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Soil quality — Guidance on the ecotoxicological characterization of soils and soil materials

Qualité du sol — Lignes directrices relatives à la caractérisation écotoxicologique des sols et des matériaux du sol

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Foreword

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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.

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ISO 15799 was prepared by Technical Committee ISO/TC 190, *Soil quality*, Subcommittee SC 7, *Soil and site assessment*.

Introduction

The majority of existing ecotoxicological test methods (biotests) being internationally harmonized were developed to describe the ecotoxic potential of a test substance when added to a soil or soil material. These methods can be used, with some modification, for the ecotoxicological characterization of soils and soil materials with respect to their function and depending on the intended use. However, in such cases, users of the methods need to be aware that the validation of the methods is not complete.

For substances with properties resulting in toxic effects, biotests are a complement to conventional chemical analysis. Results from chemical analysis can be used for ecotoxicological assessments based on information on the substances identified, including properties of the chemicals, e.g. their bioaccumulation potential. This information is often scarce (if it exists at all) and does not include possible interactions (synergy/antagonism) between chemicals and the complex soil matrix. Furthermore, an exhaustive identification and quantification of substances is impractical. Therefore, ecotoxicological testing of soils can be used for investigating the potential toxicity of complex chemical mixtures. The extrapolation from laboratory tests to field conditions requires adequate consideration of important environmental factors within the test conditions and the selection of suitable ecotoxicological endpoints.

This International Standard is one of a series providing guidance on soils and soil materials in relation to certain functions and uses, including wildlife conservation, and ought to be read in conjunction with those other standards.

Soil quality — Guidance on the ecotoxicological characterization of soils and soil materials

1 Scope

This International Standard provides guidance on the selection of experimental methods for the assessment of the ecotoxic potential of soils and soil materials (e.g. excavated and remediated soils, refills, embankments) with respect to their intended use and possible adverse effects on aquatic and soil-dwelling organisms, and habitat maintenance and the retention function of the soil.

It does not cover tests for bioaccumulation. Genotoxicity tests using eukaryotic organisms in soils are not yet available. It is not applicable to the ecological assessment of uncontaminated soils with a view to natural, agricultural or horticultural use, such soils being of possible interest where they can serve as a reference for the assessment of soils from contaminated sites. Nor is the interpretation of the results gained by application of the proposed methods within its scope.

2 Terms and definitions

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

2.1 Types of soil and other soil materials

2.1.1
soil
upper layer of the Earth's crust composed of mineral particles, organic matter, water, air and organisms

[ISO 11074-1:1996, definition 5.4]

2.1.2
soil material
excavated soil, dredged materials, manufactured soils, treated soils and fill materials

[ISO 15176:2002, definition 3.1.4]

2.1.3
excavated soil
any natural material excavated from the ground, including top soil, sub soil, altered parent rock and parent rock itself

NOTE Excavated soil typically arises during construction works.

[ISO 15176:2002, definition 3.1.5]

2.1.4
standard soil
field-collected soil whose main properties (e.g. pH, texture, organic matter content) are within a known range

EXAMPLE Eurosoils[34].

2.2 Terms relating to soil characteristics

2.2.1
habitat function
ability of soils/soil materials to serve as a habitat for micro-organisms, plants, soil-living animals and their interactions (biocenoses)

2.2.2
retention function
ability of soils/soil materials to adsorb pollutants in such that they cannot be mobilized via the water pathway and translocated into the food chain

NOTE The habitat and retention functions include the following soil functions according to ISO 11074-1:

— control of substance and energy cycles as components of ecosystems;

— basis for the life of plants, animals and humans;

— carrier of genetic reservoir;

— basis for the production of agricultural products;

— buffer inhibiting movement of water, contaminants or other agents into the ground water.

2.2.3
pollutant
substance or agent present in the soil which due to its properties, amount or concentration causes adverse impact on soil functions or soil use

cf. **contaminant** (2.2.4), **potentially harmful substance** (2.2.5)

[ISO 15176:2002, definition 3.2.7]

NOTE See Introduction to ISO 11074-1:1996.

2.2.4
contaminant
substance or agent present in soil as a result of human activity

cf. **pollutant** (2.2.3), **potentially harmful substance** (2.2.5)

NOTE There is no assumption in this definition that harm results from the presence of the contaminant.

[ISO 15176:2002, definition 3.2.6]

2.2.5
potentially harmful substance
substance which, when present in sufficient concentration or amount, may be harmful to humans or the environment

NOTE It may be present as a result of human activity [**contaminant** (2.2.4)] or naturally.

[ISO 15176:2002, definition 3.2.8]

2.3 Land and sites

2.3.1
re-use
useful and harmless utilization of soil materials

NOTE In the context of this International Standard the re-use means the transfer of soil materials to another location for use in agriculture, horticulture, forestry, gardens, recreational areas and construction sites.

[ISO 15176:2002, term 3.4.1]

3 Field of application

3.1 Soils and areas of soil use where ecotoxicological tests should be considered:

— assessment of the ability of a soil to sustain a natural biocenosis or agriculture;

— assessment of the combined ecotoxicity of all bioavailable contaminants present in soils or soil materials;

— assessment of the ecotoxicity of potentially harmful substances in cases where the soil or soil material can affect the ground and surface water;

— identification of soils or soil materials (refills, embankments) having a low degree of contamination — usually within a depth of 1 m — and which can remain at the site without further treatment;

— detection of potential ecotoxicity which could not be traced by chemical analysis;

— monitoring and control of the success of soil treatment (off-site, on-site, *in situ*);

— monitoring and control of soils/soil materials that have been decontaminated and are to be applied at the surface.

3.2 Soils and areas of soil use where ecotoxicological tests are not necessary (provided groundwater contamination can be excluded):

— contaminated soils classified as hazardous waste or which can be characterized clearly by chemical/analytical parameters, in which cases ecotoxicological testing could be useful for a final investigation after remediation and for process control during biological remediation;

— commercially/industrially used areas with no prospect of horticultural/agricultural use;

— soil materials or backfilled materials in an area to be effectively sealed by covering with buildings or other forms of low permeability cover such as concrete or tarmacadam or asphalt.

4 Selection of tests according to use/re-use of soils and soil materials and soil functions

4.1 Use of ecotoxicity tests

Toxicants can affect different species (and in some cases genotypes) present within ecosystems at different concentrations. The ideal approach for the precise ecotoxicological characterization of the soil toxicity is to use a battery of tests with several species belonging to different taxonomic and trophic groups, in order to avoid false negative results owing to an adaptation of a test system (genotypic shift) to a specific contaminant as compared to uncontaminated soils. Studies using field or semi-field investigations are rarely carried out and can be very expensive.

The ideal scheme can be rendered more practicable by the adoption of simpler testing strategies and the application of safety factors to the results obtained. If, however, testing is performed on one species or function only, the high diversity in the sensitivity of species to toxicants will result in a high level of uncertainty. It is therefore recommended to test at least a microbial process, a species from the plant kingdom, and one from the animal kingdom, usually a saprophagous/detritivorous species; if more than one animal species is tested, a predatory species should be included in the test battery. The minimum number of species to be tested depends on the regulations to which the test strategy must comply. This International Standard only gives the basic principles for their use. Further considerations to the selection of tests using soil organisms are given in 4.3.

4.2 General criteria for selection of tests

Criteria for the selection of ecotoxicity tests have been established in the context of hazard assessment and classification of chemicals. These criteria should also apply for the ecotoxicological characterization of contaminated soils. Criteria reviewed were: scientific validity, ecological significance, practicability and acceptability. See [30] and [31].

The basic requirements that test protocols must meet in order to be established in international standards include reproducibility, statistical validity, general acceptance and performance.

The importance of a criterion is relative to the specific situation. Decisions have to be made as to which criteria are the most important or which tests may have to be modified, and choices made between these and more practical considerations — for example, easy culturing of test organisms in the laboratory or the availability of life stages required for a test throughout the year.

The test methods recommended here were originally designed for hazard assessment of chemicals and were in most cases internationally harmonized (e.g. by OECD, EU or ISO). In most of the methods, provisions have been made to adapt the test design to come within the scope of this International Standard. Nevertheless, in many cases experience still has to be gained using the test methods for characterizing soil quality. In addition, the selection of ecotoxicological test methods for the assessment of soils or soil materials depends on their intended use or re-use and on the soil functions to be protected, in particular the retention and habitat functions.

Table 1 gives an example of a decision scheme based on the relevant function.

Table 1 — Relevance for ecotoxicological testing to the intended re-use of the soil

Re-use of soils	Soil function		
	Retention function	Habitat function	
	Aquatic organisms	Plant growth	Soil biocenoses
	Detection of biological effects		
Below sealed areas	low[a]	low	low
Commercially and industrially used unsealed areas	high	low	low
Landfill covering	high	high	low
Green areas, parks and recreation areas	high	high	high
Areas used in horticulture or agriculture	high	high	high

[a] Applies only to the unsaturated soil zone.

4.3 Considerations for the examination of soil functions

4.3.1 Retention function

Transport via water of soluble, colloidal or particle fractions plays a dominant role in the risk assessment of contaminated soils. This is true not only because water may mobilize contaminants, but also because contaminants and metabolites in the water phase potentially have a severe effect on micro-organisms, plants and soil fauna.

Aqueous eluates are useful for testing ecotoxic effects on organisms exposed via the water-mediated transport. It should be taken into account that substances mobilized via water could be subjected to different types of changes, (e.g. metabolism or hydrolization) when transported into the groundwater and from there into surface waters, and that their concentrations are reduced by dilution. Moreover, substances may be mobilized over time due to environmental changes (e.g. pH, chemical and biological transformation). Eluates

may serve as early indicators for the contamination of pore and ground water prior to the exposure of surface and drinking water.

With these aspects, the investigation of groundwater and eluates is of the utmost importance — regardless of the proposed soil use.

4.3.2 Habitat function

4.3.2.1 Representativeness of organisms and processes

The suitability of the soil for living organisms can best be examined by means of test methods selected to include organisms and processes representative of different taxonomic groups.

4.3.2.2 Soil material used as control for bioassays on solid matrices

As a general principle in ecotoxicological testing, any end points measured in a treatment are compared with those measured in the control or controls.

When evaluating the suitability of the soil for soil-dwelling organisms, it is a prerequisite to compare the contaminated soil or soil material with a control material, which may also be used for preparing dilution series with the contaminated sample.

Several types of control material can be used:

— an uncontaminated soil with comparable pedological properties to the sample being tested;

— an inert material (e.g. quartz sand);

— a certified natural soil (e.g. standard soil);

— a standardized artificial soil (see ISO 11267, ISO 11268-1 and ISO 11268-2).

The choice between these control materials should be made depending entirely on the aims of the ecotoxicological assessment, the type of biological test being carried out and the requirements of the test organism. This recommendation cannot be generalized for all biological tests. Adding sand to a soil or a soil material can create a compact mixture incompatible with the growth and development of many organisms (e.g. plant growth tests). It is preferable to use a more complex control material (such as artificial soil) for dilution where this would have the advantage of reproducing more closely the natural environment of the organisms and even if it may interact with pollutants. Placing an organism in a medium that does not match the most important characteristics of its natural habitat may cause stress.

— If a dose-response curve is needed, one of the control materials mentioned above may be used to dilute the contaminated substrate.

— If the aim is to classify each sample of soil or soil materials in terms of ecotoxicity hazard, it is preferable to use an inert material (e.g. quartz sand), which will not interact with the pollutants present in the sample and whose composition and granulometry can be rigorously standardized.

The requirements of the control material must take into account the different soil uses and the type and origin of the soil (e.g. undisturbed soil, refilling material, excavated soil, remediated soil). Nutrient deficiency, as well as physical conditions, can cause differences in plant growth and animal behaviour that need not necessarily be caused by the pollutant situation and the hazard potential.

— If the aim is to evaluate the ecotoxicity of a soil or soil material sample from a contaminated site, the preferred method would be to use an uncontaminated control material similar to the sample being tested.

— If the aim is to evaluate the ecotoxicity of soils or soil materials which may be re-employed for certain specific uses, the preferred method would be to use as a control material any material which may in future be mixed with the soil or soil material.

4.3.2.3 Soil as substrate (medium) for soil micro-organisms

The soil microflora comprises on average 80 % of the mass of organisms living in soil. In combination with the microfauna, the main functions of the microflora are the decomposition and degradation of complex organic substances to easily available nutrients, thereby maintaining the natural substance cycles of carbon, nitrogen, phosphorus and sulfur.

Substrate-induced respiration provides an indicator of the microbial population density.

Nitrifying bacteria, which are responsible for the oxidation of ammonium to nitrite and from nitrite to nitrate, are a very sensitive group of micro-organisms. Reduced nitrification need not necessarily lead to significant changes in the ecosystem but can be used as a sensitive indicator for the inhibition of an essential soil process.

The purpose of determining the microbial biomass or other microbial processes in soils is to allow assessment of the continued maintenance of soil fertility, the potential ability to degrade organic compounds, and the effects of added materials on the soil microbial community.

4.3.2.4 Soil as substrate for plant growth

After micro-organisms, plant roots constitute the largest biological surface in soil. Their contact area with soil particles is increased by the presence of root hairs and mycorrhizal associations (VA-mycorrhiza with cultivated plants and additional ectomycoorhizae with woody plants).

As with the other bioassays proposed, tests with higher plants are designed to assess the bioavailability and effects of pollutants detected or not detected by chemical analysis, respectively. By applying a test period of at least 14 days, short-term changes in the soil by the test plant itself are included.

The accumulation of pollutants in plants, their metabolism and their effects on consumers are not investigated in these tests. They do not apply to the assessment of soil fertility and productivity.

4.3.2.5 Soil as substrate for soil living fauna

Soil animals generally fulfil the following four functions:

a) mechanical activities (drainage, aeration, mixing, mechanical comminution);

b) chemical changes (enhanced availability of nitrate and phosphate from excrements and accelerated formation of clay-humus complexes, after the substrate has passed the gut);

c) biological changes (distribution of micro-organisms in the soil matrix, synergistic effects through stimulation of microbial activity and organic matter decomposition);

d) significant links in the food web.

Short-term and long-term tests are available for examination of the effects of pollutants on soil fauna. For testing the habitat function, characterization by sublethal test parameters is particularly recommended.

Since a single test method cannot adequately represent the vast number of very diverse invertebrates, a test battery should be used. When selecting the individual test species, the following criteria should be considered:

a) trophic level — *e.e.* saprophagous and predatory species should be included;

b) taxonomic/physiological groups — in order to cover the biodiversity of soil communities, at least representatives of annelids and arthropoda have to be selected;

c) size class/exposure pathway: species of the micro-, meso- and macrofauna do not only represent various size classes but also different life-styles and therefore exposure routes (e.g. pore water versus food uptake);

d) ecological role — at least soil-dwelling and litter-inhabiting species are important to consider.

Only internationally standardized methods should be used.

5 Sampling, transport, storage and sample preparation

Before soil quality is assessed by any of the methods proposed, soil samples need to be collected from the site under investigation. Soil sampling should be carried out by trained personnel with sufficient knowledge of sampling, handling of samples and safety measures at contaminated sites and sampling locations. The sampling strategy and handling should be determined by the site to be investigated, the kind of contamination and the aim of the biological tests (e.g. quantities of soil samples could vary between 100 mg and 100 kg, depending on the tests selected).

Record all data concerning sampling, transport and sample preparation. For instructions on the design of sampling programmes, sampling techniques, safety, investigations of natural, cultivated, urban and industrial sites and on the collection, handling and storage of soil for the assessment of aerobic/anaerobic microbial processes in the laboratory, see ISO 10381-6.

6 Limitations of proposed biotests for soils/soil materials

Biological test systems are suitable for volatile pollutants only to a limited extent. Other methods should be developed for this purpose. Similarly, the impact of organic contaminants, which are easily degradable under aerobic conditions, may be detected incompletely by the methods described. In this case, alternative methods for sampling and sample preparation should be applied.

NOTE The proposed terrestrial and aquatic test methods in A.1 and A.2 were developed to assess the ecotoxic potential of chemicals. The characterization of soils or soil eluates was not their primary goal. Therefore, the methods need to be adapted to the specific requirements of soil and site assessment.