
**Soil quality — Determination of the
effects of pollutants on soil flora —**

Part 2:

**Effects of chemicals on the emergence
and growth of higher plants**

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*Qualité du sol — Détermination des effets des polluants sur la flore du
sol —*

*Partie 2: Effets des substances chimiques sur l'émergence et la
croissance des végétaux supérieurs*

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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.

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 11269-2 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:11269-2:1995) which has been technically revised.

ISO 11269 consists of the following parts, under the general title *Soil quality — Determination of the effects of pollutants on soil flora*:

- *Part 1: Method for the measurement of inhibition of root growth*
- *Part 2: Effects of chemicals on the emergence and growth of higher plants*

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Introduction

This part of ISO 11269 describes a method for the evaluation of soil quality following the addition of chemicals or after contamination of an unknown type. Before assessing the effects on plant growth of a chemical incorporated in soil, information on the solubility in water and in organic solvents, and the vapour pressure of the test substance is recommended. Preferably, the molecular formula, partition coefficient (water/octanol), and chemical and biological stability should be available to the laboratory. All physical and biological parameters should be considered when interpreting the results of the test.

The test, as written, assesses the effect on emergence and plant growth of a chemical incorporated in soil. In case of contaminated soil, the individual chemicals are unidentified and therefore correct information on solubility, vapour pressure and molecular formula etc. cannot be selected. No incorporation is recommended, but it may be necessary to dilute the contaminated soil with an uncontaminated control soil or sand before testing.

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Soil quality — Determination of the effects of pollutants on soil flora —

Part 2: Effects of chemicals on the emergence and growth of higher plants

1 Scope

This part of ISO 11269 describes a method that is applicable to the determination of possible toxic effects of solid or liquid chemicals incorporated in soil on the emergence and early stages of growth and development of a variety of terrestrial plants. It does not give an indication of damage resulting from direct contact of seedlings with the chemical in the vapour or liquid phase outside the soil environment.

The method is also applicable to the comparison of soils of known and unknown quality. Information on how to adapt the method for this purpose is given in Annex B.

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

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

3 Terms and definitions

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

3.1

emergence

appearance of the coleoptile or cotyledon above the soil

3.2

lowest observed effect concentration

LOEC

lowest concentration used in the test at which the substance is observed to have a statistically significant effect on emergence or growth as compared with the control

NOTE All test concentrations above the LOEC have a harmful effect equal to or greater than those observed at the LOEC.

3.3

no observed effect concentration

NOEC

test concentration immediately below the LOEC, which when compared to the control has no statistically significant effect ($p > 0,05$)

NOTE See also LOEC.

3.4

visual assessment

rating of visual damage based on observations of plant stand, vigour, malformation, chlorosis, necrosis and overall appearance compared with a control

NOTE Reference [2] in the Bibliography.

3.5

x % effect concentration

EC_x

concentration that results in an undesirable change or alteration of x % in the test endpoint being measured relative to the control (e.g. 25 % reduction in seedling emergence, shoot mass or increase in visual injury would constitute an EC₂₅)

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4 Units

Concentrations of test substances are expressed as milligrams per kilogram of dry soil.

Emergence is expressed as the percentage of seedlings which emerge compared with the control pots.

Effects on growth are expressed as the difference in mass of the aerial parts of the plants between the treated plants and those in the control pots.

5 Principle

This phytotoxicity test is based on the emergence and early growth response of a variety of terrestrial plant species to various concentrations of a chemical added to the test soil.

Seeds of selected species of plants are planted in pots containing soil to which the test chemical has been added and in control pots. The pots are kept under growth conditions for the test species selected. The emergence and mass (dry or fresh basis) of the shoots of the test plants are compared with those of the control plants.

6 Test plants and materials

6.1 Equipment

Suitable facilities for carrying out the test include phytotrons, plant growth rooms and greenhouses. The planting containers shall be non-porous plastics or glazed pots with a top internal diameter of between 85 mm and 95 mm. The pots should be adjusted to the size of the specific test species.

6.2 Test plants

A minimum of two species shall be selected for the test, comprising at least one species from each of the two categories shown in Table 1. Category 1 comprises monocotyledonous and category 2 dicotyledonous. It is recommended to use more than two plant species.

Table 1 — Test species

Category	Test species
Category 1	
Rye	<i>Secale cereale</i> L., Poaceae
Ryegrass, perennial	<i>Lolium perenne</i> L., Poaceae
Rice	<i>Oryza sativa</i> L., Poaceae
Oat (common or winter)	<i>Avena sativa</i> L., Poaceae
Wheat, soft	<i>Triticum aestivum</i> L., Poaceae
Barley (spring or winter)	<i>Hordeum vulgare</i> L., Poaceae
Sorghum, common (or shatter cane or durra, white or millet, great)	<i>Sorghum bicolor</i> (L.) Moench, Poaceae
Sweet corn	<i>Zea mays</i> L., Poaceae
Category 2	
Mustard, white	<i>Sinapis alba</i> , Brassicaceae
Rape [or rape (summer) or rape (winter)]	<i>Brassica napus</i> (L.) ssp. <i>napus</i> , Brassicaceae
Radish, wild	<i>Raphanus sativus</i> L., Brassicaceae
Turnip, wild	<i>Brassica rapa</i> ssp. <i>rapa</i> (DC.) Metzg., Brassicaceae
Chinese cabbage	<i>Brassica campestris</i> L. Var. <i>Chinensis</i> , Brassicaceae
Birds foot fenugreek	<i>Trifolium ornithopodioides</i> (L.), Fabaceae
Lettuce	<i>Lactuca sativa</i> L., Asteraceae
Cress, garden	<i>Lepidium sativum</i> L., Brassicaceae
Tomato	<i>Lycopersicon esculentum</i> Miller, Solanaceae
Bean	<i>Phaseolus aureus</i> Roxb., Fabaceae