



# SLOVENSKI STANDARD SIST EN ISO 11267:2024

01-junij-2024

Nadomešča:  
SIST EN ISO 11267:2014

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**Kakovost tal - Zaviranje razmnoževanja vrste Folsomia candida iz rodu skakačev (Collembola) zaradi onesnaževal v tleh (ISO 11267:2023)**

Soil quality - Inhibition of reproduction of Collembola (Folsomia candida) by soil contaminants (ISO 11267:2023)

Bodenbeschaffenheit - Hemmung der Reproduktion von Collembolen (Folsomia candida) durch Verunreinigungen (ISO 11267:2023)

Qualité du sol - Inhibition de la reproduction de Collembola (Folsomia candida) par des contaminants du sol (ISO 11267:2023)

**Ta slovenski standard je istoveten z: EN ISO 11267:2023**

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**ICS:**

13.080.30      Biološke lastnosti tal      Biological properties of soils

**SIST EN ISO 11267:2024**      en,fr,de



EUROPEAN STANDARD  
NORME EUROPÉENNE  
EUROPÄISCHE NORM

**EN ISO 11267**

August 2023

ICS 13.080.30

Supersedes EN ISO 11267:2014

English Version

**Soil quality - Inhibition of reproduction of Collembola  
(Folsomia candida) by soil contaminants (ISO  
11267:2023)**

Qualité du sol - Inhibition de la reproduction de  
Collembola (*Folsomia candida*) par des contaminants  
du sol (ISO 11267:2023)

Bodenbeschaffenheit - Hemmung der Reproduktion  
von Collembolen (*Folsomia candida*) durch  
Verunreinigungen (ISO 11267:2023)

This European Standard was approved by CEN on 30 July 2023.

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

Contents	Page
European foreword.....	3

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## European foreword

This document (EN ISO 11267:2023) 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of an identical text or by endorsement, at the latest by February 2024, and conflicting national standards shall be withdrawn at the latest by February 2024.

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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INTERNATIONAL  
STANDARD

ISO  
11267

Third edition  
2023-08

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**Soil quality — Inhibition of  
reproduction of *Collembola* (*Folsomia  
candida*) by soil contaminants**

*Qualité du sol — Inhibition de la reproduction de *Collembola*  
(*Folsomia candida*) par des contaminants du sol*

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# Contents

Page

Foreword.....	iv
Introduction.....	v
<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>3</b>
<b>5 Reagents and material.....</b>	<b>4</b>
<b>6 Apparatus.....</b>	<b>6</b>
<b>7 Procedure.....</b>	<b>6</b>
7.1 Experimental design.....	6
7.1.1 General.....	6
7.1.2 Range-finding test.....	6
7.1.3 Definitive test.....	7
7.2 Preparation of test mixture.....	7
7.2.1 Testing contaminated soil.....	7
7.2.2 Testing substances added to the test substrate.....	8
7.2.3 Preparation of control container.....	8
7.3 Addition of the biological material.....	9
7.4 Test conditions and measurements.....	9
7.5 Determination of surviving Collembola.....	9
<b>8 Calculation and expression of results.....</b>	<b>9</b>
8.1 Calculation.....	9
8.2 Expression of results.....	9
<b>9 Validity of the test.....</b>	<b>10</b>
<b>10 Statistical analysis.....</b>	<b>10</b>
10.1 General.....	10
10.2 Single-concentration tests.....	10
10.3 Multi-concentration tests.....	11
10.3.1 Range-finding test.....	11
10.3.2 Definitive test.....	11
<b>11 Test report.....</b>	<b>12</b>
<b>Annex A (informative) Techniques for rearing and breeding <i>Folsomia candida</i>.....</b>	<b>13</b>
<b>Annex B (normative) Determination of water-holding capacity.....</b>	<b>15</b>
<b>Annex C (informative) Guidance on adjustment of pH of artificial soil.....</b>	<b>16</b>
<b>Annex D (informative) Extraction and counting of Collembola.....</b>	<b>17</b>
<b>Annex E (informative) Specific information of alternative Collembolan species other than <i>Folsomia candida</i>.....</b>	<b>18</b>
<b>Bibliography.....</b>	<b>32</b>

## ISO 11267:2023(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 document 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)).

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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 third edition cancels and replaces the second edition (ISO 11267:2014), which has been technically revised.

The main change is as follows:

- addition of an annex to provide specific information when using alternative Collembola species for reproduction test.

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

Ecotoxicological test systems are applied to obtain information about the effects of contaminants in soil and are proposed to complement conventional chemical analysis (see References [2] and [4]). Reference [2] includes a list and short characterization of recommended and standardized test systems and Reference [4] gives guidance on the choice and evaluation of the bioassays. Aquatic test systems with soil eluate are applied to obtain information about the fraction of contaminants potentially reaching the groundwater by the water path (retention function of soils), whereas terrestrial test systems are used to assess the habitat function of soils.

Soil-dwelling Collembola are ecologically relevant species for ecotoxicological testing. Springtails are prey animals for a variety of endogeic and epigeic invertebrates and they contribute to decomposition processes in soils. In acidic soils they are probably the most important soil invertebrates besides enchytraeids with respect to that function, since earthworms are typically absent.<sup>[19]</sup> Additionally, Collembola represent arthropod species with a different route and a different rate of exposure compared to earthworms<sup>[1]</sup> and enchytraeids.<sup>[3]</sup> Various species were used in bioassays of which four species were used most commonly, *Folsomia candida* Willem, *Folsomia fimetaria* L., *Onychiurus armatus*, and *Orchesella cincta*.<sup>[20]</sup> Numerous soil toxicity tests supported by Environment Canada (EC) resulted in the development and standardization of a biological test method for determining the lethal and sublethal toxicity of samples of contaminated soil to Collembola.<sup>[10]</sup> The method prepared by EC includes four species, *Orthonychiurus folsomi*, *Proisotoma minuta*, *F. candida*, and *F. fimetaria*. As standardized test systems using Collembola as indicator organisms for the habitat function of soil, another two methods exist. One is designed for assessing the effects of substances on the reproductive output of the Collembola, *F. fimetaria* and *F. candida* in soil<sup>[19],[21]</sup>, and the other method described here, focuses on testing contaminated soil. Optionally the method can be used for testing substances added to standard soils (e.g. artificial soil) for their sublethal hazard potential to Collembola.

This document describes a method that is based on the determination of sublethal effects of contaminated soils to adult Collembola of the species *Folsomia candida* Willem. The species is distributed worldwide. It plays a similar ecological role to *F. fimetaria*<sup>[10],[19]</sup>. *F. candida* reproduces parthenogenetically and is an easily accessible species as it is commercially available and easy to culture. *F. candida* is considered to be a representative of soil arthropods and Collembola in particular. Background information on the ecology of springtails and their use in ecotoxicological testing is available in Reference [22].

Distinct Collembolan species inhabit various ecological niches at different soil depths and in different soil types across the globe. Although considered a surrogate species and therefore frequently used in ecotoxicological reproduction tests, *F. candida* is not common in most natural soils.<sup>[28]</sup> Furthermore, species specific morphological adaptations can influence exposure and toxic effects of chemicals on organisms.<sup>[102]</sup> Thus, the use of a variety of Collembolan species representing different morphological adaptations can be advantageous to obtain a broad spectrum of sensitivities for this group. Therefore, other species like *F. fimetaria* (euedaphic, distributed worldwide and found in agricultural soils<sup>[28]</sup>), *Onychiurus yodai* (an euedaphic Asian species,<sup>[31]</sup> *Proisotoma minuta* (hemiedaphic, distributed worldwide and inhabiting agricultural soils<sup>[31],[36]</sup>), *Protaphorura fimata* (euedaphic, occurring through mild temperate to cold zones<sup>[31],[37]</sup>), and *Sinella curviseta* (epedaphic, distributed from North America to Europe, Southeast Asia and Japan<sup>[42]</sup>) were added as potential alternative test species (Annex E). These species have been used as ecotoxicological test species before, but available testing experience is limited.

Effects of substances are assessed using a standard soil, preferably a defined artificial soil substrate. For contaminated soils, the effects are determined in the soil to be tested and in a control soil. According to the objective of the study, the control and dilution substrate (dilution series of contaminated soil) are either an uncontaminated soil comparable to the soil to be tested (reference soil) or a standard soil (e.g. artificial soil).

**NOTE** The stability of the test substance cannot be ensured over the test period. No provision is made in the test method for monitoring the persistence of the substance under test.



# Soil quality — Inhibition of reproduction of *Collembola (Folsomia candida)* by soil contaminants

## 1 Scope

This document specifies one of the methods for evaluating the habitat function of soils and determining effects of soil contaminants and substances on the reproduction of *Folsomia candida* Willem by dermal and alimentary uptake. This document also provides information on how to use this method for testing substances under temperate conditions.

The chronic test described is applicable to soils and soil materials of unknown quality, e.g. from contaminated sites, amended soils, soils after remediation, industrial, agricultural or other sites of concern and waste materials.

The method is not applicable to volatile substances, i.e. substances for which  $H$  (Henry's constant) or the air/water partition coefficient is greater than 1, or for which the vapour pressure exceeds 300 Pa at 25 °C.

## 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 10390, *Soil, treated biowaste and sludge – Determination of pH*

ISO 10694, *Soil quality — Determination of organic and total carbon after dry combustion (elementary analysis)*

ISO 11260, *Soil quality — Determination of effective cation exchange capacity and base saturation level using barium chloride solution*

ISO 11277, *Soil quality — Determination of particle size distribution in mineral soil material — Method by sieving and sedimentation*

ISO 11465, *Soil quality — Determination of dry matter and water content on a mass basis — Gravimetric method*

ISO 18400-206, *Soil quality — Sampling — Part 206: Collection, handling and storage of soil under aerobic conditions for the assessment of microbiological processes, biomass and diversity in the laboratory*

## 3 Terms and definitions

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

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

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

### 3.1

#### **contaminant**

substance or agent present in the soil as a result of human activity

## ISO 11267:2023(E)

### 3.2

#### EC<sub>x</sub>

effect concentration for  $x$  % effect

concentration (mass fraction) of a test sample or a test substance that causes  $x$  % of an effect on a given end-point within a given exposure period when compared with a control

**EXAMPLE** An EC<sub>50</sub> is a concentration estimated to cause an effect on a test end-point in 50 % of an exposed population over a defined exposure period.

Note 1 to entry: The EC<sub>x</sub> is expressed as a percentage of soil to be tested (dry mass) per soil mixture (dry mass). When substances are tested, the EC<sub>x</sub> is expressed as mass of the test substance per dry mass of soil in milligrams per kilogram.

### 3.3

#### ER<sub>x</sub>

effect rate for  $x$  % effect

rate of a contaminated soil that causes  $x$  % of an effect on a given end-point within a given exposure period when compared with a control

### 3.4

#### limit test

single concentration test consisting of at least four replicates each, the soil to be tested without any dilution or the highest concentration of test substance mixed into the *control soil* (3.11) and the control

### 3.5

#### LOEC

#### lowest observed effect concentration

lowest test substance concentration that has a statistically significant effect ( $p < 0,05$ ) when compared with the control

Note 1 to entry: In this test, the LOEC is expressed as a mass of test substance per dry mass of the soil to be tested. All test concentrations above the LOEC should usually show an effect that is statistically different from the control.

### 3.6

#### LOER

lowest observed effect rate

lowest rate of a contaminated soil tested in a *control soil* (3.11) that has a statistically significant effect ( $p < 0,05$ ) when compared with the control

### 3.7

#### NOEC

no observed effect concentration

highest test substance concentration immediately below the *LOEC* (3.5) at which no statistically significant effect is observed when compared to the control

Note 1 to entry: In this test, the concentration corresponding to the NOEC has no statistically significant effect ( $p < 0,05$ ) within a given exposure period when compared with the control.

### 3.8

#### NOER

#### no observed effect rate

highest rate of a contaminated soil to be tested immediately below the *LOER* (3.6) at which no statistically significant effect is observed when compared to the control

### 3.9

#### reference soil

uncontaminated soil with comparable pedological properties (nutrient concentrations, pH, organic carbon content and texture) to the soil being studied