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Microbiology of the food chain - Methods for the detection of Anisakidae L3 larvae in fish and fishery products - Part 2: Artificial digestion method (ISO 23036-2:2021)

EESTI STANDARDI EESSÕNA

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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 and Accreditation.
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Standard on kättesaadav Eesti Standardimis- ja Akrediteerimiskeskusest.	The standard is available from the Estonian Centre for Standardisation and Accreditation.

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ICS 07.100.30

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EUROPEAN STANDARD

NORME EUROPÉENNE

EUROPÄISCHE NORM

EN ISO 23036-2

May 2021

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English Version

Microbiology of the food chain - Methods for the detection of Anisakidae L3 larvae in fish and fishery products - Part 2: Artificial digestion method (ISO 23036-2:2021)

Microbiologie de la chaîne alimentaire - Méthodes de recherche des larves L3 d'Anisakidae dans les poissons et produits de la pêche - Partie 2: Méthode de digestion artificielle (ISO 23036-2:2021)

Mikrobiologie der Lebensmittelkette - Verfahren zum Nachweis von Anisakidae L3-Larven in Fisch und Fischereierzeugnissen - Teil 2: Verfahren der künstlichen Verdauung (ISO 23036-2:2021)

This European Standard was approved by CEN on 24 March 2021.

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Ref. No. EN ISO 23036-2:2021 E

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European foreword

This document (EN ISO 23036-2:2021) has been prepared by Technical Committee ISO/TC 34 "Food products" in collaboration with Technical Committee CEN/TC 463 “Microbiology of the food chain” 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of an identical text or by endorsement, at the latest by November 2021, and conflicting national standards shall be withdrawn at the latest by November 2021.

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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Endorsement notice

The text of ISO 23036-2:2021 has been approved by CEN as EN ISO 23036-2:2021 without any modification.

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Contents

Page

Foreword ... **iv**
Introduction ... **v**
1 Scope ... **1**
2 Normative references ... **1**
3 Terms and definitions ... **1**
4 Principle ... **2**
4.1 General ... 2
4.2 Sample size ... 2
4.3 Sample preparation ... 2
4.4 Digestion of the sample ... 2
4.5 Filtration of the digest fluid ... 2
4.6 Verification of findings ... 2
5 Reagents ... **3**
6 Equipment and consumables ... **3**
7 Sampling ... **4**
8 Procedure ... **4**
8.1 Preparation of the sample ... 4
8.2 Preparation of the digest fluid ... 4
8.3 Digestion of the sample in the glass beaker ... 5
8.4 Filtration of the digest fluid ... 5
8.5 Microscopic examination ... 5
9 Expression of the results ... **5**
10 Performance characteristics of the method ... **6**
11 Test report ... **6**
12 Quality assurance ... **6**
Annex A (informative) **Sample collection** ... **7**
Annex B (informative) **Example of a laboratory worksheet for recording data when testing fish fillets with the artificial digestion method** ... **8**
Bibliography ... **9**

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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).

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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.

This document was prepared by Technical Committee ISO/TC 34, *Food products*, Subcommittee SC 9, *Microbiology*, in collaboration with the European Committee for Standardization (CEN) Technical Committee CEN/TC 275, *Food analysis — Horizontal methods*, in accordance with the Agreement on technical cooperation between ISO and CEN (Vienna Agreement).

A list of all parts in the ISO 23036 series can be found on the ISO website.

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Introduction

Nematodes of the Anisakidae family have a complex life cycle involving a high number of hosts. Adult stages of Anisakidae reside in the stomach of marine mammals, where they are embedded in the mucosa. Unembryonated eggs produced by adult females are released with the faeces of marine mammals and become embryonated in seawater, where first-stage larvae (L1) develop in the eggs. The larvae moult to become free-swimming second-stage larvae (L2) and, if ingested by crustaceans, mature into third-stage larvae (L3). This stage is infective to fish and squid, and larvae are transferred between fishes through predation, maintaining the L3 stage. Some larvae migrate from the abdominal cavity into muscle tissues. Humans are incidental hosts and can be infected after ingesting raw or undercooked infected fish or cephalopods containing viable L3.

Nematodes of the family Anisakidae are the causative agents of human anisakidosis, a disease that is not only a public health hazard affecting humans, but also represents an economic problem in fishery and food safety (the term "anisakiasis", designating the disease caused by members of the genus *Anisakis*, is also sometimes used). Worldwide, marine and wild anadromous fishes are intermediate hosts of Anisakidae, whereas marine mammals are the definitive hosts.

Visual inspection procedures for the detection of Anisakidae larvae in fish are employed to minimize the risk that contaminated fish will reach the consumer,[1]][2] thus preventing human anisakidosis.

The UV-press and the artificial digestion of the fish muscular tissue are the methods specifically designed to detect nematode larvae in fish and to evaluate the infestation level of a batch, and have been validated and tested in multicentre collaborative studies[3] (see Clause 10).

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Microbiology of the food chain — Methods for the detection of Anisakidae L3 larvae in fish and fishery products —

Part 2: Artificial digestion method

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1 Scope

This document specifies a method for the detection of Anisakidae L3 larvae commonly found in marine and anadromous fishes. The method is applicable to fresh fish and/or frozen fish, as well as lightly processed fish products, such as marinated, salted or smoked. It is also suitable for visceral organs as a confirmatory method for a visual inspection scheme.

The artificial digestion method[4][5][6] is applicable to quantifying parasitic infections by estimating the number of parasites in the fish musculature and, when applied to fresh fish or lightly processed fish products (never frozen before processing), determining the viability of Anisakidae L3, which can be present.

This method does not apply to determining the species or genotype of detected parasites. Final identification is made by morphological and/or molecular methods.

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 7218, *Microbiology of food and animal feeding stuffs — General requirements and guidance for microbiological examinations*

3 Terms and definitions

For the purposes of this document, the following terms and definitions apply.

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

— ISO Online browsing platform: available at https://www.iso.org/obp

— IEC Electropedia: available at http://www.electropedia.org/

3.1
Anisakidae L3 larvae
third-stage larvae (L3) belonging to the Anisakidae family, in particular to *Anisakis*, *Contracaecum* and *Pseudoterranova* genera

Note 1 to entry: For practical purposes, the genus *Hysterothylacium* belonging to Raphidascarididae family and already classified as Anisakidae, can be also included.

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