

Foodstuffs - Determination of T-2 toxin and HT-2 toxin in cereals and cereal products for infants and young children by LC-MS/MS after SPE cleanup

EESTI STANDARDI EESSÕNA

NATIONAL FOREWORD

See Eesti standard EVS-EN 16923:2017 sisaldab Euroopa standardi EN 16923:2017 ingliskeelset teksti.	This Estonian standard EVS-EN 16923:2017 consists of the English text of the European standard EN 16923:2017.
Standard on jõustunud sellekohase teate avaldamisega EVS Teatajas	This standard has been endorsed with a notification published in the official bulletin of the Estonian Centre for Standardisation.
Euroopa standardimisorganisatsioonid on teinud Euroopa standardi rahvuslikele liikmetele kättesaadavaks 24.05.2017.	Date of Availability of the European standard is 24.05.2017.
Standard on kättesaadav Eesti Standardikeskusest.	The standard is available from the Estonian Centre for Standardisation.

Tagasisidet standardi sisu kohta on võimalik edastada, kasutades EVS-i veebilehel asuvat tagasiside vormi või saates e-kirja meiliaadressile standardiosakond@evs.ee.

ICS 67.060, 67.230

Standardite reprodutseerimise ja levitamise õigus kuulub Eesti Standardikeskusele

Andmete paljundamine, taastekitamine, kopeerimine, salvestamine elektroonsesse süsteemi või edastamine ükskõik millises vormis või millisel teel ilma Eesti Standardikeskuse kirjaliku loata on keelatud.

Kui Teil on küsimusi standardite autorikaitse kohta, võtke palun ühendust Eesti Standardikeskusega:
Koduleht www.evs.ee; telefon 605 5050; e-post info@evs.ee

The right to reproduce and distribute standards belongs to the Estonian Centre for Standardisation

No part of this publication may be reproduced or utilized in any form or by any means, electronic or mechanical, including photocopying, without a written permission from the Estonian Centre for Standardisation.

If you have any questions about copyright, please contact Estonian Centre for Standardisation:

Homepage www.evs.ee; phone +372 605 5050; e-mail info@evs.ee

English Version

Foodstuffs - Determination of T-2 toxin and HT-2 toxin in
cereals and cereal products for infants and young children
by LC-MS/MS after SPE cleanup

Produits alimentaires - Dosage des toxines T-2 et HT-2
dans les céréales et les produits céréaliers pour
nourrissons et enfants en bas âge par CL-SM/SM après
purification par SPE

Lebensmittel - Bestimmung von T-2-Toxin und HT-2-
Toxin in Getreide und Säuglings- und
Kleinkindernahrung auf Getreidebasis mit LC-MS/MS
nach SPE-Reinigung

This European Standard was approved by CEN on 27 February 2017.

CEN members are bound to comply with the CEN/CENELEC Internal Regulations which stipulate the conditions for giving this European Standard the status of a national standard without any alteration. Up-to-date lists and bibliographical references concerning such national standards may be obtained on application to the CEN-CENELEC Management Centre or to any CEN member.

This European Standard exists in three official versions (English, French, German). A version in any other language made by translation under the responsibility of a CEN member into its own language and notified to the CEN-CENELEC Management Centre has the same status as the official versions.

CEN members are the national standards bodies of Austria, Belgium, Bulgaria, Croatia, Cyprus, Czech Republic, Denmark, Estonia, Finland, Former Yugoslav Republic of Macedonia, France, Germany, Greece, Hungary, Iceland, Ireland, Italy, Latvia, Lithuania, Luxembourg, Malta, Netherlands, Norway, Poland, Portugal, Romania, Serbia, Slovakia, Slovenia, Spain, Sweden, Switzerland, Turkey and United Kingdom.



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

CEN-CENELEC Management Centre: Avenue Marnix 17, B-1000 Brussels

Contents	Page
European foreword	3
Introduction	4
1 Scope	5
2 Normative references	5
3 Principle	5
4 Reagents	5
5 Apparatus and equipment	7
6 Procedure	9
7 Calculation	10
8 Precision	11
9 Test report	12
Annex A (informative) Example chromatograms (API 4000™)	13
Annex B (informative) Example conditions for suitable LC-MS/MS systems	17
Annex C (informative) Precision data	24
Bibliography	26

European foreword

This document (EN 16923:2017) 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 November 2017, and conflicting national standards shall be withdrawn at the latest by November 2017.

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 mandate given to CEN by the European Commission and the European Free Trade Association.

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, Former Yugoslav Republic of Macedonia, France, Germany, Greece, Hungary, Iceland, Ireland, Italy, Latvia, Lithuania, Luxembourg, Malta, Netherlands, Norway, Poland, Portugal, Romania, Serbia, Slovakia, Slovenia, Spain, Sweden, Switzerland, Turkey and the United Kingdom.

Introduction

The mycotoxin T-2 toxin and its metabolite HT-2 toxin belong to the group of trichothecenes which are produced by various *Fusarium* species. Cereals like maize, wheat, barley, oats, and rye are most likely to be affected.

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, Regulation (EC) No 1907/2006 [3], should be taken into account as well as appropriate National statements e.g. such as in [4].

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.

WARNING 3 — T-2 toxin and its metabolite HT-2 toxin are known to have carcinogenic effects.

1 Scope

This European Standard describes a method for the determination of T-2 toxin and HT-2 toxin in cereals and cereal based products e.g. oats, intended for nutrition of infants and young children by high performance liquid chromatography (HPLC) coupled with tandem mass spectrometry (MS/MS) after cleanup by solid phase extraction (SPE) [5].

The method has been validated for HT-2 toxin in oat flour at levels of 9,3 µg/kg and 28,1 µg/kg, oat flakes at levels of 16,5 µg/kg and 21,4 µg/kg, and breakfast cereals (containing oat flakes) at a level of 8,1 µg/kg and for T-2 toxin in oat flour at levels of 4,4 µg/kg and 8,3 µg/kg, oat flakes at levels of 4,9 µg/kg and 6,6 µg/kg and breakfast cereals (containing oat flakes) at a level of 3,5 µg/kg.

Laboratory experiences [6] have shown that the method is also applicable to highly swelling materials (dry cereal based porridges and modified starches), but these were not examined in the method validation study. Details are outlined in 6.3.

The method can also be applied to oat-by-products at higher levels of T-2- and HT-2 toxin. In this case, the dilution steps need to be considered [6].

The method can also be applied to cereals and cereal products for infants and young children based on e.g. wheat, barley, and rice. In this case, the method needs to be in-house-validated for each material. At the time of the interlaboratory study, planned range was 10 µg/kg to 100 µg/kg, and it is known from the pre-study that the method works well in the whole range, although final validation was only done in the range from 3,5 µg/kg to 28,1 µg/kg.

2 Normative references

The following documents, in whole or in part, are normatively referenced in this document and are indispensable for its application. 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 Principle

T-2 toxin and HT-2 toxin are extracted with acetonitrile-water mixture and by shaking manually or with a laboratory blender. A solid phase extraction column or a pass through column is used to clean up and concentrate the filtered and diluted extract, see also [7]. The toxins are determined by HPLC coupled with tandem mass spectrometry.

4 Reagents

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

4.1 Acetonitrile, HPLC grade.

4.2 Methanol, HPLC grade.

4.3 Solvent mixture.

Mix 20 parts of acetonitrile (4.1) and 80 parts of water (20+80, v+v).

4.4 Extraction mixture.

Mix 84 parts of acetonitrile (4.1) and 16 parts of water (84+16, v+v).