 455 seeks to improve the reliability of the supply chain, thereby improving the confidence of farmers, industry, and consumers in biostimulants, and will promote and support commercialisation of the European biostimulant industry.

Biostimulants used in agriculture can be applied in multiple ways: on soil, on plant, as seed treatment, etc. A microbial plant biostimulant consists of a microorganism or a consortium of microorganisms, as referred to in Component Material Category 7 of Annex II of the EU Fertilising Products Regulation.

This document is applicable to all microbial biostimulants in agriculture.

The Table 1 below summarizes many of the agro-ecological principles and the role played by biostimulants.

Table 1 — Agro-ecological principles and the role played by biostimulants

Increase biodiversity
By improving soil microorganism quality/quantity
Reinforce biological regulation and interactions
By reinforcing plant-microorganism interactions
- symbiotic exchanges i.e. Mycorrhizae
- symbiotic exchanges i.e. Rhizobiaceae/Faba
- secretions mimicking plant hormones (i.e. Trichoderma)
By regulating plant physiological processes
- for e.g. growth, metabolism, plant development
Improve biogeochemical cycles
- improve absorption of nutritional elements
- improve bioavailability of nutritional elements in the soil
- stimulate degradation of organic matter

WARNING — Persons using this document should be familiar with normal laboratory practice. This document does not purport to address all of the safety problems, if any, associated with its use. It is the responsibility of the user to establish appropriate safety and health practices and to ensure compliance with any national regulatory conditions.

IMPORTANT — It is absolutely essential that tests conducted in accordance with this document be carried out by suitably trained staff.

1 Scope

This document provides a method for the detection of *Listeria monocytogenes* in microbial plant biostimulants for verifying that the concentration of this human pathogen does not exceed the respective limits outlined in the EU Regulation on Fertilising Products [1].

2 Normative references

The following documents are referred to in the text in such a way that some or all of their content constitutes requirements of this document. For dated references, only the edition cited applies. For undated references, the latest edition of the referenced document (including any amendments) applies.

CEN/TS 17724, Plant biostimulants — Terminology

CEN/TS 17708, Plant biostimulants — Preparation of sample for microbial analysis

EN ISO 11290-1:2017, Microbiology of the food chain — Horizontal method for the detection and enumeration of Listeria monocytogenes and of Listeria spp. — Part 1: Detection method (ISO 11290-1:2017)

EN ISO 11133,¹ Microbiology of food, animal feed and water — Preparation, production, storage and performance testing of culture media

EN ISO 7218, Microbiology of food and animal feeding stuffs — General requirements and guidance for microbiological examinations (ISO 7218)

3 Terms and definitions

For the purposes of this document, the terms and definitions are given in CEN/TS 17724 and the following apply.

3.1

Listeria monocytogenes

microorganisms which form typical colonies on solid selective media described and which display the morphological, physiological and biochemical characteristics described when the analysis is carried out in accordance with this document

[SOURCE: EN ISO 11290-1:2017, 3.1]

4 Principle

4.1 General

The detection of *Listeria monocytogenes* requires four successive stages as specified in Annex A.

NOTE *L. monocytogenes* can be present in small numbers and is often accompanied by considerably larger numbers of bacteria belonging to different taxonomic groups or different *Listeria* species. Pre-enrichment is used to permit the detection of low numbers of *L. monocytogenes* or injured *L. monocytogenes*.

4.2 Pre-enrichment in non-selective liquid medium

Half-Fraser broth (225 ml) at ambient temperature is inoculated with the test portion sample (25 g or 25 ml), then incubated at 30 °C \pm 1 °C for 24 h to 26 h.

¹ As impacted by EN ISO 11133:2014/A1:2018 and EN ISO 11133:2014/A2:2020.