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**Karakterizacija odpadkov - Navodilo za uporabo ekotoksikoloških preskusov za odpadke**

Characterization of waste - Guidance on the use of ecotoxicity tests applied to waste

Charakterisierung von Abfällen - Anleitung zur Anwendung von Ökotoxizitätsprüfungen auf Abfälle

Caractérisation des déchets - Recommandations sur l'utilisation des essais d'écotoxicité appliqués aux déchets

**Ta slovenski standard je istoveten z: FprCEN/TR 16110**

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**FprCEN/TR 16110**

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Will supersede CEN/TR 16110:2010

English Version

## Characterization of waste - Guidance on the use of ecotoxicity tests applied to waste

Caractérisation des déchets - Lignes directrices pour l'utilisation des essais d'écotoxicité appliqués aux déchets

Charakterisierung von Abfällen - Anleitung zur Anwendung von Ökotoxizitätsprüfungen auf Abfälle

This draft Technical Report is submitted to CEN members for Vote. It has been drawn up by the Technical Committee CEN/TC 444.

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COMITÉ EUROPÉEN DE NORMALISATION  
EUROPÄISCHES KOMITEE FÜR NORMUNG

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**FprCEN/TR 16110:2023(E)****European foreword**

This document (FprCEN/TR 16110:2023) has been prepared by Technical Committee CEN/TC 444 “Environmental characterization of solid matrices”, the secretariat of which is held by NEN.

This document is currently submitted to the vote on TR.

This document will supersede CEN/TR 16110:2010.

FprCEN/TR 16110:2023 includes the following significant technical changes with respect to CEN/TR 16110:2010:

- The reference to the assessment of hazardous property HP 14 “Ecotoxic” in place of basic ecotoxicological characterization
- The inclusion of an annex that summarizes the national practices to assess the hazardous property HP 14 'Ecotoxic' and the addition of the main conclusions of this survey in subclause 8.1.

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## Introduction

Ecotoxicity can be estimated using two approaches: a chemical-specific approach and a toxicity-based approach. In the first situation, chemical analyses are compared to quality criteria or threshold values to estimate toxicity. In the second case, toxicity is measured directly using toxicity tests. These two approaches complement each other. However, determination of pollutants in complex mixtures of unknown composition (a characteristic of many wastes) does not allow a relevant estimation of toxicity. For such samples, the toxicity-based approach is usually recognized to be the best approach to assess toxicity. Ecotoxicity tests integrate the effects of all contaminants including additive, synergistic and antagonistic effects. They are sensitive to the bioavailable fraction of the contaminants only and integrate the effects of all contaminants, including those, not considered or detected by chemical analyses.

Ecotoxicity tests can be applied to wastes to identify their hazardous properties (*i.e.* HP 14) in accordance with Council Regulation (EU) 2017/997 [22] or to assess the risk related to a site-specific exposure scenario. Determining the hazard classification of waste for hazardous property HP 14 “Ecotoxic” by applying calculation formulae, generic cut-off values, as defined in Regulation (EC) No 1272/2008 [24] is out of the scope of this document.

The majority of existing ecotoxicity tests being internationally harmonized were developed to describe the ecotoxic potential of a test substance when added to water or to soil/soil material, of wastewater or of eluates. These methods can be applied with some modifications for the ecotoxicological characterization of wastes. Nevertheless, users of these methods should be aware that the validation of the methods is not complete. Several studies as well as an International ring test have been conducted to validate some test methods for waste samples and the results have been used as background information.

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## FprCEN/TR 16110:2023(E)

### 1 Scope

Ecotoxicity tests can be applied to wastes to identify their potential hazardous properties with respect to the environment or to assess the risk related to a site-specific exposure scenario. This document provides guidance for the selection and use of ecotoxicity tests for both applications.

This document focuses on the following selected field of applications:

- a) hazardous properties (*i.e.* HP 14);
- b) Site-specific exposure scenario;
- c) Landfill management:
  - a. monitoring of leachates;
  - b. mineral waste going to non-controlled landfill sites.
- d) Re-use of waste:
  - c. use of sludge in agriculture;
  - d. use of mineral waste in road construction.

Other fields of application can also be covered by ecotoxicological testing not being in the scope of the document. The ecotoxicological assessment of waste within other scenarios might need the development of other test strategies.

With regard, more specifically, to the assessment of hazardous properties, this document focuses on the ecotoxicological characterization of waste using biotests.

Depending on the waste type and the assessment goal, relevant criteria are described for the selection of a test strategy and the suitable ecotoxicity test(s).

This document also provides guidance for ecotoxicity test protocols to meet the specific demands of waste testing (e.g., limitations, test design, confounding factors). The proposed tests represent a minimum test battery that can be completed by additional tests or even be replaced by others according to the waste, the intended use or protection goal envisaged.

### 2 Normative references

There are no normative references in this document.



### 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

##### **eluate**

aqueous solution recovered from a leaching test

#### 3.2

##### **waste**

any substance or object in the categories which the holder discards or intends or is required to discard

[Directive 2006/12/EC]

Note 1 to entry: See Annex I.

#### 3.3

##### **leachant**

liquid used in a leaching test

Note 1 to entry: For the purpose of this Technical report the leachant is water

#### 3.4

##### **leaching test**

test during which a waste is put into contact with a leachant and some constituents of the material are extracted

#### 3.5

##### **leachate**

any liquid percolating through the deposited waste and emitted from or contained within a landfill

#### 3.6

##### **inert waste**

waste that does not undergo any significant physical, chemical or biological transformations

Note 1 to entry: Inert waste does not dissolve, burn or otherwise physically or chemically react, biodegrade, or adversely affect other matter with which it comes into contact in a way likely to give rise to environmental pollution or harm human health. The total leachability and pollutant content of the waste and the ecotoxicity of the leachate must be insignificant, and in particular not endanger the quality of surface and/or groundwater. [adapted from Directive 1993/31/EC].

#### 3.7

##### **EC<sub>x</sub> (effective concentration)**

x % effect concentration of the test sample at which the measured effect (e.g., mobility, growth, reproduction) is reduced by x % compared to the control

**FprCEN/TR 16110:2023(E)****3.8****LID (lowest Ineffective Dilution (LID))**

dilution expressed as the reciprocal value of the volume fraction of the test sample in the control and dilution medium

Note 1 to entry: (e.g., if the waste eluate content is 1 in 4 (25 % volume fraction) the dilution level is  $D = 4$ )

**4 General information provided by the content of the guidance document**

This document gives guidance for the selection of a suitable test design e.g., determination of concentration/effect relationship, single concentration test (limit test). Additionally, leaching tests are suggested for each field of application and, for some ecotoxicity tests, information about confounding factors or interactions with the test material is given.

Where test strategy and selection of test methods for basic characterization is favoured by standardization and harmonization, a site-specific scenario might require a more tailor-made approach. Comparisons with other locations and situations become less important, which open the way for the use of less known organisms, specific exposure scenarios or non-standardized methods. Other criteria as sensitivity, practicability and cost efficiency however still holds.

For some site-specific scenarios, choices regarding test strategy and test methods might strongly depend on available information. For example, it is advised to consider the outcome of the assessment before treatment in the re-assessment of treated waste. In other situations, emphasis should be paid to expected changes in the waste material after application. For example, in the reuse of sludge as well as for the reuse of mineral waste, changes in physico-chemical composition after application can be expected which influence availability of toxicants and as a consequence possible ecological effects. It is important that the overall strategy covers these aspects.

In most other cases of site-specific characterization of waste material, the principles for the definition of test strategy and selection of test methods do not differ strongly from those proposed for the site-specific assessment of contaminated soils in general. For guidance on these aspects, reference is therefore made to general textbooks, reviews and guidelines (e.g., ISO 17616, ISO 18772).

**5 Sampling, transport, storage and sample preparation**

Before waste is assessed by any of the methods proposed, samples need to be collected from the site of waste origin and, depending on the potential reuse scenario, from the site of future reapplication. Sampling of waste is carried out by trained operators with sufficient knowledge of sampling, handling of samples and safety measures at waste treatment plants, industrial sites or contaminated locations. The sampling strategy and handling are linked to the waste or site to be investigated, the kind of contamination and the aim of the biological tests.

Instructions on the design of sampling plan, transport, storage and sample preparation are given in the documents EN 14899, CEN/TR 15310-1 to -5 and EN 14735.

Addition of preservatives (e.g., acids, basic solutions, biocides) in order to retard chemical and biological activity is prohibited.

## 6 Leaching procedures for ecotoxicological testing

### 6.1 Overview of methods

#### 6.1.1 General

For the selected fields of application, a key aspect regards the assessment of ecotoxic effects on organisms exposed via the water phase. Indeed, the release of soluble constituents upon contact with water can be regarded as a main mechanism of release which results in a potential risk to the environment during the reuse or disposal of waste materials.

In order to generate a water extract from a solid material, several methods have been developed and a wide variety of test protocols is available in literature. The following subclause does not intend to describe all types of extraction methods but gives some relevant information on the selection of the appropriate method according to the aim of the study.

The leaching of contaminants from waste is controlled by several parameters and external factors. These factors include the physical/chemical nature of the waste especially in terms of pH, reducing properties and degradable organic matter content, the nature of the leachant, the contact time of the leachant with the waste, the particle size and the liquid to solid ratio (L/S). The outcome of which also influences the ecotoxicological response of the waste leachate (see Van der Sloot and van Zomeren, 2009 [16]; Postma et al., 2009 [17]).

European Standards have been developed to investigate mainly inorganic constituents from waste. They do not take into account the particular characteristics of non-polar organic constituents nor the consequences of microbiological processes in organic degradable wastes and need to be adapted in some cases (e.g., Table 3). Some information is provided in EN ISO/TS 21268-3 and EN ISO/TS 21268-4 to address organic contaminants.

Leaching tests, performed to characterize waste materials according to the Landfill Directive, can be divided into three categories: basic characterization, compliance tests and on-site verification tests.

#### 6.1.2 Basic characterization

Leaching tests belonging to this category are used to obtain information on the short- and long-term leaching behaviour and characteristic properties of waste materials. They allow characterizing the source term in accordance with a given scenario which can either be generic or site specific. Liquid/solid (L/S) ratios, leachant composition, factors controlling leachability such as pH, redox potential, complexing capacity and physical parameters are addressed in these tests. They can be subdivided as follows:

- **Parametric tests.** These tests are intended for measuring an intrinsic property of a material or the effects (correlated) of specific parameters on release on the basis of a contaminated material in a defined scenario. CEN/TS 14429 is a typical parametric test.
- Temperature, pH-value, liquid/solid ratio, redox potential, chemical properties or leaching agent flow rate are examples of specific parameters which influence the behaviour towards leaching.
- **Multiparametric tests.** These tests are intended to measure the combined effect of different parameters on release from a contaminated material in a relevant scenario. For a typical multiparametric leaching test see CEN/TS 14405.

**NOTE** A methodology for the determination of the leaching behaviour of waste has been developed within TC 292 and formulated in EN 12920.

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### 6.1.3 Compliance tests

Tests belonging to this category are used to determine whether the waste complies with specific reference values. They also allow intercomparison and classification of different types of wastes. These tests focus on key variables and leaching behaviour identified by basic characterization tests. In contrast to characterization tests, this type of tests is relatively simple and quick. It is not designed to provide information on leaching mechanisms and controlling factors. However, it is important to link the information obtained with such tests to the more elaborated characterization tests.

Batch leaching tests developed originally by CEN/TC 292: EN 12457-1 to EN 12457-4 belong to this category and are based on different liquid to solid (L/S) ratios (2 - 10) and different particle sizes (4 mm - 10 mm).

### 6.1.4 "On-site verification"

Tests belonging to this category are used as a rapid check to confirm that the waste is the same as that which has been subjected to the compliance test(s).

## 6.2 Selection of leaching tests

In addition to those mentioned above, it is important to consider some other parameters when selecting leaching procedures for ecotoxicological testing such as duration of leaching test, amount of eluate needed to perform ecotoxicological tests and separation of solid and liquid phases.

For example, for the leaching test procedure described in CEN/TS 14405, a linear leachant velocity of 15 cm/day has been fixed. This enables the test to be carried out to a final L/S = 10 l/kg in approximately 30 days and to reach L/S = 2 l/kg within approximately one week. This however leads to extended storage period of eluate (for batch leaching tests, the contact time does not exceed 24 h) and possibly to significant changes in its toxicity before performing ecotoxicity tests.

The advantages and limitations of the available techniques are given in Table 1, Table 2 and Table 3.

It is important to use the same leaching protocol for chemical analyses and for ecotoxicity tests in order to facilitate the interpretation of the results.

For more information on the L/S ratio for batch tests and other leaching protocols refer also to ISO 18772.

**Table 1 — Advantages and limitations of leaching procedures for ecotoxicity testing**

	Parameter	Advantages	Limitations
Type of test	Static test (as EN 12457-1 to -4)	<ul style="list-style-type: none"> <li>- Covering the water available fraction, simulation of leaching depending on L/S ratio.</li> <li>- Quick, most experience, reproducible.</li> </ul>	<ul style="list-style-type: none"> <li>- Separation could result in a loss of compounds</li> <li>- Substantial higher release of organic contaminants observed in batch L/S=10 compared to cumulative release in percolation test.</li> </ul>
	Dynamic test (as CEN/TS 14405)	<ul style="list-style-type: none"> <li>- Simulation of leaching, covering leaching in the field, risk assessment in terms of contact time.</li> <li>- Separation of solid materials is not needed (no turbidity).</li> <li>- Contact time can be adapted to scenario conditions.</li> <li>- More realistic test conditions possible.</li> </ul>	<ul style="list-style-type: none"> <li>- The time required to obtain a sufficient volume of eluate (depending on column diameter and L / S ratio) can be long. It could lead to changing characteristics of the eluate during the storage period prior to ecotoxicity tests.</li> </ul>