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Rapeseed and rapeseed meals - Determination of glucosinolates content - Method using high-performance liquid chromatography (ISO 9167:2019)

Táto norma obsahuje anglickú verziu európskej normy.  
This standard includes the English version of the European Standard.

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English Version

## Rapeseed and rapeseed meals - Determination of glucosinolates content - Method using high-performance liquid chromatography (ISO 9167:2019)

Graines et tourteaux de colza - Dosage des  
glucosinolates - Méthode par chromatographie liquide  
à haute performance (ISO 9167:2019)

Rapssamen und Rapsschrot - Bestimmung des  
Glucosinolatgehaltes - Verfahren mittels  
Hochleistungsflüssigchromatographie (ISO 9167:2019)

This European Standard was approved by CEN on 4 May 2019.

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**EN ISO 9167:2019 (E)**

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## **European foreword**

This document (EN ISO 9167:2019) has been prepared by Technical Committee ISO/TC 34 "Food products" in collaboration with Technical Committee CEN/TC 307 "Oilseeds, vegetable and animal fats and oils and their by-products - Methods of sampling and analysis" the secretariat of which is held by AFNOR.

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of an identical text or by endorsement, at the latest by December 2019, and conflicting national standards shall be withdrawn at the latest by December 2019.

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.

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According to the CEN-CENELEC Internal Regulations, the national standards organizations of the following countries are bound to implement this European Standard: 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 North Macedonia, Romania, Serbia, Slovakia, Slovenia, Spain, Sweden, Switzerland, Turkey and the United Kingdom.

## **Endorsement notice**

The text of ISO 9167:2019 has been approved by CEN as EN ISO 9167:2019 without any modification.

**INTERNATIONAL  
STANDARD**

**ISO  
9167**

First edition  
2019-05

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**Rapeseed and rapeseed meals —  
Determination of glucosinolates  
content — Method using high-  
performance liquid chromatography**

*Graines et tourteaux de colza — Dosage des glucosinolates —  
Méthode par chromatographie liquide à haute performance*



Reference number  
ISO 9167:2019(E)

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# ISO 9167:2019(E)

## Foreword

ISO (the International Organization for Standardization) is a worldwide federation of national standards bodies (ISO member bodies). The work of preparing International Standards is normally carried out through ISO technical committees. Each member body interested in a subject for which a technical committee has been established has the right to be represented on that committee. International organizations, governmental and non-governmental, in liaison with ISO, also take part in the work. ISO collaborates closely with the International Electrotechnical Commission (IEC) on all matters of electrotechnical standardization.

The procedures used to develop this document and those intended for its further maintenance are described in the ISO/IEC Directives, Part 1. In particular, the different approval criteria needed for the different types of ISO documents should be noted. This document was drafted in accordance with the editorial rules of the ISO/IEC Directives, Part 2 (see [www.iso.org/directives](http://www.iso.org/directives)).

Attention is drawn to the possibility that some of the elements of this document may be the subject of patent rights. ISO shall not be held responsible for identifying any or all such patent rights. Details of any patent rights identified during the development of the document will be in the Introduction and/or on the ISO list of patent declarations received (see [www.iso.org/patents](http://www.iso.org/patents)).

Any trade name used in this document is information given for the convenience of users and does not constitute an endorsement.

For an explanation of the voluntary nature of standards, the meaning of ISO specific terms and expressions related to conformity assessment, as well as information about ISO's adherence to the World Trade Organization (WTO) principles in the Technical Barriers to Trade (TBT) see [www.iso.org/iso/foreword.html](http://www.iso.org/iso/foreword.html).

This document was prepared by Technical Committee ISO/TC 34, *Food products*, Subcommittee SC 2, *Oleaginous seeds and fruits and oilseed meals*.

This first edition cancels and replaces ISO 9167-1:1992, which has been technically revised. It also incorporates the amendment ISO 9167-1:1992/Amd.1:2013. The main changes are as follows:

- rapeseed meals have been added to the scope with the addition of a new collaborative trial;
- in [9.2](#), methanol 70 % has been replaced by ethanol 50 % for lower toxicity<sup>[6]</sup>;
- in [9.2](#), only one extraction is carried out instead of two;
- in [10.2](#) and [E.5.1](#), the term “relative proportionality factor” has been used instead of “response factor”;
- the isocratic mode has been added in [Annex E](#).

Any feedback or questions on this document should be directed to the user's national standards body. A complete listing of these bodies can be found at [www.iso.org/members.html](http://www.iso.org/members.html).



## Introduction

The glucosinolates in rapeseed can be analysed by chromatographic, enzymatic or spectroscopic methods. This document describes a chromatographic method with two conditions (gradient and isocratic) of elution for qualitative and quantitative analysis of individual glucosinolates in rapeseed and rapeseed meals. The method with gradient elution is considered as the reference method whereas the method with isocratic elution is considered as a simplified method and is presented in [Annex E](#) as information.

This document specifies a method using high-performance liquid chromatography (HPLC) with gradient elution as reference method. For the isocratic mode, the choice of the internal standard, the chromatographic conditions and the separation results are different from the reference method. These aspects are discussed in [Annex E](#).

# Rapeseed and rapeseed meals — Determination of glucosinolates content — Method using high-performance liquid chromatography

## 1 Scope

This document specifies a method for the determination of the individual glucosinolates content in rapeseeds and rapeseed meals using high-performance liquid chromatography with gradient elution.

This method was tested on rapeseeds and rapeseed meals (*Brassica rapa*, *Brassica napus* and *Brassica juncea*) but is applicable to other plant materials, on the condition that the occurring glucosinolates previously identified are described in this document. On the contrary, the quantitative analysis of the concerned glucosinolate(s) is not carried out.

**NOTE** This method does not determine glucosinolates that are substituted on the glucose molecule, but these compounds are of little importance in commercial rapeseed and rapeseed meal.

[Annex A](#) presents the results of the interlaboratory trials for the gradient elution HPLC method. [Annex B](#) presents how to check the titre of the prepared internal standard solution. [Annex C](#) presents how to prepare and test the purified sulfatase solution and how to check the desulphation step on the ion exchange column. [Annex D](#) presents the HPLC and column performance criteria qualification.

The analysis of glucosinolates content in rapeseed can also be done using an isocratic elution mode. This requires some modifications of the method (internal, standard, HPLC column and HPLC buffers), as described in [Annex E](#).

## 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.

ISO 664, *Oilseeds — Reduction of laboratory sample to test sample*

ISO 665, *Oilseeds — Determination of moisture and volatile matter content*

ISO 771, *Oilseed residues — Determination of moisture and volatile matter content*

ISO 3696, *Water for analytical laboratory use — Specification and test methods*

ISO 5502, *Oilseed residues — Preparation of test samples*

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