



NSAI
Standards

Irish Standard
I.S. EN ISO 11268-2:2015

Soil quality - Effects of pollutants on earthworms - Part 2: Determination of effects on reproduction of *Eisenia fetida*/*Eisenia andrei* (ISO 11268-2:2012)

I.S. EN ISO 11268-2:2015

Incorporating amendments/corrigenda/National Annexes issued since publication:

The National Standards Authority of Ireland (NSAI) produces the following categories of formal documents:

I.S. xxx: Irish Standard — national specification based on the consensus of an expert panel and subject to public consultation.

S.R. xxx: Standard Recommendation — recommendation based on the consensus of an expert panel and subject to public consultation.

SWiFT xxx: A rapidly developed recommendatory document based on the consensus of the participants of an NSAI workshop.

This document replaces/revises/consolidates the NSAI adoption of the document(s) indicated on the CEN/CENELEC cover/Foreword and the following National document(s):

NOTE: The date of any NSAI previous adoption may not match the date of its original CEN/CENELEC document.

This document is based on:

EN ISO 11268-2:2015

Published:

2015-08-12

This document was published under the authority of the NSAI and comes into effect on:

2015-08-29

ICS number:

13.080.30

NOTE: If blank see CEN/CENELEC cover page

NSAI
1 Swift Square,
Northwood, Santry
Dublin 9

T +353 1 807 3800
F +353 1 807 3838
E standards@nsai.ie
W NSAI.ie

Sales:
T +353 1 857 6730
F +353 1 857 6729
W standards.ie

Údarás um Chaighdeáin Náisiúnta na hÉireann

National Foreword

I.S. EN ISO 11268-2:2015 is the adopted Irish version of the European Document EN ISO 11268-2:2015, Soil quality - Effects of pollutants on earthworms - Part 2: Determination of effects on reproduction of *Eisenia fetida*/*Eisenia andrei* (ISO 11268-2:2012)

This document does not purport to include all the necessary provisions of a contract. Users are responsible for its correct application.

Compliance with this document does not of itself confer immunity from legal obligations.

In line with international standards practice the decimal point is shown as a comma (,) throughout this document.

This page is intentionally left blank

EUROPEAN STANDARD

EN ISO 11268-2

NORME EUROPÉENNE

EUROPÄISCHE NORM

August 2015

ICS 13.080.30

English Version

**Soil quality - Effects of pollutants on earthworms - Part 2:
Determination of effects on reproduction of *Eisenia fetida*/*Eisenia
andrei* (ISO 11268-2:2012)**

Qualité du sol - Effets des polluants vis-à-vis des vers de terre - Partie 2: Détermination des effets sur la reproduction de *Eisenia fetida*/*Eisenia andrei* (ISO 11268-2:2012)

Bodenbeschaffenheit - Wirkungen von Schadstoffen auf Regenwürmer - Teil 2: Bestimmung der Wirkung auf die Reproduktionsleistung von *Eisenia fetida*/*Eisenia andrei* (ISO 11268-2:2012)

This European Standard was approved by CEN on 6 August 2015.

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, 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

EN ISO 11268-2:2015 (E)

Contents	Page
European foreword.....	3

European foreword

The text of ISO 11268-2:2012 has been prepared by Technical Committee ISO/TC 190 “Soil quality” of the International Organization for Standardization (ISO) and has been taken over as EN ISO 11268-2:2015 by Technical Committee CEN/TC 345 “Characterization of soils” 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 2016, and conflicting national standards shall be withdrawn at the latest by February 2016.

Attention is drawn to the possibility that some of the elements of this document may be the subject of patent rights. CEN [and/or CENELEC] shall not be held responsible for identifying any or all such patent rights.

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, Slovakia, Slovenia, Spain, Sweden, Switzerland, Turkey and the United Kingdom.

Endorsement notice

The text of ISO 11268-2:2012 has been approved by CEN as EN ISO 11268-2:2015 without any modification.

This page is intentionally left blank

**Soil quality — Effects of pollutants on
earthworms —**

Part 2:

**Determination of effects on reproduction
of *Eisenia fetida*/*Eisenia andrei***

Qualité du sol — Effets des polluants vis-à-vis des vers de terre —

*Partie 2: Détermination des effets sur la reproduction de Eisenia fetida/
Eisenia andrei*





COPYRIGHT PROTECTED DOCUMENT

© ISO 2012

All rights reserved. Unless otherwise specified, no part of this publication may be reproduced or utilized in any form or by any means, electronic or mechanical, including photocopying and microfilm, without permission in writing from either ISO at the address below or ISO's member body in the country of the requester.

ISO copyright office
Case postale 56 • CH-1211 Geneva 20
Tel. + 41 22 749 01 11
Fax + 41 22 749 09 47
E-mail copyright@iso.org
Web www.iso.org

Published in Switzerland

Contents		Page
Foreword		iv
Introduction		v
1 Scope		1
2 Normative references		1
3 Terms and definitions		2
4 Principle		3
5 Reagents and material		4
6 Apparatus		5
7 Procedure		6
7.1 Experimental design		6
7.2 Preparation of test mixture		7
7.3 Addition of the earthworms		8
7.4 Test conditions and measurements		8
7.5 Reference substance		9
8 Calculation and expression of results		9
8.1 Calculation		9
8.2 Expression of results		9
9 Validity of the test		9
10 Statistical analysis		9
10.1 General		9
10.2 Single-concentration tests		10
10.3 Multi-concentration tests		10
11 Test report		11
Annex A (informative) Determination of the chronic toxicity of chemicals on earthworms under tropical conditions		13
Annex B (informative) Culturing of <i>Eisenia fetida</i> and <i>Eisenia andrei</i>		15
Annex C (informative) Determination of water holding capacity of artificial soil		16
Annex D (informative) Techniques for counting juvenile worms hatched from cocoons		17
Annex E (informative) Performance of the method		18
Bibliography		20

ISO 11268-2:2012(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.

International Standards are drafted in accordance with the rules given in the ISO/IEC Directives, Part 2.

The main task of technical committees is to prepare International Standards. Draft International Standards adopted by the technical committees are circulated to the member bodies for voting. Publication as an International Standard requires approval by at least 75 % of the member bodies casting a vote.

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.

ISO 11268-2 was prepared by Technical Committee ISO/TC 190, *Soil quality*, Subcommittee SC 4, *Biological methods*.

This second edition cancels and replaces the first edition (ISO 11268-2:1998), which has been technically revised.

ISO 11268 consists of the following parts, under the general title *Soil quality — Effects of pollutants on earthworms*:

- *Part 1: Determination of acute toxicity to Eisenia fetida/Eisenia andrei*
- *Part 2: Determination of effects on reproduction of Eisenia fetida/Eisenia andrei*
- *Part 3: Guidance on the determination of effects in field situations*

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 ISO 15799 ^[34] and ISO 17616 ^[35]). ISO 15799 includes a list and short characterization of recommended and standardized test systems and ISO 17616 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. As standardized test systems using earthworms as indicator organisms for the habitat function of soil, an acute test for survival and a chronic test for reproduction are available.

This part of ISO 11268 describes a method that is based on the determination of sublethal effects of contaminated soils on adult earthworms of the species *Eisenia fetida* (Savigny 1826) and *Eisenia andrei* (André 1963). Optionally, the method can be used for testing chemicals added to standard soils (e.g. artificial soil) for their sublethal hazard potential to earthworms. Finally, information is provided on how to use this method for testing chemicals under tropical conditions (see Annex A).

Eisenia fetida and *Eisenia andrei* are considered to be representatives of soil fauna and earthworms in particular. Background information on the ecology of earthworms and their use in ecotoxicological testing is available. Other species, e.g. *Aporrectodea caliginosa*, *Lumbricus rubellus* and *Lumbricus terrestris*, have also been used as test organisms. These or other species have not been proven to be more sensitive in general, and the database and experience in testing soils is small ^{[16][17]}.

This part of ISO 11268 has been drawn up taking into consideration test procedures adopted by the Organization for Economic Cooperation and Development ^{[27][28]} and by the European Union ^[11].

Soil quality — Effects of pollutants on earthworms —

Part 2:

Determination of effects on reproduction of *Eisenia fetida*/ *Eisenia andrei*

WARNING — Contaminated soils may contain unknown mixtures of toxic, mutagenic, or otherwise harmful chemicals or infectious microorganisms. Occupational health risks may arise from dust or evaporated chemicals during handling and incubation. Precautions should be taken to avoid skin contact.

1 Scope

This part of ISO 11268 specifies one of the methods for evaluating the habitat function of soils and determining the effects of soil contaminants and chemicals on the reproduction of *Eisenia fetida*/*Eisenia andrei* by dermal and alimentary uptake. This chronic test is applicable to soils and soil materials of unknown quality, e.g. from contaminated sites, amended soils, soils after remediation, agricultural or other sites concerned, and waste materials.

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

Information is provided on how to use this method for testing chemicals under temperate as well as under tropical conditions.

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 0,013 3 Pa at 25 °C.

This method does not take into account the persistence of the substance during the test.

2 Normative references

The following referenced documents are indispensable for the application 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 10381-6, *Soil quality — Sampling — Part 6: Guidance on the collection, handling and storage of soil under aerobic conditions for the assessment of microbiological processes, biomass and diversity in the laboratory*

ISO 10390, *Soil quality — 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 11268-1, *Soil quality — Effects of pollutants on earthworms — Part 1: Determination of acute toxicity to Eisenia fetida/Eisenia andrei*

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*

This is a free preview. Purchase the entire publication at the link below:

[Product Page](#)

-
- [Looking for additional Standards? Visit Intertek Inform Infostore](#)
 - [Learn about LexConnect, All Jurisdictions, Standards referenced in Australian legislation](#)
-