



Mildew, Mixed Fungal Resistance – Mixed Culture Method

1 Introduction

Note: Nothing in this standard supercedes applicable laws and regulations.

Note: In the event of conflict between the English and domestic language, the English language shall take precedence.

This standard describes a procedure used to evaluate antifungal effectiveness of plastics, coated fabrics and other polymeric materials.

1.1 Purpose. To detect Polyvinyl Chloride (PVC) exterior grades compatible for exterior use

1.2 Applicability. Integral colored exterior grade PVC trim.

2 References

Note: Only the latest approved standards are applicable unless otherwise specified.

2.1 External Standards/Specifications.

None

2.2 GM Standards/Specifications.

None

3 Resources

3.1 Facilities.

3.1.1 Calibration. The test facilities and equipment shall be in good working order and shall have a valid calibration label.

3.1.2 Alternatives. Alternative test facilities and equipment may also be used. However, all measuring variables as specified in this standard shall be determined correctly with respect to their physical definition.

3.2 Equipment.

3.2.1 Petri dishes, 100 mm.

3.2.2 Sterile cotton tipped swabs.

3.2.3 Sterile distilled water.

3.2.4 Incubator capable of maintaining $29\text{ }^{\circ}\text{C} \pm 1\text{ }^{\circ}\text{C}$.

3.2.5 Autoclave capable of maintaining a pressure of 100 kPa at a temperature of $121\text{ }^{\circ}\text{C} \pm 2\text{ }^{\circ}\text{C}$.

3.2.6 Atomizer: DeVilbiss Number 15 or equivalent.

3.2.7 Culture Media. The test medium composition shall be as follows. Do not adjust the pH:

Ammonium nitrate (NH_4NO_3)	3.0 g
Potassium dihydrogen orthophosphate (KH_2PO_4)	2.0 g
Magnesium sulfate ($\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$)	0.5 g
Potassium chloride (KCl)	0.5 g
Dextrose	1.0 g
Agar	15.0 g
Distilled water	1000 mL

3.2.7.1 Heat to boiling to dissolve mixture completely before adding dextrose. Sterilize test medium by autoclaving at 100 kPa and a temperature of $121\text{ }^{\circ}\text{C} \pm 2\text{ }^{\circ}\text{C}$ for 15 minutes. Pour sufficient test medium into petri dishes to give a layer 15 mm in depth and allow to solidify at room temperature.

3.2.8 Test Organism. See Table 1.

Table 1: Test Organism

Culture Collection Number	Type Culture	Quartermaster
Aspergillus niger	9642	386
Aspergillus flavus	9643	380
Penicillium funiculisum	9644	391
Chaetomium globosum	6265	459

3.2.8.1 All stock cultures, except for Chaetomium globosum, should be maintained separately on appropriate medium such as potato dextrose agar, or on the test medium with 0.05% yeast extract added. Chaetomium globosum should be maintained on a sterile strip of craft paper placed on the test medium without dextrose. Stock cultures are stored in refrigerators at $3\text{ }^{\circ}\text{C}$ to $7\text{ }^{\circ}\text{C}$ and transfers made at six month intervals. Subcultures incubated at $28\text{ }^{\circ}\text{C} \pm 2\text{ }^{\circ}\text{C}$ for 7 to 14 days should be used in preparing the spore suspension.

3.2.8.2 Mixed Fungal Spore Suspension.

3.2.8.2.1 A spore suspension is prepared by pouring into one subculture of each organism, a 10 mL of sterile distilled water containing 0.05 mL/L of non-toxic wetting agent such as sodium dioctyl sulfosuccinate. Use a sterile platinum or nichrome inoculating loop to gently scrape the surface growth from the agar medium.

3.2.8.2.2 Pour the spore mixtures into a sterile jar containing ten to fifteen 5 mm diameter solid glass beads, and shake vigorously to break up the spore lumps.

3.2.8.2.3 Filter the shaken suspension through a thin layer of sterile glass wool in a sterile funnel into a sterile bottle. Add distilled water to make up a volume of 100 mL.

3.2.8.2.4 The spore suspension may be prepared fresh each day or kept in a refrigerator at $3\text{ }^{\circ}\text{C}$ to $10\text{ }^{\circ}\text{C}$ for not more than 7 days.

3.3 Test Vehicle/Test Piece. Not applicable.

3.4 Test Time.

Calendar time: 14 to 16 days

Test hours: 0 hours

Coordination hours: 0 hours

3.5 Test Required Information. Not applicable.

3.6 Personnel/Skills. Not applicable.

4 Procedure

4.1 Preparation. Not applicable.

4.2 Conditions.

4.2.1 Environmental Conditions. Not applicable.

4.2.2 Test Conditions. Deviations from the requirements of this standard shall have been agreed upon. Such requirements shall be specified on component drawings, test certificates, reports, etc.

4.3 Instructions.

4.3.1 Test Specimen. Cut a 25 mm diameter disc from each sample and place it on the test medium. For coated fabrics, two discs cut from each sample are tested with the plastic side and fabric side each in firm contact with the agar. It is important to establish good contact between the sample surface and the agar surface. Sterile