



SLOVENSKI STANDARD
SIST EN 15457:2007

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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 vérifier l'efficacité des préservateurs du feuill d'un revêtement contre les champignons

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ICS:

87.040

Barve in laki

Paints and varnishes

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ICS 87.040

English Version

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

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

This European Standard was approved by CEN on 21 June 2007.

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This European Standard exists in three official versions (English, French, German). A version in any other language made by translation under the responsibility of a CEN member into its own language and notified to the CEN Management Centre has the same status as the official versions.

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

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Foreword

This document (EN 15457:2007) has been prepared by Technical Committee CEN/TC 139 "Paints and varnishes", the secretariat of which is held by DIN.

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of an identical text or by endorsement, at the latest by January 2008, and conflicting national standards shall be withdrawn at the latest by January 2008.

According to the CEN/CENELEC Internal Regulations, the national standards organizations of the following countries are bound to implement this European Standard: Austria, Belgium, Bulgaria, Cyprus, Czech Republic, Denmark, Estonia, Finland, France, Germany, Greece, Hungary, Iceland, Ireland, Italy, Latvia, Lithuania, Luxembourg, Malta, Netherlands, Norway, Poland, Portugal, Romania, Slovakia, Slovenia, Spain, Sweden, Switzerland and United Kingdom.

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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 ingredient with regard to its inclusion in Annex A of the Biocidal Products Directive 98/8/EC (Directive 98/8/EC of the European Parliament and of the Council of 16 February 1998 concerning the placing of biocidal products on the market – BPD).

NOTE 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 European standard specifies a laboratory test method for determining the biocidal/biostatic efficacy of film preservatives in a coating against fungal growth. This standard does not apply to coatings not susceptible to fungal growth. The test method comprises only 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 wood and masonry coatings. It is not applicable to marine coatings.

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

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.

EN 12469, *Biotechnology — Performance criteria for microbiological safety cabinets*

EN 23270, *Paints and varnishes and their raw materials — Temperatures and humidities for conditioning and testing (ISO 3270:1984)*

EN ISO 1513, *Paints and varnishes — Examination and preparation of samples for testing (ISO 1513:1992)*

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3 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 specimen after incubation. The method described here is a semi-quantitative, comparative method between coatings, with and without film preservatives.

4 Apparatus and materials

- 4.1 **Cutting device** for preparing the specimens (coated filter paper with a diameter of 55 mm).
- 4.2 **Autoclave**
- 4.3 **Incubator** capable of maintaining (24 ± 2) °C.
- 4.4 **Pipette**, in the range between 100 µl to 1 000 µl, with sterile tips or combi-tips of 12,5 ml.
- 4.5 **Filter paper** without fungicidal effect (e.g. cellulose with a pore size of 0,45 µm and a thickness of 650 µm).
- 4.6 **Automatic welding apparatus** to seal the bags.
- 4.7 **Sterilised glass bottles** (100 ml).
- 4.8 **Laboratory balance**, capable of weighing to an accuracy of 0,1 g.
- 4.9 **Microscope**

- 4.10 **Device to determine cell count** (commercially available counting chamber, e.g. Thoma chamber).
- 4.11 **Wetting agent** (e.g. N-Methyltaurine).
- 4.12 **Device for applying the coating**
- 4.13 **Sterilized test tubes or other sterilized glassware** for preparing slant agar cultures.
- 4.14 **Sterile Drigalski spatula**
- 4.15 **Sterile platinum loop**
- 4.16 **Sterile glass funnel with cotton wool**
- 4.17 **Sterile Petri dishes** with a diameter of 94 mm, and a height of 16 mm).
- 4.18 **Sterile tweezers**
- 4.19 **Sterile water**
- 4.20 **Class 2 microbiological safety cabinet** according to EN 12469.

5 Fungi

5.1 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)

5.2 Fungi more likely to grow in an interior environment

- a) *Aspergillus versicolor* (DSM 1943)
- b) *Aspergillus niger* (DSM 12634)
- c) *Stachybotris chartarum* (DSMZ 2144)
- d) *Penicillium purpurogenum* (DSM 62866)
- e) *Rhodotorula mucilaginosa* (DSM 70825)

The spore suspension used for the test shall be a mixture containing two fungi selected from the first group (5.1) and two fungi selected from the second group (5.2).

1) DSM = DSMZ = Deutsche Sammlung für Mikroorganismen und Zellkulturen (German collection of micro organisms and cell cultures), Braunschweig, Germany
2) IMI = CABI = Bioscience Genetic Resource Collection, Egham, UK

6 Sampling and preparation of test samples and of specimens

6.1 Sampling

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

6.2 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.

6.3 Conditioning of the test samples

Condition the test sample in a horizontal position for at least 5 days at $(23 \pm 2) ^\circ\text{C}$ and $(50 \pm 5) \%$ relative humidity, in accordance with EN 23270.

NOTE The conditioning time may be different according to the coating material and end use corresponding to information given by the manufacturer.

6.4 Preparation and number of specimens

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

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

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7 Procedure

7.1 Preparation of the Petri dishes with the culture medium

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

7.2 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 $(24 \pm 2) ^\circ\text{C}$ 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 $3 ^\circ\text{C}$ to $7 ^\circ\text{C}$ for a period of 3 months.

7.3 Preparation of the spore suspension

For preparing the spore suspension a well-sporulating sub-culture is used. To this sub-culture 5 ml of sterile distilled water shall 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.