

Irish Standard I.S. EN 17252:2020

Foodstuffs - Determination of phomopsin A in lupin seeds and lupin derived products by HPLC-MS/MS

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I.S. EN 17252:2020

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National Foreword

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Foodstuffs - Determination of phomopsin A in lupin seeds and lupin derived products by HPLC-MS/MS

Produits alimentaires - Détermination de la teneur en phomopsine A dans les graines de lupin et les produits dérivés du lupin par CL-SM/SM Lebensmittel - Bestimmung von Phomopsin A in Lupinensamen und Lupinenerzeugnissen mit LC-MS/MS

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EN 17252:2020 (E)

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EN 17252:2020 (E)

European foreword

This document (EN 17252:2020) has been prepared by Technical Committee CEN/TC 275 "Food analysis - Horizontal methods", the secretariat of which is held by DIN.

This European Standard shall be given the status of a national standard, either by publication of an identical text or by endorsement, at the latest by July 2020, and conflicting national standards shall be withdrawn at the latest by July 2020.

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EN 17252:2020 (E)

Introduction

Phomopsins are mycotoxins produced by the fungus *Diaporthe toxica*. There are several phomopsins of which phomopsin A is the major toxic congener. The main host of the fungus are lupins (*Lupinus L*.). Lupin seeds are being used as food ingredients and therefore phomopsin A might occur in food ingredients and food products containing lupin seeds or lupin flour.

WARNING 1 — Suitable precaution and protection measures need to be taken when carrying out working steps with harmful chemicals. The latest version of the hazardous substances ordinance (EU) 1907/2006 [3] should be taken into account as well as appropriate national statements.

WARNING 2 — The use of this document can involve hazardous materials, operations and equipment. This document does not purport to address all the safety problems associated with its use. It is the responsibility of the user of this document to establish appropriate safety and health practices and determine the applicability of regulatory limitations prior to use.

1 Scope

This document specifies a procedure for the determination of phomopsin A in lupin seeds and lupin-derived products based on liquid chromatography with tandem mass spectrometry (LC-MS/MS). Several phomopsins exist, i.e. phomopsin A, B, C and D, but the method only deals with the quantitative measurement of phomopsin A due to lack of commercially available analytical reference standards for the other phomopsins.

The method has been validated for phomopsin A in naturally contaminated lupin seeds, lupin flour and crisp bread at levels ranging from approximately 5 μ g/kg to 60 μ g/kg.

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.

EN ISO 3696, Water for analytical laboratory use — Specification and test methods (ISO 3696)

3 Terms and definitions

No terms and definitions are listed in this document.

ISO and IEC maintain terminological databases for use in standardization at the following addresses:

- IEC Electropedia: available at http://www.electropedia.org/
- ISO Online browsing platform: available at https://www.iso.org/obp

4 Principle

Phomopsin A is extracted from the homogenized sample material by shaking with a mixture of acetonitrile/water/acetic acid (80+19+1, v+v+v). After centrifugation, an aliquot of the extract is diluted with water, optionally filtered, and analysed by liquid chromatography coupled to tandem mass spectrometry (LC-MS/MS). Phomopsin A is quantified by multi-level matrix-matched calibration.

5 Reagents

Use only reagents of recognized analytical grade and water complying with grade 1 of EN ISO 3696, unless otherwise specified. Solutions shall be of quality for LC analysis, unless otherwise specified.

- **5.1 Water,** deionised.
- **5.2 Water,** LC-MS grade.
- **5.3** Acetonitrile, pro analysis (p.a.).
- **5.4 Methanol,** LC-MS grade.
- **5.5** Acetic acid, purity greater than mass fraction $w \ge 98 \%$.
- **5.6** Ammonium formate, p.a.



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