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Laboratory test method - assessment of the susceptibility of thermal insulation products to mould growth

Bewertung des Widerstands von Wärmedämmprodukten gegen Schimmelbildung

Évaluation de la résistance des produits d'isolation contre le développement des moisissures

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Laboratory test method - assessment of the susceptibility of thermal insulation products to mould growth

Évaluation de la résistance des produits d'isolation
contre le développement des moisissures

Bewertung des Widerstands von
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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 17886:2022) has been prepared by Technical Committee CEN/TC 88 “Thermal insulating materials and products”, the secretariat of which is held by DIN.

This document is currently submitted to the CEN Enquiry.

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Introduction

Insulation materials used in buildings can be subjected to high humidity either periodically or permanently and thus be affected by mould growth.

The main factor that affect the growth of mould is an increased availability of moisture on the surface of materials. In buildings, this results typically from condensation of water vapour and/or an unintended increase of material moisture content through building defects, etc.

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1 Scope

This document describes a laboratory test method to determine the susceptibility of thermal insulation products used for construction against mould growth under specified climatic conditions.

The method is applicable to both factory-made products and *in situ* formed products. Factory-made products include panels, mats, and rolls. *In situ* formed products are usually those that are delivered loose and installed by blowing-in, poring, or spraying-on, using water and/or binder, whether or not they are also treated using additives.

Depending on the insulation manufacturer, the test is carried out with one of the conditions in Table 1. This condition is linked to worst case scenarios selected from real hygrothermal conditions that reflect the end use conditions experienced by insulation products, and to short test duration for mould test.

This test method determines the susceptibility of a thermal insulation material to mould growth, but does not determine the suitability for use in a given design (wall, roof, etc.).

This method does not predict the resistance of an insulation product to accidental water exposure resulting in saturation of the product (water damage).

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 ISO 846:2019, *Plastics - Evaluation of the action of microorganisms (ISO 846:2019)*

EN ISO 3696, *Water for analytical laboratory use - Specification and test methods (ISO 3696)*

EN ISO 7218, *Microbiology of food and animal feeding stuffs - General requirements and guidance for microbiological examinations (ISO 7218)*

3 Terms and definitions

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 <https://www.electropedia.org/>
- ISO Online browsing platform: available at <https://www.iso.org/obp>

3.1

colony-forming unit

CFU

unit used in microbiology which estimates the number of bacteria or fungal cells in a sample which are viable; cell or aggregate of cells which gives rise to a single colony (a population of the cells visible to the naked eye) in a semi-solid growth medium

3.2

test specimen

sample made specially for the test from the insulation product

prEN 17886:2022 (E)**3.3****mould growth**

presence of either mycelium, germinated spores or fruiting bodies

4 Principle

The test specimens are prepared from insulating material test samples and are sterilized, then inoculated with a defined quantity of fungal spores. The samples are then incubated under specific conditions of temperature and relative humidity. At the end of the test, the fungal growth is assessed by visual observation and, where appropriate, by determining the number of CFU present. The presence or absence of fungal growth makes it possible to rule on the intrinsic characteristic of susceptibility of the tested product.

5 Test materials and apparatus**5.1 Fungal species**

The fungal species referred to in normative documents shall be used to inoculate the insulating product to be tested. The fungal species shall have been obtained from official culture collections (reference stocks).

Because of the handling of biological agents, it is recommended to use personal protective equipment and/or proceed with caution using appropriate equipment such as a microbiological safety cabinet.

Compulsory fungal strains:

- *Aspergillus niger* (MNHN 48-521, or DSM 1957)
- *Trichoderma viride* (MNHN 88-3354 or IHEM 4146)
- *Penicillium funiculosum* (MNHN 56-1527 or DSM 1944)
- *Chaetomium globosum* (ATCC 6205)
- *Paecilomyces variotii* (MNHN LCP 79-3210 or DSM 1961)
- *Aspergillus versicolor* (ATCC 11730)

This test may be done with additional fungal strains. In this case it shall be done with an additional separate set of test specimens.

5.2 Products and reagents

The following products and reagents shall be used:

- Culture medium:
 - Malt powder (2 % w/w) and agar (2 % w/w) to be prepared in water quality 3.
- Solvents and thinners:
 - Water quality 3 in accordance with EN ISO 3696.
 - Wetting agent such as Tween 80.
- Binder-free glass fibre pre-filters that do not contain any cellulose, 70 mm in diameter.

5.3 Apparatus

The following apparatus shall be used:

- Climate chamber with controlled temperature and relative humidity maintained (mean values recorded) at either:
 - 28 °C ± 2 °C and 95 % ± 4 %; or
 - 28 °C ± 2 °C and 85 % ± 4 %; or
 - 20 °C ± 2 °C and 85 % ± 4 %; or
 - 14 °C ± 2 °C and 95 % ± 4 %.
- Conditioning chamber or drying oven at 70 °C ± 3 °C.
- Sterile Petri dishes.
- Atomiser capable of providing $1,5 \times 10^4$ spores / cm³.
- Stomacher (optional) and sterile bags.
- Nylon-based filter with a pore size of 20 µm to allow the preparation of the spore suspension.
- Autoclave.
- Microscope or binocular magnifier at both 20 × and 50 × magnification.
- Material specifically used for the preparation of the spore suspension to be sprayed:
 - Glass beads 3 to 5 mm in diameter;
 - Centrifuge or filtration unit;
 - Counting chamber. The Malassez cell or a chamber with a Thoma ruling (0,1 mm depth) is appropriate for counting fungal spores.
- Microbiological safety cabinet.

6 Sampling of the insulating products

For loose-fill insulating products, three manufacturing batches of products are required. The samples shall be taken from those three batches. For insulating products made of boards or rolls, three boards/rolls coming from three different manufacturing batches are required.

7 Insulating product specimens

7.1 Number of test specimens

Three series of specimens from the insulating product to be tested shall be prepared:

- **Series 1 (S1): 3 specimens** dedicated to the assessment of the quantity of viable spores settled at the beginning of the test (Time T0), the day of inoculation (one test specimen from each batch).
- **Series 2 (S2): 9 specimens** (three test specimens from each batch) dedicated to the visual inspection and, where appropriate, to the assessment of the quantity of CFU – at the end of the test – after 4 weeks (Time T4 weeks) or 8 weeks (Time T8 weeks) incubation (according to the test condition in Table 1).
- **Series 3 (S3): 3 specimens** (one test specimen from each batch) dedicated to the determination of the water absorption by the test specimens at the end of the test – after 4 weeks or 8 weeks incubation (according to the test condition in Table 1).

7.2 Preparation of the test specimens

For insulation products made of rolls or boards, the specimens shall be cut preferably from the surface away from the edge. The area of each test specimen shall be at least 20 cm² and its thickness shall range between 8 and 10 mm. During preparation, the material shall not be damaged, to ensure that its structure and density are maintained.

For loose-fill insulating products, enough material shall be used to be gently tamped down to obtain the smoothest surface possible. This will facilitate microscopic examination. The area of each test specimen shall be at least 20 cm² and its thickness shall range between 8 and 10 mm.

For final products such as the External Thermal Insulation Composite System (ETICS), a thickness exceeding 10 mm may be tested. The size and the volume of all the test specimens shall be the same.

This method may be applied to wet-sprayed insulation products and insulation products sprayed with a binder using conditions that are relevant to the water or binder content representative of their installation.

If insulation materials are not homogeneous and have surface layers, both “materials” shall be tested (surface layer + matrix).

7.3 Observation of the test specimens when delivered

The insulation test specimens shall be inspected with the naked eye and through the microscope ($\times 50$ magnification minimum) so as to make sure that no fungi have grown after delivery and before the start of the test.

At this stage, any material likely to have been the subject of fungal growth shall not be tested.

8 Control specimens

Control specimens are prepared using three binder-free glass fibre pre-filters (5.2).

The glass fibre pre-filters are sterilized by gamma radiation (9.1).

The following solution is prepared and sterilized for 30 min \pm 2 min at 115 °C \pm 1 °C in the autoclave.

The solution's composition is detailed in EN ISO 846:2019, 5.2.3.1.

NaNO ₃	2,0 g
KH ₂ PO ₄	0,7 g
K ₂ HPO ₄	0,3 g
KCl	0,5 g
MgSO ₄ · 7H ₂ O	0,5 g
FeSO ₄ · 7H ₂ O	0,01 g
Glucose	30 g
H ₂ O	1 000 ml

After sterilization, a specified amount of solution is put on each control specimen (9.2.3).

Once the solution is put on the control specimens, they are dried on a rack and then used in the test in sterile Petri dishes.

9 Procedure

9.1 Sterilization by ionizing radiation

The insulation product specimens to be tested S1, S2 and S3 and the control specimens (glass fibre pre-filters- before the addition of the solution described in Clause 8) shall be sterilized. The specimens are placed in individual bags made from polyethylene. The bags are then closed by heat-welding, and then sterilized by ionizing radiation (25kGy to 50kGy from a 60 Co source or 50 kGy to 100 kGy using a particle accelerator).

9.2 Inoculation

9.2.1 Preparation of the spore suspension

Follow each stage described below carefully for each fungal strain, and separately, as follows.

The strains described in 5.1 are first grown on malt (2 % w/w) agar (2 % w/w) for 1 to 4 weeks of culture at 22°C to 28 °C ± 2 °C.

Sterile water quality 3 (10 mL in addition to 0,05 % (w/v) wetting agent and NaCl at 0,9 % (w/v)) is to be added to each Petri dish containing a fungal culture and agitated lightly using a sterile inoculating loop. The resulting suspension of spores and hyphal fragments is then transferred to a sterile Erlenmeyer flask. The flask shall contain previously sterilized glass beads (3 to 5 mm in diameter). The spore solution shall be agitated vigorously and then filtered using a nylon-based filter (porosity of 20 µm) or glass wool. The spore solution shall then be centrifuged at 2000 g for 20 min. The supernatant is then removed and the residue re-suspended in 20 mL of sterile water containing 0,9 % (w/v) NaCl and 0,05 % (w/v) wetting agent. This centrifugation process is then repeated. After the second centrifugation, 10 mL of sterile water containing 0,9 % NaCl (w/v) and 0,05 % wetting agent (w/v) is to be added to the tube and the contents agitated vigorously to produce a uniformly dispersed suspension of spores.

The six fungal spore solutions are diluted and combined so as to obtain an inoculum containing a total of 1×10^6 spores / mL. Thus, this inoculum solution contains $1,67 \times 10^5$ spores/mL of each one of the six strains.

The number of spores in each of the final suspensions is determined using a counting chamber (Malassez or Thoma) for each fungal species.