

**Aerospace series - Paints and varnishes - Determination  
of resistance to microbial growth**

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ICS 49.040

English Version

## Aerospace series - Paints and varnishes - Determination of resistance to microbial growth

Série aérospatiale - Peintures et vernis - Détermination de la résistance à l'action des microorganismes

Luft- und Raumfahrt - Beschichtungsstoffe - Bestimmung der Widerstandsfähigkeit gegen Schimmelwachstum

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## Foreword

This document (EN 4159:2011) has been prepared by the Aerospace and Defence Industries Association of Europe - Standardization (ASD-STAN).

After enquiries and votes carried out in accordance with the rules of this Association, this Standard has received the approval of the National Associations and the Official Services of the member countries of ASD, prior to its presentation to CEN.

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of an identical text or by endorsement, at the latest by June 2012, and conflicting national standards shall be withdrawn at the latest by June 2012.

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## Introduction

Certain fungi are known to be capable of proliferating in fuel systems which can cause corrosion and blockage. Conidiospores are the dispersal form of these fungi. Germination of conidia is the first stage in proliferation of the fungus. If the conidiospore cannot germinate, there can be no proliferation and no blockage of fuel lines, ducts etc.

This method should be performed only by persons qualified in the microbiology of fungi.

The standard can be used to assess the effectiveness of new candidate coating systems in inhibiting microbial (fungal) growth.

## 1 Scope

This European Standard specifies a method to assess the ability of biocide-containing coatings to prevent the germination of conidiospores of certain fungi known to be capable of proliferating in fuel systems for aerospace applications.

## 2 Terms and definitions

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

### 2.1

#### **Conidiospores**

single-celled structures produced by the mycelial [mould] form of the fungus

**NOTE** Conidiospores are spherical or nearly spherical resting cells, i.e. cells which may be dispersed readily but which do not proliferate. However, conidiospores may germinate if they encounter suitable conditions of moisture and nutrients. On germination, a conidiospore produces a long tube-like outgrowth which then forms dense branching structures [mycelia] which may block fuel ducts etc. A suitable coating will prevent germination of conidiospores. A coating which prevents germination of conidiospores is considered to have fungistatic activity. This fungistatic activity may be assessed quantitatively by assessing the success rate of germination of conidiospores under standard conditions (see below) to determine whether the test coating delays or prevents germination of conidiospores when compared with a coating which is known to possess no fungistatic activity. Laboratories which undertake work to this method should first obtain the test fungi (see 5) and perform control experiments to satisfy themselves that they can follow the process of germination of conidiospores. These initial experiments may be performed by placing the agarose gel (see below) on the surface of sterile plastic petri dishes rather than on the surface of coated test panels, as is done in the present method.

## 3 Principle

**3.1** Conidiospores are placed on a gel within a few millimetres of the panel/coating under test. Under the test conditions a high proportion of these conidia germinate [begin growth] rapidly unless some material in the coating diffuses through the gel and prevents germination.

**3.2** The success rate of germination, after any given interval of exposure to the coating, is expressed as the number of cells that have germinated divided by the number of cells examined (germinated + nongerminated). The success rate of germination is determined from time to time, beginning when the conidiospores are first exposed to the coating under test. Examination is made using a microscope [100×]. This allows ready distinction between ungerminated conidiospores [approximately spherical] and the long filamentous outgrowth that is the result of germination.

**3.3** The results obtained with conidiospores exposed to test coatings are to be compared with results of conidiospores exposed to coatings that contain no inhibitor.

## 4 Apparatus

**4.1** Incubator, capable of maintaining  $(25 \pm 1) ^\circ\text{C}$ .

**4.2** Autoclave suitable for sterilization of microbiological growth media, i.e. capable of heating the media to  $121 ^\circ\text{C}$  for 15 min.

**4.3** Water bath, set to  $(45 \pm 1) ^\circ\text{C}$ .

**4.4** Microscope, magnification 100×, and glass microscope slides.

**4.5** Plastic disposable petri dishes -- 90 mm to 100 mm diameter.

**4.6** Sterile microbiological loops. Commercially available disposable plastic loops (stated to carry 10 µl) are suitable.

**4.7** Haemocytometer (blood cell counting chamber) Neubauer ruling.