



**SLOVENSKI STANDARD**  
**SIST EN ISO 23611-4:2023**

**01-marec-2023**

**Nadomešča:**

**SIST EN ISO 23611-4:2012**

---

**Kakovost tal - Vzorčenje nevretenčarjev v tleh - 4. del: Vzorčenje, ekstrakcija in identifikacija nematod iz tal (ISO 23611-4:2022)**

Soil quality - Sampling of soil invertebrates - Part 4: Sampling, extraction and identification of soil-inhabiting nematodes (ISO 23611-4:2022)

Bodenbeschaffenheit - Probenahme von Wirbellosen im Boden - Teil 4: Probenahme, Extraktion und Bestimmung von Boden bewohnenden Nematoden (ISO 23611-4:2022)

Qualité du sol - Prélèvement des invertébrés du sol - Partie 4 : Prélèvement, extraction et identification des nématodes du sol (ISO 23611-4:2022)

**Ta slovenski standard je istoveten z: EN ISO 23611-4:2022**

---

**ICS:**

13.080.30      Biološke lastnosti tal      Biological properties of soils

**SIST EN ISO 23611-4:2023**      **en,fr,de**



EUROPEAN STANDARD

EN ISO 23611-4

NORME EUROPÉENNE

EUROPÄISCHE NORM

August 2022

ICS 13.080.30; 13.080.05

Supersedes EN ISO 23611-4:2011

English Version

## Soil quality - Sampling of soil invertebrates - Part 4: Sampling, extraction and identification of soil-inhabiting nematodes (ISO 23611-4:2022)

Qualité du sol - Prélèvement des invertébrés du sol -  
Partie 4 : Prélèvement, extraction et identification des  
nématodes du sol (ISO 23611-4:2022)

Bodenbeschaffenheit - Probenahme von Wirbellosen  
im Boden - Teil 4: Probenahme, Extraktion und  
Bestimmung von Boden bewohnenden Nematoden  
(ISO 23611 4:2022)

This European Standard was approved by CEN on 16 July 2022.

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, 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, Türkiye and United Kingdom.



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

CEN-CENELEC Management Centre: Rue de la Science 23, B-1040 Brussels

Contents	Page
European foreword.....	3

**iTeh STANDARD PREVIEW**  
**(standards.iteh.ai)**

SIST EN ISO 23611-4:2023

<https://standards.iteh.ai/catalog/standards/sist/087ed233-87e4-4612-a1d6-43a88279fe4b/sist-en-iso-23611-4-2023>

## European foreword

This document (EN ISO 23611-4:2022) has been prepared by Technical Committee ISO/TC 190 "Soil quality" in collaboration with Technical Committee CEN/TC 444 "Environmental characterization of solid matrices" the secretariat of which is held by NEN.

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 February 2023, and conflicting national standards shall be withdrawn at the latest by February 2023.

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 supersedes EN ISO 23611-4:2011.

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

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, Türkiye and the United Kingdom.

(standards.iteh.ai)

## Endorsement notice

The text of ISO 23611-4:2022 has been approved by CEN as EN ISO 23611-4:2022 without any modification.



INTERNATIONAL  
STANDARD

ISO  
23611-4

Second edition  
2022-08

---

---

**Soil quality — Sampling of soil  
invertebrates —**

**Part 4:  
Sampling, extraction and  
identification of soil-inhabiting  
nematodes**

*Qualité du sol — Prélèvement des invertébrés du sol —*

*Partie 4: Prélèvement, extraction et identification des nématodes du  
sol*

[SIST EN ISO 23611-4:2023](https://standards.iteh.ai/catalog/standards/sist/087ed233-87e4-4612-a1d6-43a88279fe4b/sist-en-iso-23611-4-2023)

<https://standards.iteh.ai/catalog/standards/sist/087ed233-87e4-4612-a1d6-43a88279fe4b/sist-en-iso-23611-4-2023>



Reference number  
ISO 23611-4:2022(E)

© ISO 2022

# iTeh STANDARD PREVIEW (standards.iteh.ai)

SIST EN ISO 23611-4:2023

<https://standards.iteh.ai/catalog/standards/sist/087ed233-87e4-4612-a1d6-43a88279fe4b/sist-en-iso-23611-4-2023>



## **COPYRIGHT PROTECTED DOCUMENT**

© ISO 2022

All rights reserved. Unless otherwise specified, or required in the context of its implementation, no part of this publication may be reproduced or utilized otherwise in any form or by any means, electronic or mechanical, including photocopying, or posting on the internet or an intranet, without prior written permission. Permission can be requested from either ISO at the address below or ISO's member body in the country of the requester.

ISO copyright office  
CP 401 • Ch. de Blandonnet 8  
CH-1214 Vernier, Geneva  
Phone: +41 22 749 01 11  
Email: [copyright@iso.org](mailto:copyright@iso.org)  
Website: [www.iso.org](http://www.iso.org)

Published in Switzerland



<b>Contents</b>		Page
<b>Foreword</b> .....		<b>iv</b>
<b>Introduction</b> .....		<b>v</b>
<b>1 Scope</b> .....		<b>1</b>
<b>2 Normative references</b> .....		<b>1</b>
<b>3 Terms and definitions</b> .....		<b>1</b>
<b>4 Principle</b> .....		<b>2</b>
<b>5 Reagents</b> .....		<b>3</b>
<b>6 Apparatus</b> .....		<b>3</b>
6.1 Sampling.....		3
6.2 Extraction.....		4
6.3 Counting.....		4
6.4 Fixation and preparation of mass slides.....		5
6.5 Identification.....		5
<b>7 Procedure</b> .....		<b>5</b>
7.1 General.....		5
7.2 Sampling.....		5
7.3 Extraction.....		6
7.4 Counting.....		7
7.5 Fixation and preparation of mass slides.....		8
7.6 Identification.....		8
<b>8 Data assessment</b> .....		<b>8</b>
<b>9 Test report</b> .....		<b>9</b>
<b>Annex A (informative) Figures of equipment and methods for nematological research</b> .....		<b>11</b>
<b>Annex B (informative) Information about the availability of the Oostenbrink elutriator</b> .....		<b>14</b>
<b>Annex C (informative) Information about the Baermann funnel/tray extraction method</b> .....		<b>17</b>
<b>Annex D (informative) Examples of the use of soil invertebrates in soil monitoring programmes (including presentation of their results)</b> .....		<b>19</b>
<b>Bibliography</b> .....		<b>24</b>

## ISO 23611-4:2022(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 190, *Soil quality*, Subcommittee SC 4, *Biological characterization*, in collaboration with the European Committee for Standardization (CEN) Technical Committee CEN/TC 444, *Environmental characterization of solid matrices*, in accordance with the Agreement on technical cooperation between ISO and CEN (Vienna Agreement).

This second edition cancels and replaces the first edition (ISO 23611-4:2007), which has been technically revised. The main changes are as follows:

- examples of the use of nematodes in soil monitoring programmes have been added (including presentation of their results) as an informative annex (see [Annex D](#)).

A list of all parts in the ISO 23611 series can be found on the ISO website.

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

This document has been drawn up since there is a growing need for the standardization of terrestrial zoological field methods. Such methods, mainly covering the sampling, extraction and handling of soil invertebrates, are necessary for the following purposes:

- biological classification of soils including soil quality assessment [37],[42],[57];
- terrestrial bio-indication and long-term monitoring [25],[28],[31],[50];
- evaluation of the effects of chemicals on soil animals in the field (see ISO 11268-3[4]).

Data for these purposes are gained by standardized methods since they can form the basis for far-reaching decisions (e.g. whether a given site should be remediated or not). In fact, the lack of such standardized methods is one of the most important reasons why bio-classification and bio-assessment in terrestrial (i.e. soil) habitats has so far been relatively rarely used in comparison to aquatic sites.

Nematodes are an important and major part of the soil fauna. Some authors estimate that this group is probably the most dominant one of the multicellular organisms (Metazoa) on earth[52]. Nematodes occur from the Antarctic to the tropics and from deep sea sediments to mountain regions. They are active in every place with sufficient water and organic material. The species diversity and functional variety are impressive[14]. Nematodes are commonly known as parasites of animals and plants, but the major part of the nematode fauna participates in decomposition processes by feeding on bacteria and fungi.

Nematodes occur in high numbers ( $0,2 \times 10^6 \text{ m}^{-2}$  to  $9 \times 10^6 \text{ m}^{-2}$ ) and with a high (10 to 100 species) diversity in almost every soil sample[12]. Moreover, there is a broad ecological spectrum of feeding types and food web relations among the nematodes such as bacterivores, fungivores, herbivores, predators and omnivores[57],[58]. These factors make the group highly suitable as indicators for ecological soil quality[56], but standardization of methods is urgently needed for comparison and combination of results.

In the past 100 years, nematology has developed strongly from the viewpoint of agriculture, advisory sampling and phytosanitary regulations because some terrestrial nematodes cause a lot of damage in crops. With respect to methods, there are several “schools” in different parts of the world with their own history, practical advantages and disadvantages. A comprehensive overview is given by Oostenbrink[14] and Southey[48],[49]. The more recently described methods (or variants) are often developed with special interest to certain plant parasitic species. Within the past 20 years new methods have evolved that allow a DNA-based taxonomic identification of nematode species[21],[34],[54]. This opens the taxonomic analysis of nematodes to a broader community of non-specialists.

Since Bongers[16] introduced the Maturity Index, the use of nematodes in bio-indication for soil quality has increased rapidly[56]. Nematodes are now used for ecological soil research and monitoring in several countries all over the world. Monitoring activities make special demands on methodology, for instance, that a large number of soil samples is processed on a routine basis against reasonable costs. Some of the methods originally developed for advisory sampling in agriculture are very suitable for ecological research. They form the basis for specific variants described in this document.

The nematodes that are characterized by the proposed procedure are all the free-living forms of nematodes found in soil. They include non-plant-feeding nematodes as well as ectoparasitic plant-feeding nematodes and free-living stage of endoparasitic nematodes. The quantification of obligate plant-feeding nematodes in roots requires specific methods. Basic information on the ecology of nematodes and their use as bio-indicators can be found in the bibliography.

