

SLOVENSKI STANDARD oSIST prEN 15457:2020

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Barve in laki - Laboratorijska metoda za preskušanje učinkovitosti konzervansov filma v premazih proti glivam

Paints and varnishes - Laboratory method for testing the efficacy of film preservatives in a coating against fungi

Beschichtungsstoffe - Laborverfahren für die Prüfung der Wirksamkeit von Filmkonservierungsmitteln in einer Beschichtung gegen Pilze

Peintures et vernis - Méthode d'essai en laboratoire permettant de déterminer l'efficacité des préservateurs du feuil d'un revêtement contre les champignons

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ICS:

87.040 Barve in laki Paints and varnishes

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English Version

Paints and varnishes - Laboratory method for testing the efficacy of film preservatives in a coating against fungi

Peintures et vernis - Méthode d'essai en laboratoire permettant de déterminer l'efficacité des préservateurs du feuil d'un revêtement contre les champignons Beschichtungsstoffe - Laborverfahren für die Prüfung der Wirksamkeit von Filmkonservierungsmitteln in einer Beschichtung gegen Pilze

This draft European Standard is submitted to CEN members for enquiry. It has been drawn up by the Technical Committee CEN/TC 139.

If this draft becomes a European Standard, 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.

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Recipients of this draft are invited to submit, with their comments, notification of any relevant patent rights of which they are aware and to provide supporting documentation.

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EUROPEAN COMMITTEE FOR STANDARDIZATION COMITÉ EUROPÉEN DE NORMALISATION EUROPÄISCHES KOMITEE FÜR NORMUNG

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European foreword

This document (prEN 15457:2020) has been prepared by Technical Committee CEN/TC 139 "Paints and varnishes", the secretariat of which is held by DIN.

This document is currently submitted to the CEN Enquiry.

This document will supersede EN 15457:2014.

The main changes compared to EN 15457:2014 are as follows:

- a) new terms and definitions added;
- b) use of terms and definitions throughout the document corrected;
- c) procedure in 8.4 corrected: the suspension is spread out onto the test specimen;
- d) Figure A.1 corrected in accordance with corrected procedure in 8.4;
- e) document editorially revised.

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Introduction

This document identifies criteria to assess efficacy of film preservatives in a coating against fungi. The results of the method allow evaluation of an active substance with regard to its inclusion in Annex I of the Biocidal Product Regulation 528/2012 (Regulation EU No 528/2012 of the European Parliament and of the Council of 22 May 2012 concerning the placing of biocidal products on the market – BPR).

The characteristics of the biocide treated coating material should conform to national regulations with regard to health, safety and the environment.

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1 Scope

This document specifies a laboratory test method for determining the biocidal/biostatic efficacy of single active substances or combinations thereof used in film preservatives in a coating against fungal growth. This document does not apply to coatings not susceptible to fungal growth. The test method comprises only active substances for film preservation, not the protection of the substrate itself, e.g. wood, which is dealt with in another standard. The test method is applicable for active substances used for wood and masonry coatings. It is not applicable to marine coatings.

Safety, health and environmental aspects are not in the scope of this document.

Determination of the performance of film preservatives in coatings by applying ageing procedures is not within the scope of this document.

2 Normative references

The following documents are referred to in the text in such a way that some or all of their content constitutes requirements 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.

EN 12469, Biotechnology — Performance criteria for microbiological safety cabinets

EN 16492:2014, Paints and varnishes — Evaluation of the surface disfigurement caused by fungi and algae on coatings

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EN 23270, Paints and varnishes and their raw materials—Temperatures and humidities for conditioning and testing (ISO 3270)

EN ISO 1513, Paints and varnishes — Examination and preparation of test samples (ISO 1513) https://standards.iteh.ai/catalog/standards/sist/23e80eec-9c66-46f4-bbce-

3 Terms and definitions 7032b8b1021d/osist-pren-15457-2020

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

ISO and IEC maintain terminological databases for use in standardization at the following addresses:

- IEC Electropedia: available at http://www.electropedia.org/
- ISO Online browsing platform: available at http://www.iso.org/obp

3.1

active substance

substance or micro-organism that has an action on or against harmful organisms

[SOURCE: Biocidal Product Regulation (BPR, Regulation (EU) 528/2012), Article 3.1 (c), modified – the article "a" between "or" and "micro-organism" was deleted]

3.2

sample

portion of coating material to be tested

3.3

test sample

strip of filter paper without biocidal effect coated with the coating material to be tested

Note 1 to entry: See also Figure A.1.

3.4

test specimen

punched-out portion of a test sample

Note 1 to entry: See also Figure A.1.

4 Principle

To determine the fungicidal efficacy of film preservatives in a coating, the coating material is applied to a substrate conditioned according to EN 23270, placed onto an agar surface, inoculated with a standard fungal spore suspension and incubated. Conclusions can be drawn to the fungicidal efficacy of the film preservatives in a coating from the intensity of the fungal growth on the surface of the test specimen after incubation. The method described here is a semiquantitative, comparative method between coatings, with and without film preservatives.

5 Apparatus and materials

- **5.1 Cutting device** for preparing the test specimens (coated filter paper with a diameter of 55 mm).
- 5.2 Autoclave
- **5.3 Incubator** capable of maintaining (24 ± 2) °C.
- **5.4 Pipette**, in the range between $100 \mu l$ to $1000 \mu l$, with sterile tips or combitips of 12,5 ml.
- 5.5 Filter paper without fungicidal effect (e.g. cellulose with a pore size of 0,45 μ m and a thickness of 650 μ m).

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- **5.6 Automatic welding apparatus** to is eal the bags dards/sist/23e80eec-9c66-46f4-bbce-7032b8b1021d/osist-pren-15457-2020
- 5.7 Sterilized glass bottles (100 ml).
- **5.8 Laboratory balance**, capable of weighing to an accuracy of 0,1 g.
- 5.9 Microscope
- **5.10 Device to determine cell count** (commercially available counting chamber, e.g. Thoma chamber).
- **5.11 Wetting agent** (e.g. N-Methyltaurine).
- 5.12 Device for applying the coating
- **5.13 Sterilized test tubes** or **other sterilized glassware** for preparing slant agar cultures.
- 5.14 Sterile Drigalski spatula
- 5.15 Sterile platinum loop
- 5.16 Sterile glass funnel with cotton wool
- **5.17 Sterile Petri dishes** (with a diameter of 94 mm, and a height of 16 mm).
- 5.18 Sterile tweezers

5.19 Sterile water

5.20 Class 2 microbiological safety cabinet according to EN 12469.

6 Fungi

6.1 Fungi more likely to grow in an exterior environment

- a) Aureobasidium pullulans (DSM¹⁾ 2404)
- b) Alternaria alternata (DSM 62010)
- c) Cladosporium cladosporioides (DSM 62121)
- d) Phoma violaceae (IMI²⁾ 49948ii)
- e) Ulocladium atrum (IMI 79906 or DSMZ 63068)

6.2 Fungi more likely to grow in an interior environment

- a) Aspergillus versicolor (DSM 1943)
- b) Aspergillus niger (DSM 12634)

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c) Stachybotrys chartarum (DSMZ 2144)

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- d) Penicillium purpurogenum (DSM 62866)
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e) Rhodotorula mucilaginosa (DSM₁79825)_{tandards/sist/23e80eec-9c66-46f4-bbce-7032b8b1021d/osist-pren-15457-2020}

The spore suspension used for the test shall be a mixture containing two fungi selected from the first group (6.1) and two fungi selected from the second group (6.2).

7 Sampling and preparation of test samples and of test specimens

7.1 Sampling

Take a representative sample of the coating material or of the coating system for testing in accordance with EN ISO 1513.

7.2 Preparation of test samples (see Annex A)

Coat a strip of filter paper without biocidal effect with the coating material to be tested. The application rate shall be that recommended by the coating manufacturer for normal use.

7.3 Conditioning of the test samples

Condition the test sample in a horizontal position for at least 5 days at (23 ± 2) °C and (50 ± 5) % relative humidity, in accordance with EN 23270.

¹⁾ DSM = DSMZ = Deutsche Sammlung für Mikroorganismen und Zellkulturen (German collection of microorganisms and cell cultures), Braunschweig, Germany.

²⁾ IMI = CABI = Bioscience Genetic Resource Collection, Egham, UK.

NOTE The conditioning time might vary according to the coating material and end use corresponding to information given by the manufacturer.

7.4 Preparation and number of test specimens

After conditioning, three test specimens each with a diameter of 55 mm shall be prepared from the test samples. The test specimens shall be sealed in a plastics bag and sterilized using gamma radiation of ≥ 10 kGy. Other methods of sterilization may be agreed between the parties.

For each test series three test specimens coated with coating material containing the film preservative, three test specimens coated with the same coating material without film preservative and three test specimens of the uncoated substrate shall be tested.

8 Procedure

8.1 Preparation of the Petri dishes with the culture medium

A malt (3 %)-agar(1,5 %)-medium shall be sterilized in the autoclave. After cooling the medium to 55 $^{\circ}$ C to 60 $^{\circ}$ C, 20 ml shall be poured into each sterile Petri dish under aseptic conditions.

8.2 Preparation of stock cultures and sub-cultures

Sub-cultures shall be obtained by inoculating spore material from a stock culture to freshly prepared agar slope culture media and shall be used for preparing the spore suspension. From these sub-cultures further sub-cultures can be derived in sufficient number. After the inoculation the sub-cultures shall be incubated at (24 ± 2) °C until good sporulation has been achieved. This might require 3 days to 7 days, depending on the fungal species used for testing. The sub-cultures can be stored satisfactorily at 3 °C to 7 °C for a period of 3 months.

8.3 Preparation of the spore suspension SIST prEN 15457:2020

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For preparing the spore suspension a well-sporulating sub-culture is used. To this sub-culture 5 ml of sterile deionized water should be added (if required also add a surfactant – e.g. 0,1 % N-Methyltaurine). The spores shall be carefully washed down from the agar slope, using a platinum loop as an aid, filtered through a sterile glass funnel with cotton wool and collected in a sterile glass bottle.

After counting the spore concentration by using a commercially available counting chamber and diluting each with sterile water to 10^6 to 10^7 spores/ml all spore suspensions intended for testing shall be mixed in equal parts.

8.4 Inoculation and incubation (see Annex A)

In addition to the coated and uncoated test specimens (see 7.4) a further three Petri dishes containing the nutritive agar medium only shall be inoculated. The sterilized test specimens shall be placed centrally onto the surface of the culture media using sterile tweezers. The coated surface of the specimen shall be face up and there shall be full contact without air bubbles between the specimens and the surface of the culture medium.

Should the coating lead to an undulation of the filter paper the paper should be kept even and in close contact with the agar by appropriate means. Otherwise a different substrate may be used. Check that the substrate does not inhibit growth of each selected test organism under the test conditions. If the substrate does inhibit growth it cannot be used.

Under aseptic conditions in a safety cabinet the specimens shall be uniformly inoculated with 0,2 ml each of the mixed spore suspension using a suitable pipette. It is permissible to use up to an additional 0,8 ml of diluent to ensure an even distribution over the surface. Afterwards the suspension is spread