## INTERNATIONAL STANDARD

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Fine ceramics (advanced ceramics, advanced technical ceramics) — Test method for antialgal activity of semiconducting photocatalytic materials

Céramiques techniques — Méthode d'essai pour l'activité algicide des matériaux photocatalytiques semi-conducteurs





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#### **Foreword**

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The committee responsible for this document is ISO/TC 206, Fine ceramics.

#### Introduction

Under ultraviolet (UV) light illumination, photocatalysts have diverse functions, such as prevention of fouling, antifogging effects, antibacterial effects, deodorization, and decomposition of air and water contaminants, and their applications have grown in recent years. Since products utilizing these photocatalytic functions are commercialized in large quantities, a method to evaluate and determine photocatalytic effects is required. This International Standard is intended to provide a method for objective evaluation of activity in controlling algae, which are a primary producer of microbial contamination (in the form of environmental biofilms) of outdoor structures, under illumination with ultraviolet light to simulate the outdoor environment and to contribute, via control of algae, to conservation of urban landscapes, prevention of member corrosion, and prevention of fouling of water tank window materials.

# Fine ceramics (advanced ceramics, advanced technical ceramics) — Test method for antialgal activity of semiconducting photocatalytic materials

#### 1 Scope

This International Standard specifies a test method for evaluating anti-algal activities in outdoor structures, specifically flat photocatalytic materials (for example, window panes and water tank glasses, films, guardrails, etc.) under irradiation of ultraviolet light. It does not include powder, granular or porous photocatalytic materials.

#### 2 Normative references

The following documents, in whole or in part, are normatively referenced in this document and are indispensable for its application. For dated references, only the edition cited applies. For undated references, the latest edition of the referenced document (including any amendments) applies.

ISO 27447, Fine ceramics (advanced ceramics, advanced technical ceramics) — Test method for antibacterial activity of semiconducting photocatalytic materials

#### 3 Terms and definitions

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

#### 3.1

#### photocatalyst

substance with many functions based on oxidization and reduction reactions under *photoirradiation* (3.3), including decomposition and removal of contaminants, deodorization, antibacterial actions, and prevention of fouling

#### 3.2

#### photocatalytic materials

materials in which or on which the *photocatalyst* (3.1) is added by coating, impregnation, mixing, etc.

Note 1 to entry: Photocatalytic materials are to be used for building and road construction materials to obtain the above-mentioned functions.

#### 3.3

#### photoirradiation

irradiation of UV light at wavelength 300 nm to 380 nm

#### 3.4

#### antialgal activity

activity in suppressing growth of algae over a material surface

#### 3.5

#### photocatalytic antialgal treatment

support of a *photocatalyst* (3.1) by means of various methods, including coating, impregnation, blending, and others, for utilization of the antialgal activity of a photocatalyst

#### 3.6

#### photocatalytic antialgal activity based on three-point absorbance spectrum method

photocatalytic *antialgal activity* (3.4) derived from the ratio of the absorbance of material treated with *photocatalyst* (3.1) after *photoirradiation* (3.3) of UV light to that of non-treated material after photoirradiation

#### 4 Algae used in the test

The type of algae to be used in the test with photocatalytic-treated antialgal materials shall be *Chlorella vulgaris* (NIES-227). The strain of algae to be used in the test shall be the same strain stored in agencies affiliated with the World Federation for Culture Collections or the Japan Society for Culture Collection.

#### 5 Preparation for the test

#### 5.1 Conditions for handling of algae

The test shall be performed in a laboratory with equipment needed to prevent mixing with other microorganisms.

#### 5.2 Chemicals, materials, and apparatus

Unless otherwise specified, the chemicals, materials, and apparatus to be used in the test shall be as follows. Test tubes, flasks, pipettes, tweezers, etc. shall be carefully cleaned with alkaline or neutral detergent, rinsed thoroughly with water, dried, and sterilized by hot air or high-pressure steam before use.

- **5.2.1 Purified water**, the water used for the preparation of all solutions and culture media and for all determinations shall be distilled or deionized and shall have a conductivity of  $<1 \mu S/cm$ .
- **5.2.2 Ethanol for disinfection**, solution whose ethanol concentration has been adjusted to 77 % to 82 % in volume fraction by means of purified water.
- **5.2.3 Autoclave**, capable of maintaining the temperature at 121 °C  $\pm$  1 °C (equivalent to a pressure of 103 kPa).
- **5.2.4 Dry sterilizer**, capable of maintaining the temperature at 160 °C to 180 °C.
- **5.2.5 Spectrophotometer**, capable of measurement within the wavelength range of 400 nm to 800 nm.
- **5.2.6 Refrigerator**, capable of maintaining the temperature at 4 °C ± 1 °C.
- **5.2.7 Platinum loop**, with an end loop of approximately 4 mm.
- **5.2.8 Glass plate**, made from material not affecting the growth of microorganisms.
- **5.2.9 Cotton plug**, plug of non-degreased cotton, or silicon plug, metal plug, or molten plug.
- **5.2.10 Test tube mixer**, for microorganism test.
- **5.2.11 Contact film**, film of material not affecting the growth of microorganisms, which is free of water-absorbing properties and features satisfactory adherence, and in which transmittance in the range of 340 nm to 380 nm, as measured according to the method specified in ISO 27447, accounts for 85 % or more.

- **5.2.12 Moisture retention glass**, glass with thickness of 1,1 mm or less, in which transmittance of 340 nm to 380 nm, as measured according to the method specified in ISO 27447, accounts for 85 % or more and which has been cut to a size capable of covering the entire surface of a Petri dish.
- **5.2.13 Storage Petri dish**, dish with the inside diameter of about 90 mm.
- **5.2.14 Moisture-conditioning filter paper**, filter paper not affecting the growth of microorganisms, which has been cut to a size appropriate for placement in the container in which the test piece is to be set.
- **5.2.15 Disposable cuvette**, plastic cuvettes (about  $12 \text{ mm} \times 12 \text{ mm} \times 45 \text{ mm}$ ) appropriate for placement in the storage container.
- **5.2.16 Tooth brush**, soft type of brush made of polyamide.
- **5.2.17 UV fluorescent lamp**, 20-W black-light blue type UV fluorescent lamp (BLB lamp) that provides UV-A ranging from 300 nm to 400 nm with a peak emission at 351 nm.
- **5.2.18 Fluorescent lamp**, 20-W starter and straight-type white (symbol: W) lamp.
- **5.2.19 UV irradiator**, system comprised of one UV fluorescent lamp, which can shield light from the surrounding region.
- **5.2.20 UV light radiometer**, radiometer with a detector whose sensitivity peak is at 351 nm and provide limit, e.g.  $\pm 10$  nm, shall be used. The radiometer shall be calibrated for the light source to be used or corrected to ascertain sensitivity within the wavelength range to be absorbed by the photocatalytic test piece with suitable approach.
- **5.2.21 Illuminometer**, general type AA-grade illuminometer.
- **5.2.22 Filter unit**, in which the membrane filter is made from a less-absorbent material, such as hydrophilic polyvinylidene fluoride, hydrophilic polytetrafluoroethylene, etc., and the filter pore size is  $0.22 \, \mu m$  or  $0.45 \, \mu m$ .
- **5.2.23 Centrifuge**, capable of the centrifugal force to 1 500 × g for preparation of test solutions.

#### 5.3 Sterilization and filtration methods

#### 5.3.1 Hot-air sterilization

The apparatus to be sterilized shall be placed in a hot-air sterilizer at 160 °C to 180 °C and held at this temperature for 30 min to 60 min. If the sterilized sample exhibits any wet cotton plug or package water after completion of hot-air sterilization, the apparatus shall not be used.

#### 5.3.2 Hot-steam sterilization

An autoclave shall be filled with water and the apparatus to be sterilized shall be placed in a wire cage and placed on the autoclave shelf. The autoclave shall be closed with the lid and heated and held at a temperature of 121  $^{\circ}$ C (equivalent to a pressure of 103 kPa) for 15 min to 20 min. After discontinuing heating and allowing the temperature to cool to 100  $^{\circ}$ C or less, the exhaust valve shall be opened to bleed steam and the lid shall be opened. The apparatus sterilized in this fashion shall be taken out and, if necessary, allowed to cool in the safety cabinet.

In order to maintain cleanness to prevent contamination by culture media and processing chemicals, the autoclave shall be cleaned with neutral detergent as required and rinsed with water thoroughly.

#### 5.3.3 Flaming

The apparatus to be sterilized shall be subjected to gas or alcohol flame treatment. Platinum loops shall be subjected to flame treatment until they become red hot, while test tubes shall be subjected to flame treatment for two to three seconds.

#### 5.3.4 Sterilization with alcohol

Absorbent cotton or gauze shall be soaked with ethanol for disinfection and squeezed lightly, for use in wiping both hands.

#### 5.3.5 Sterilization through filtration

Liquid to be sterilized shall be filtered with the filter unit.

#### 5.4 Culture media

#### 5.4.1 General

Culture media to be used shall be of the composition shown in <u>Table 1</u> to <u>Table 3</u>.

Table 1 — C culture medium

Tris (hydroxymethyl) aminomethane	500 mg
Ca(NO <sub>3</sub> ) <sub>2</sub> ·4H <sub>2</sub> O	150 mg
KNO <sub>3</sub>	100 mg
Glyceric acid disodium 5.5 hydrate	50 mg
MgSO <sub>4</sub> ·7H <sub>2</sub> O	40 mg
PIV metallic salt solution	3 ml
Cyanocobalamin (vitamin B <sub>12</sub> ) solution	1 ml
Biotin solution	1 ml
Thiamine hydrochloride (vitamin B <sub>1</sub> ) solution	1 ml
Vitamin mixture	0,1 ml
Distilled water (purified water)	994 ml

The contents shall be thoroughly dissolved.

Table 2 — PIV metallic salt solution

Ethylene diamine tetra-acetic acid dihydrogen disodium dihydrate (ED-TA2Na·2H <sub>2</sub> O)	1 000 mg
FeCl <sub>3</sub> ·6H <sub>2</sub> O	196 mg
MnCl <sub>2</sub> ·4H <sub>2</sub> O	36 mg
ZnSO <sub>4</sub> ·7H <sub>2</sub> O	22 mg
CoCl <sub>2</sub> ·6H <sub>2</sub> O	4,0 mg
Na <sub>2</sub> MoO <sub>4</sub> ·2H <sub>2</sub> O	2,5 mg
Distilled water (purified water)	1 000 ml

Table 3 — Vitamin solutions

Cyanocobalamin (vitamin B <sub>12</sub> ) solution	1,0 mg/l
Biotin solution	1,0 mg/l
Thiamine hydrochloride (vitamin $B_1$ ) solution	100 mg/l

As for the PIV metallic salt and vitamin solutions, after thorough dissolution of the contents and subsequent sterilization through filtration, the required quantity shall be dispensed into a sterilized bottle, which shall then be plugged tightly and stored in a dark place at 4 °C. The stored sample shall be used within three months.

#### 5.4.2 C culture medium

After thorough dissolution, the pH shall be adjusted to pH 7,5 with HCl. Finally, sterilization shall be performed by the autoclaving.

#### 5.4.3 Slant medium

In 1 000 ml of the C culture medium, 15,0 g of agar powder shall be mixed. The contents shall be allowed to dissolve thoroughly in a boiling water bath (C agar culture medium). Approximately 10 ml of dissolved C agar culture medium shall be poured into the test tube, which shall then be plugged with a cotton plug and subjected to sterilization with high-pressure steam. After completion of sterilization, the test tube shall be left inclined at about 15° to 30° relative to the horizontal surface in the clean room. The content allowed to solidify in this state is called slant medium.

The slant medium may also be prepared by pouring the high-pressure steam-sterilized C agar culture medium into the test tube previously sterilized with hot air and by allowing the content thus prepared to solidify in the inclined test tube as described above.

#### 5.4.4 Washing solution

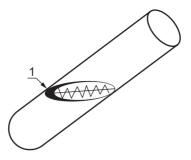
Prepared by adding and dissolving 3,04 g of  $NaH_2PO_4 \cdot 2H_2O$  and 10,92 g of  $Na_2HPO_4 \cdot H_2O$  in 1 000 ml of purified water, followed by sterilization with high-pressure steam.

#### 6 Cultivation of algae

#### 6.1 Transplantation and cultivation of algae

The Chlorella to be tested shall be transplanted aseptically in the clean bench or in an environment with cleanliness equivalent to that of the clean room. The test tube for base strain and that into which transplantation is to be performed shall have their cotton plug and neck wiped with absorbed cotton or gauze soaked with ethanol for disinfection and then set in the tube rack and placed in the clean bench. The test tube with base strain and that with slant medium to which the sample Chlorella is to be transplanted shall be held in one hand and the platinum loop in the other. Cotton plugs shall be removed with the other hand holding the loop, with the mouth of test tube sterilized by flame. Then, the platinum loop shall be sterilized by flame and inserted into a slant medium portion containing condensed water for thorough cooling. The cooled platinum loop shall be placed in the test tube for base strain to scrape off one loopful from the *Chlorella* growth surface and this amount sprayed into the new slant medium. The mouth of the test tube shall be sterilized again by flame and plugged by the cotton plug as originally. After use, the platinum loop shall be sterilized by frame. The slant medium to which *Chlorella* has been transplanted shall be cultivated at a temperature of 25 °C ± 1 °C and under visible light at an intensity of 5 μmol/m<sup>2</sup>s to 10 μmol/m<sup>2</sup>s (about 500 lx to 1 000 lx). When one loopful of Chlorella is to be sprayed over the slant medium, Chlorella shall be dispersed in condensed water as shown in Figure 1 and a straight line shall be drawn from there upward along the slope. An end of the platinum loop shall be lifted temporarily from the medium, immersed again in condensed air, and shall this time draw a meandering line upward along the slope. Do not use the Chlorella cultivated on slant medium for over three months.

NOTE See Figure 1.



#### Key

1 condensed water

Figure 1 — Transplanting Chlorella into slant medium

#### 6.2 Preparation of test solution

One loopful of biomass shall be scraped off from the slant medium described in <u>6.1</u> and placed in an Erlenmeyer flask of 300 ml capacity that contains 150 ml C medium. With a temperature of 25 °C  $\pm$  2 °C, visible light intensity of 20  $\mu$ mol/m²s to 25  $\mu$ mol/m²s (2 000 lx to 2 500 lx) and supply of air through the filter (about 50 ml/min) and gently stirring, cultivation shall be aseptically performed for one week.

50 ml of biomass solution shall be taken out and centrifuged (1  $500 \times g$ , 5 min), then the supernatant shall be removed. The sample fluid shall be re-suspended with 5 ml of distilled water. This process shall be repeated one more time. Then, the cell suspension shall be transferred into an Erlenmeyer flask of 300 ml capacity that contains 150 ml C medium to be 0,2 of the absorbance (1 cm absorption cell) at 530 nm ( $0D_{530}$ ). Liquid culture shall then be performed for one week under similar conditions.

20 ml to 30 ml of culture fluid shall be sampled and centrifuged (1  $500 \times g$ , 5 min), then the supernatant shall be removed. The sample fluid shall be re-suspended with 30 ml of distilled water. This process shall be repeated one more time.

The fluid shall be suspended by adding distilled water and the *Chlorella* concentration shall be adjusted so that  $OD_{530}$  becomes  $10 \pm 0.1$  (about  $10^8$  cells/ml). The solution thus obtained shall be the test solution. When not used immediately, the test solution shall be stored in light-shielded condition in a refrigerator (4 °C  $\pm$  1 °C), but shall be used within one week.

#### 7 Photoirradiation method

## 7.1 Measurement of UV light intensity and preparation of the test piece installation location

The light sensor of the UV radiometer shall be placed on the floor of the UV irradiation system and the film and glass to be used in the test shall be placed over the light sensor. The position at which the value stipulated in 7.2 is obtained shall be determined while reading the indication and this value shall be the position where the test piece is to be placed. When the UV light radiant intensity is measured, the light source of the UV irradiation system shall be kept ON preliminarily for 15 min or longer to obtain stable values.

#### 7.2 Photoirradiation conditions

Photoirradiation shall be performed at room temperature of 25 °C  $\pm$  2 °C, UV radiant intensity of 1,0 mW/cm<sup>2</sup>  $\pm$  0,025 mW/cm<sup>2</sup> for several hours (24 h at the maximum).

#### 8 Test

#### 8.1 Preparation of test piece

The flat portion of photocatalytic-treated antialgal materials shall be cut into a square shape of  $50 \text{ mm} \pm 2 \text{ mm}$  (thickness 10 mm or less), which shall be the standard-sized test piece. The following test pieces shall be prepared: nine test pieces without photocatalytic antialgal treatment (test pieces cut from the base material without photocatalytic antialgal treatment or glass plate) and six photocatalytic-treated antialgal test pieces.

NOTE 1 Of nine test pieces without photocatalytic antialgal treatment, three are used for determination of absorbance immediately after inoculation of test algae solution, three for determination of absorbance after photoirradiation for the specified period of time, and remaining three for determination of absorbance after having been left in a dark place for the specified period of time.

NOTE 2 Of six photocatalytically-treated antialgal test pieces, three are for determination of absorbance after photoirradiation for the specified period of time while remaining three are for determination of absorbance after having been left for the specified period of time in a dark place.

If test pieces without photocatalytic antialgal treatment are not available, glass plates may be used. During preparation of test pieces, due care shall be taken to prevent contamination with microorganism, and cross-contamination and fouling among materials. It is best to prepare test pieces from the material itself. However, if the material is shaped in such a way as to make test piece preparation difficult, these test pieces may be prepared from the material separately processed to a flat plate from the same raw materials by the same processing method.

If the photocatalytcally-treated antialgal material is difficult or impossible to cut into a square of 50 mm  $\pm$  2 mm (thickness 10 mm or less), test pieces having the shape and size other than those specified in this International Standard may be used, provided that their size allows coverage with the film with a surface area of 400 mm<sup>2</sup> to 1 600 mm<sup>2</sup>. If the sample surface is contaminated with organics, preliminary steps may be performed to remove such organics by irradiating the surface with a light source of about 1,0 mW/cm<sup>2</sup> for a maximum of 24 h.

#### 8.2 Cleaning and installation of test pieces

#### 8.2.1 Cleaning of test pieces

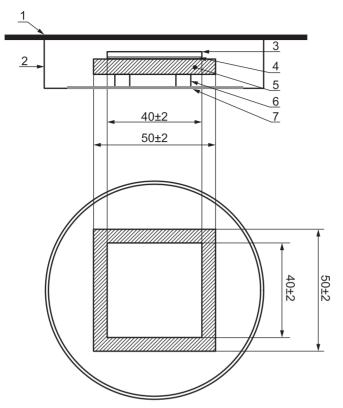
The entire surface of the test pieces described in <u>8.1</u> shall be wiped two or three times lightly with gauze or absorbed cotton soaked with ethanol and dried thoroughly. If this type of treatment can cause changes, such as softening of test pieces, dissolution of surface paint, or elution of contents, which can have adverse effects on test results, the test pieces shall be cleaned according to another appropriate method or shall be used as they are without any cleaning.

The moisture-retaining glass shall be wiped lightly two or three times over the entire glass surface with gauze or absorbed cotton soaked with ethanol and dried thoroughly.

#### 8.2.2 Installation of test pieces

Sterilized moisture-conditioning filter paper shall be placed in the bottom of the sterilized storage petri dish, to which an adequate amount (~8 ml) of sterilized water shall be added. The cuvettes shall be placed so that the test piece and moisture-conditioning filter paper do not make contact with each other. Then, the test piece shall be placed on the cuvettes with the photocatalytic-treated antibacterial surface facing upward (see Figure 2). The surface of photocatalytic-treated material shall be used as the test surface and the cut area shall never be used as a test surface even when the material has been photocatalytic-treated on the inside.

Dimensions in millimetres



#### Key

- 1 moisture-preservation glass
- 2 petri dish (vessel)
- 3 adhesive film
- 4 algae suspension
- 5 test piece
- 6 disposable cuvette
- 7 moisturizing filter paper

Figure 2 — Arrangement of test piece and vessel

#### 8.3 Inoculation of test solution

A 0,1 ml portion of the test solution described in <u>6.2</u> shall be sampled correctly with a pipette and dripped onto each test piece described in <u>8.2.2</u>. Contact film shall be placed to cover the dripped test solution and shall be held down lightly, with care not to cause spillage of the solution from edges of the film, so that the solution is distributed over the entire film. Then, the moisture-retaining glass shall be placed on the film (see <u>Figure 2</u>). Test pieces, excluding those used for determination of absorbance immediately after inoculation of test solution, shall be subject to the tests described in <u>7.2</u> and <u>8.7</u>.

For test pieces of substandard size, the amount of solution to be inoculated shall be divided proportionally according to the area ratio of covering contact film. Even standard size test pieces are used, inoculation of specified amount of solution can result in failure of distribution over the entire area of the film or in spillage of emulsion of bacteria from film edges. In such event, the amount of inoculation solution may be adjusted to give the same cell density on the surface of test piece.

The standard size of contact film shall be  $40~\text{mm}^2 \pm 2~\text{mm}^2$ . When the test piece size is substandard, the size shall be adjusted so that the contact film is 2,5 mm or more inside from the test piece, although the contact film size shall never be smaller than  $400~\text{mm}^2$ . For the film to be used in the test, either that cut previously shall be wiped with alcohol or a sterilized one shall be cut aseptically. When the test piece

is not shaped flat, making close contact with the contact film difficult, the procedure used for covering the film may be omitted.

#### 8.4 Washing of test solution immediately after inoculation of test solution

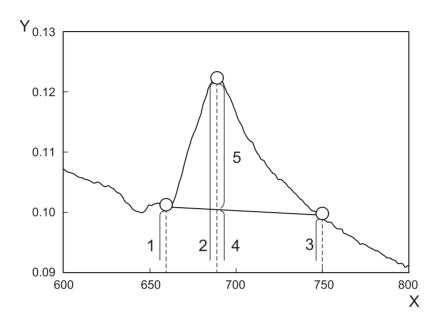
Using tweezers, three test pieces without photocatalytic antialgal treatment immediately after inoculation of test solution shall be placed in a sterilized petri dish to which 5 ml of washing solution has been added. The contact film shall be peeled off with tweezers, drained of solution fully, and placed temporarily on the lid of the petri dish. After immersion of a toothbrush in the washing solution, the test pieces without photocatalytic antialgal treatment shall be brushed lightly with this toothbrush to collect the *Chlorella* into the solution. In this case, they shall be brushed lightly while changing direction several times. The film left on the lid of the petri dish shall be returned to the dish. While the film is held at one end with tweezers, the film side in contact with the test solution shall be brushed lightly with the toothbrush, of which toothbrush shall be lightly rinsed with the same solution and this solution shall be subjected to determination of absorbance (8.6 and 9.2) immediately.

#### 8.5 Washing of test solution after test

For the six test pieces described in 7.2 or six test pieces in 8.7, algae shall be washed out in a fashion similar to that described in 8.4 and this washing solution shall be immediately measured according to the procedure described in 8.6 or 8.7.

#### 8.6 Measurement according to the three-point absorbance method

About 4 ml of washing solution described in 5.4.4 shall be placed in the optical cell and the baseline shall be measured within a range of 400 nm to 800 nm. Then, 4 ml of washing solution obtained in 8.4 or 8.5 shall be placed in the cell for measurement of the continuous absorption spectrum of Chlorophyll in the *Chlorella* (Figure 3) and the values at 660 nm (peak left end), 692 nm (peak maximum value), and 750 nm (peak right end) shall be recorded. When a spectrophotometer incapable of measuring the continuous spectrum is to be used, the meter shall be set to 660 nm and the zero point then measured using the washing solution of 5.4.4. A 4 ml portion of washing solution obtained in 8.4 or 8.5 shall be placed in the optical cell for measurement at 660 nm. The same procedure shall be repeated to determine the values for wavelengths of 692 nm and 750 nm.



#### Key

- 1  $W_{660}$
- $2 W_{692}$
- $W_{750}$
- 4 Wbase692
- 5  $W_{\text{peak}}$
- X wavelength (nm)
- Y absorbance

Figure 3 — Explanatory chart for the three-point absorbance method (8.6 and 9.2)

#### 8.7 Storage in the dark of test piece on which the test solution has been inoculated

Of the test pieces prepared in <u>8.3</u>, three photocatalytic-treated antialgal test pieces and three test pieces without photocatalytic antialgal treatment shall be stored in the dark for a period similar to that described in <u>7.2</u>. In this case, to protect the test piece from light, the storage petri dish may be shielded from light using aluminium foil or may be stored in a dark box.

#### 9 Test results

#### 9.1 General

The test results shall be as follows.

## 9.2 Calculation of antialgal activity on the basis of absorption spectrum method for photocatalytic antialgal materials

Using Formula (4), the antialgal activity, expressed in %, of photocatalyst determined on the basis of absorption spectrum of photocatalytically-treated antialgal materials shall be determined by rounding off to the nearest integer:

$$W_{\text{base}692} = W_{660} + \left(W_{750} - W_{660}\right) / \left(750 - 660\right) \times \left(692 - 660\right) \tag{1}$$

where

 $W_{\rm base692}$  is the absorbance at 692 nm above the base line which is connected absorbance at 660 nm and 750 nm;

 $W_{660}$  is the absorbance at 660 nm;

 $W_{750}$  is the absorbance at 750 nm;

 $W_{692}$  is the absorbance at 692 nm.

$$W_{\text{peak}(0h)} = W_{692} - W_{\text{base}692}$$
 (2)

where

 $W_{\text{peak(0h)}}$  is the difference between absorbance at 692 nm of test piece without photocatalytic antialgal treatment immediately after inoculation of test solution;

 $W_{\rm base 692}$  is the absorbance at 692 nm peak determined according to the three-point method.

$$W_{\text{peak}} = W_{692} - W_{\text{base}692} \tag{3}$$

where

 $W_{\text{peak}}$  is the difference between absorbance at 692 nm of photocatalytic-treated antialgal test piece after photoirradiation test solution;

 $W_{\rm base 692}$  is the absorbance at 692 nm peak determined according to the three-point method.

$$R_{\rm S} = \left(1 - W_{\rm peak} / W_{\rm peak(0h)}\right) \times 100 \tag{4}$$

where

 $R_{\rm S}$  is the antialgal activity, in %, of photocatalyst determined according to the absorption spectrum method.

#### 10 Recording of test results

Recording shall be performed for the type and size of photocatalytic-treated antialgal test pieces and those without such treatment, conditions of preliminary irradiation if such irradiation is performed, type and size of contact film, test bacterial strain, algae storage numbers, amount of incubation of test solution, its concentration, type (name of manufacturer, part number) of light source used, type (name of manufacturer, part number) of UV radiometer used to measure UV irradiation, type (name of manufacturer, part number) of illuminometer, photoirradiation conditions, and respective values of  $R_{\rm S}$ ,  $W_{\rm 692}$ ,  $W_{\rm 750}$ ,  $W_{\rm 660}$ ,  $W_{\rm base692}$ ,  $W_{\rm peak}$ , and  $W_{\rm peak(0h)}$  when the three-point absorbance method is used.

### **Bibliography**

- [1] ISO 80000-1, Quantities and units Part 1: General
- [2] ISO 13125, Fine ceramics (advanced ceramics, advanced technical ceramics) Test method for antifungal activity of semiconducting photocatalytic materials

