

**Soil Quality - Determination of dehydrogenase activity in  
soils - Part 2: Method using iodotetrazolium chloride  
(INT) (ISO 23753-2:2005)**

## EESTI STANDARDI EESSÕNA

## NATIONAL FOREWORD

Käesolev Eesti standard EVS-EN ISO 23753-2:2011 sisaldab Euroopa standardi EN ISO 23753-2:2011 ingliskeelset teksti.

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Euroopa standardimisorganisatsioonide poolt rahvuslikele liikmetele Euroopa standardi teksti kättesaadavaks tegemise kuupäev on 13.07.2011.

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ICS 13.080.30

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English Version

**Soil Quality - Determination of dehydrogenase activity in soils -  
Part 2: Method using iodotetrazolium chloride (INT) (ISO 23753-  
2:2005)**

Qualité du sol - Détermination de l'activité des  
déshydrogénases dans les sols - Partie 2: Méthode au  
chlorure de iodotétrazolium (CIT) (ISO 23753-2:2005)

Bodenbeschaffenheit - Bestimmung der  
Dehydrogenaseaktivität in Böden - Teil 2: Verfahren mit  
Iodotetrazoliumchlorid (INT) (ISO 23753-2:2005)

This European Standard was approved by CEN on 17 June 2011.

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EUROPÄISCHES KOMITEE FÜR NORMUNG

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## Foreword

The text of ISO 23753-2:2005 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 23753-2:2011 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of an identical text or by endorsement, at the latest by January 2012, and conflicting national standards shall be withdrawn at the latest by January 2012.

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### Endorsement notice

The text of ISO 23753-2:2005 has been approved by CEN as a EN ISO 23753-2:2011 without any modification.

## Contents

Page

Foreword .....	iv
Introduction .....	v
1     Scope .....	1
2     Normative references .....	1
3     Principle .....	1
4     Reagents and materials .....	2
5     Apparatus .....	3
6     Procedure .....	3
7     Calculation .....	3
8     Test report .....	4
Bibliography .....	5

## Introduction

The soil microflora is responsible for the decomposition and conversion of organic substances, aggregation stability and the carbon, nitrogen, sulfur and phosphorus cycles. Dehydrogenases, as respiratory chain enzymes, play a major role in the energy production by organisms. They oxidize organic compounds by transferring two hydrogen atoms. Dehydrogenases are essential components of the enzyme system of microorganisms. Dehydrogenase activity can therefore be used as an indicator of biological redox systems and as a measure of microbial activity in the soil.

# Soil quality — Determination of dehydrogenase activity in soils —

## Part 2: Method using iodotetrazolium chloride (INT)

### 1 Scope

This part of ISO 23753 specifies a method for determining soil dehydrogenase activity using 2-(4-iodophenyl)-3-(4-nitrophenyl)-5-phenyltetrazolium chloride (INT). As the INT reduction is less sensitive to O<sub>2</sub>, the method is more reproducible than the TTC-method described in ISO 23753-1.

It is not applicable for determining the dehydrogenase activity in upper layers (L, F, H horizons) of forest humus forms with low microbial activity (e.g. mor), or in soils showing reducing properties (e.g. waterlogged soils).

### 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 for the assessment of aerobic microbial processes in the laboratory*

ISO 10390, *Soil quality — Determination of pH*

ISO 11259, *Soil quality — Simplified soil description*

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

### 3 Principle

INT solution is added to a soil sample and the mixture is incubated at 30 °C for 18 h. The iodonitrotetrazolium formazan (INTF) released is extracted with acetone (in the case of humic soil) and determined by photometry at a wavelength of 485 nm.

NOTE 1 The method is based on a modified version of the method reported in Reference [1].

NOTE 2 Acetone is used as extractant and samples are not extracted to completion.

In the case of soil having reducing characteristics (e.g. waterlogged soil), dehydrogenase activity should not be used as a measure of the biological activity in the soil [3]. Abiotic components, such as iron(II) compounds or sulfides can reduce INT.