International Standard

ISO 23675

First edition
2024-12

Cosmetics — Sun protection test methods — In vitro determination of sun protection factor (SPF)

Cosmétiques — Méthodes d'essai de protection solaire — Détermination in vitro du facteur de protection solaire (FPS)

Reference number
ISO 23675:2024(en)

ISO copyright office
CP 401 • Ch. de Blandonnet 8
CH-1214 Vernier, Geneva
Phone: +41 22 749 01 11
Email: copyright@iso.org
Website: www.iso.org

Published in Switzerland

Contents

Page

Foreword

ISO (the International Organization for Standardization) is a worldwide federation of national standards bodies (ISO member bodies). The work of preparing International Standards is normally carried out through ISO technical committees. Each member body interested in a subject for which a technical committee has been established has the right to be represented on that committee. International organizations, governmental and non-governmental, in liaison with ISO, also take part in the work. ISO collaborates closely with the International Electrotechnical Commission (IEC) on all matters of electrotechnical standardization.

The procedures used to develop this document and those intended for its further maintenance are described in the ISO/IEC Directives, Part 1. In particular, the different approval criteria needed for the different types of ISO document should be noted. This document was drafted in accordance with the editorial rules of the ISO/IEC Directives, Part 2 (see www.iso.org/directives).

ISO draws attention to the possibility that the implementation of this document may involve the use of (a) patent(s). ISO takes no position concerning the evidence, validity or applicability of any claimed patent rights in respect thereof. As of the date of publication of this document, ISO had not received notice of (a) patent(s) which may be required to implement this document. However, implementers are cautioned that this may not represent the latest information, which may be obtained from the patent database available at www.iso.org/patents. ISO shall not be held responsible for identifying any or all such patent rights.

Any trade name used in this document is information given for the convenience of users and does not constitute an endorsement.

For an explanation of the voluntary nature of standards, the meaning of ISO specific terms and expressions related to conformity assessment, as well as information about ISO's adherence to the World Trade Organization (WTO) principles in the Technical Barriers to Trade (TBT), see www.iso.org/iso/foreword.html.

This document was prepared by Technical Committee ISO/TC 217, *Cosmetics*, in collaboration with the European Committee for Standardization (CEN) Technical Committee CEN/TC 392, *Cosmetics*, in accordance with the Agreement on technical cooperation between ISO and CEN (Vienna Agreement).

Any feedback or questions on this document should be directed to the user's national standards body. A complete listing of these bodies can be found at www.iso.org/members.html.

Introduction

Chronic exposure to solar ultraviolet radiation (UVR) is the main environmental source of damage to human skin. Consumer protection against exposure to solar UVB and UVA radiation is, therefore, an important public health issue. The use of sunscreens is a critical part of holistic programs of consumer UVR protection, including the use of appropriate clothing, hats and minimising exposure to the sun around its zenith.

The in vivo sun protection factor (SPF) is historically measured by an in vivo method (see ISO 24444) to communicate the amplitude of protection offered by sunscreens from erythemally-effective solar UVR.[1][2] In recent years, additional test methods have been developed to measure the breadth of protection from solar UVR, namely the in vivo human persistent pigment darkening (PPD) test[3] (and associated UVA-PF) and an in vitro equivalent.[4][5][6][7]

Invasive methods based on tests conducted on human beings are ethically problematic, time-consuming and very costly. Therefore, it has for long been a desire to develop an in vitro SPF test method,[8][9][10][11][12][13][14][15][16][17] recognising the potential advantages of such methodology, including:

a) the use of a non-human model,

b) the significant improvements in speed and cost,

c) the improved repeatability and reproducibility,

d) the elimination of technically-challenging procedures (e.g., MED determination) and

e) the use of a method which is significantly more amenable to continuous improvement.

This in vitro SPF method is based on UVR transmittance spectroscopy, whereby spectrophotometric measurement of UVR transmission through appropriate UVR-transparent substrates, allows prediction of in vivo SPF values.[18][19][20][21][22] This in vitro SPF method revealed a strong reproducibility and correlation with in vivo SPF values.[23][24][25]

Cosmetics — Sun protection test methods — In vitro determination of sun protection factor (SPF)

1 Scope

This document specifies a method for the in vitro determination of sun protection factor (SPF). This method is applicable to sunscreen products in form of an emulsion or alcoholic one-phase formulation, excluding in form of a loose or compressed powder or stick. Specifications are given to enable determination of the spectral absorbance characteristics of SPF protection in a reproducible manner.

Use of this method is strictly for the determination of a static sun protection factor. It is not applicable for the determination of water-resistance properties of a sun protection product.

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.

ISO 24444, *Cosmetics — Sun protection test methods — In vivo determination of the sun protection factor (SPF)*

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:

— ISO Online browsing platform: available at https://www.iso.org/obp

— IEC Electropedia: available at http://www.electropedia.org/

3.1
sunscreen product
product containing any component able to absorb, reflect or scatter UV rays, which are intended to be placed on the surface of human skin with the purpose of protecting against erythema and other ultraviolet induced damage

3.2
emulsion
fine dispersion of minute droplets of one liquid in other(s) in which it is not soluble or miscible

3.3
in vitro sun protection factor
$SPF_{in\ vitro}$
protection factor of a sun protection product against erythema-inducing radiation calculated with spectral modelling between 290 nm and 400 nm

3.4
reference solar spectrum
$I_{sol}(\lambda)$
spectral irradiance of mid-summer sunlight in the spectral range of 290 nm to 400 nm, at a latitude of 40 °N, a solar zenith angle of 20° and an ozone layer thickness of 0,305 cm, as defined in Annex A

3.5
solar UVR simulator
solar ultraviolet radiation simulator
light source emitting a continuous spectrum [S(λ)] with no gaps or extreme peaks of emission in the UV region

Note 1 to entry: The solar simulator has a spectral quality that complies with the required acceptance limits in Annex A.

3.6
erythemal action spectrum
***E*(λ)**
relative effects of individual spectral bands of an exposure source for an erythema response

Note 1 to entry: The symbol for the erythemal action spectrum is defined as $s_{er}(\lambda)$ in ISO/CIE 17166 and $E(\lambda)$ in the ISO 24443.

Note 2 to entry: This entry was numbered 17-401 in CIE S 017:2011.

[SOURCE: CIE-ILV 17-26-065]

3.7
spectrophotometer
instrument for measuring the ratio of 2 values of a radiometric quantity at the same wavelength

Note 1 to entry: This entry was numbered 17-1235 in CIE S 017:2011.

[SOURCE: CIE-ILV 17-25-008]

3.8
monochromatic absorbance
***A*(λ)**
sunscreen absorbance at wavelength λ calculated as logarithm to base 10 of the reciprocal of the spectral internal transmittance, $T(\lambda)$

$$A(\lambda) = -\log 10\ T(\lambda)$$

Note 1 to entry: This entry was numbered 17-1207 in CIE S 017:2011.

[SOURCE: CIE-ILV 17-24-090]

3.9
irradiance at a point of surface
***I*(λ)**
quotient of the radiant flux $d\Phi e$ incident on an element of the surface containing the point, by the area dA of that element

Note 1 to entry: Expressed in $W \cdot m^{-2}$.

Note 2 to entry: Note that the symbol for the irradiance is defined as E in CIE-ILV 017:2020 but because it could be confused with the symbol used in ISO 24443:2021 for the erythemal action spectrum, here we use $I(\lambda)$.

Note 3 to entry: This entry was numbered 17-608 in CIE S 017:2011.

[SOURCE: CIE-ILV 17-21-053]

3.10
spectroradiometer
instrument for measuring radiometric quantities in narrow wavelength intervals over a given spectral region

Note 1 to entry: This entry was numbered 17-1236 in CIE S 017:2011.

[SOURCE: CIE-ILV 17-25-007]

3.11
radiometer
instrument for measuring the intensity of electromagnetic radiation (UV radiation specifically for this standard)

Note 1 to entry: In the context of this document, a UV radiometer measures the irradiance for the UV spectral range from 290 nm to 400 nm.

Note 2 to entry: This entry was numbered 17-1031 in CIE S 017:2011.

[SOURCE: CIE-ILV 17-25-006]

3.12
reference sunscreen formula
product used to validate the testing procedure

3.13
dose
UV exposure dose ($J \cdot m^{-2}$) for pre-irradiation of sunscreen products

Note 1 to entry: The UV dose is the product of UV intensity (expressed as energy per unit surface area) and time.

3.14
plate
piece of polymethylmethracylate (PMMA) on which test product is to be applied for absorbance measurements

Note 1 to entry: See Annex B.

3.15
erythemal irradiance
$I_{ER}(\lambda)$
effective irradiance computed from the product of the spectral irradiance, $I(\lambda)$ and the erythema spectral weighting function, $s_{er}(\lambda)$

Note 1 to entry: Expressed in $W \cdot m^{-2}$.

Note 2 to entry: This entry was numbered 17-403 in CIE S 017:2011.

[SOURCE: CIE-ILV 17-26-067]

3.16
UVB
electromagnetic radiation in the range of 290 nm to 320 nm

3.17
UVA
electromagnetic radiation in the range of 320 nm to 400 nm

3.18
UVA-I
electromagnetic radiation in the range of 340 nm to 400 nm

3.19
UVA-II
electromagnetic radiation in the range of 320 nm to 340 nm

3.20
percentage relative cumulative erythemal effectiveness
% RCEE
description of the spectral distribution of the solar simulator in terms of cumulative erythemal effective irradiance by successive wavelength bands, as defined in Annex A

4 Principles

The test is based on the assessment of UV-transmittance through a thin film of sunscreen spread on at least three moulded PMMA plates and on at least three sandblasted surface PMMA plates, before and after exposure to a controlled dose of radiation from a solar simulator. Samples submitted for testing should not have a SPF or UVA-PF target or other protection category description.

5 Reagents and/or materials

5.1 Sample substrate — Double plate

Moulded and sandblasted PMMA plates shall be used for sunscreen application according to this method (see Annex B).

5.2 Reference sunscreen

The formulae details and manufacturing instructions for the reference formulations are given in Annex C. At least once a month, the following reference standards shall be tested: P2 or P3 reference standard, and P5 or P6 reference standard, and P8 reference standard. The results shall be within the respective acceptance ranges given in Table C.1, Annex C.

5.3 Finger-cot

Finger-cots should be manufactured from untextured and un-powdered latex. As example, for a probe as F.2, a finger-cot of a medium size should be used. If alternative finger-cots are used, a validated equivalent result shall be demonstrated in this method. The finger-cot should be tight on the robot finger probe without ripples or breaks where product can get caught.

5.4 Blank

Glycerin or a modified glycerin solution (see Table B.1), or white petroleum in accordance with Annex D shall be used for blank measurement.

6 Apparatus

6.1 Spectrophotometers

6.1.1 Specification

The spectrophotometer shall span the spectral range from 290 nm to 400 nm. The wavelength increment step shall be 1 nm. A spectrophotometer that does not use a monochromator to analyse the reflected or transmitted radiation from the test sample should employ a fluorescence rejection filter. Its input optics should be designed for diffuse illumination and/or diffuse collection of the transmitted irradiance through the roughened polymethylmethacrylate (PMMA)plate, with and without the sunscreen layer spread on its surface. The size of the diameter of the entrance port of the spectrophotometer probe should be smaller than the size of the light spot to be measured at the sample level (in order to account for stray light). The area of each reading site shall be at least 0,5 cm^2 in order to reduce the variability between readings and to compensate for the lack of uniformity in the product layer. The wavelength should be accurate to within 1 nm, as checked using a holmium-doped filter (see Annex E). The ability of an instrument to accurately measure absorbance is limited by the sensitivity of the instrument. The minimum required dynamic range for this methodology is 2,2 absorbance units as determined in accordance with Annex E. The maximum measured absorbance should be within the dynamic range of the device used. If the test measurements yield absorbance curves that exceed the determined upper limit of the spectrophotometer, the product should be re-tested using an instrument with increased sensitivity and dynamic range.

In order to minimise scatter, the distance between the closest side of the plate and the emission source or the integrating sphere should not be more than 2 mm. The plate shall be positioned in a horizontal plane during all steps including UV measurement steps.

The lamp in the spectrophotometer (or spectroradiometer) that is used to measure the absorbance shall emit a continuous spectrum of radiation (with no gaps or extreme peaks of emission in the UV region) over the range of 290 nm to 400 nm, and the level of irradiance should be sufficiently low, so that the photostability of the product is not unduly challenged, wherein the UV dose during one measurement cycle should not exceed 0,2 $J \cdot cm^{-2}$.

6.1.2 Monitoring

The spectrophotometer shall be validated every month by measurements of reference materials.

A four-fold test is required, as described in Annex E:

— dynamic range of the spectrophotometer;

— linearity test of the spectrophotometer;

— wavelength trueness test;

— absolute transmission trueness.

6.2 Automatic positive-displacement pipette

Positive displacement pipettes, micro-pipettes, automatic pipettes or any similar device should use piston-driven displacement and shall be capable of delivering accurate and repeatable aliquots of approximately 1,6 mg to 1,8 mg of a sunscreen product (as described in 7.3.1).

NOTE 1,6 mg to 1,8 mg correspond to approximately 1,6 µl to 1,8 µl, respectively.

6.3 Analytical balance

A laboratory balance with at least 10^{-4} g precision shall be used.

6.4 Robot

6.4.1 Specifications

The robot shall be in accordance with Annex F and shall have:

a) positional repeatability of at least ±0,1 mm in *x*, *y* and *z* axes,

b) degrees of freedom equal to at least 6 rotating joints,

c) a payload of at least 0,5 kg,

d) a vertical force (*z* axis), measured in the centre of the plate (with the finger tool and finger cot, without *x* and *y* axis movement), of (6,0 ± 0,5) N.

6.4.2 Monitoring

The robotic appliance shall be checked by a suitably qualified expert at regular intervals (at least every twelve months) to ensure compliance to the mechanical and spreading specifications given in 6.4.1.

The finger tool shall be replaced after every cycle of 400 spreading operations or when damaged (e.g. cracks, etc.).

6.5 Solar simulator

6.5.1 General

A xenon arc solar simulator with appropriate filters should be used and shall conform with the spectral specifications described in Table A.1 and Figure A.1. It shall be able to maintain a stable plate temperature of (27 ± 2) °C.

6.5.2 Quality of solar simulator radiation

The output from the solar UVR simulator shall be continuous, uniform, stable, with no gaps or extreme peaks of emission in the UVR region and suitably filtered to create a spectral quality that complies with the required acceptance limits (see Table A.1).

To ensure that appropriate amounts of UVA radiation are included in the spectrum of the solar UV simulator, the total radiometric proportion of UVA-II irradiance of the simulator (320 nm to 340 nm) shall be ≥ 20,0 % of total UVR irradiance (290 nm to 400 nm) in accordance with ISO 24444 which requires the same solar irradiance. Additionally, UVA-I region (340 nm to 400 nm) irradiance shall be ≥ 60 % of total UVR irradiance. The source spectral specification is described in terms of cumulative erythemal effective irradiance by successive wavelength bands, 290 nm to 400 nm. The erythemal effective irradiance of each wavelength band is expressed as a percentage of total erythemal effective irradiance, 290 nm to 400 nm, or as percentage relative cumulative erythemal effectiveness (%RCEE). The calculation of %RCEE values shall be in accordance with Annex A, where acceptance limits are shown in Table A.1.

Total irradiance shall not exceed 200 $W \cdot m^{-2}$. The output of the solar simulator shall be measured with a broad-spectrum sensor (capable of measuring between 280 nm and 1 600 nm) calibrated against a standard reference source over the range of 280 nm to 1 600 nm. Alternatively, the source may be measured with a calibrated spectroradiometer over this same wavelength range to determine the total irradiance.

In broad-beam UV-sources, spectra from different locations under the beam shall be recorded over at least 5 different locations (a location is defined for each plate) in order to account for uniformity.

The uniformity shall be ≥ 90 % as calculated by Formula (1):

$$U = (1\text{-}(\max\text{-}\min)/(\bar{X})) \quad (1)$$

where

U is the uniformity in percentage;

$\bar{X}$ is the average.

If the uniformity is less than 90 %, then optical components should be adjusted (and a new beam uniformity control shall be performed) or appropriate compensation for different irradiance shall be made in the exposure time on each plate.

6.5.3 Maintenance and monitoring the solar simulator

The emission of the UV exposure source used for exposure shall be checked for compliance with the given acceptance limits by a suitably qualified expert (at least) every 12 months, or 2 500 h of lamp running time and after changing any significant physical (optical) component of the solar simulator (including the bulb only if the bulb was not already previously calibrated with the associated solar simulator). The inspection should be conducted with a spectroradiometer that has been calibrated against a standard lamp that is traceable to a national or an international calibration standard. Prior to the UV exposure of sample, the UV intensity of the exposure source output shall be measured and recorded with a spectroradiometer (as detailed in 6.1) or an erythema weighted radiometer cross-calibrated against a spectroradiometric measurement of each assigned solar simulator output as detailed in 6.5.2. Optical alignment shall be configured to ensure accurate radiometer alignment and reproduction of the assigned simulator output at the same optical reference

plane measured with the spectroradiometer. A calibration factor Y for each radiometer with assigned solar simulator output shall be determined by Formula (2):

$$Y = I_{ersp}/I_{err} \quad (2)$$

where

Y is the calibration factor for each radiometer;

I_{ersp} is $I(\lambda) \times s_{er}(\lambda)$ measured by the spectroradiometer;

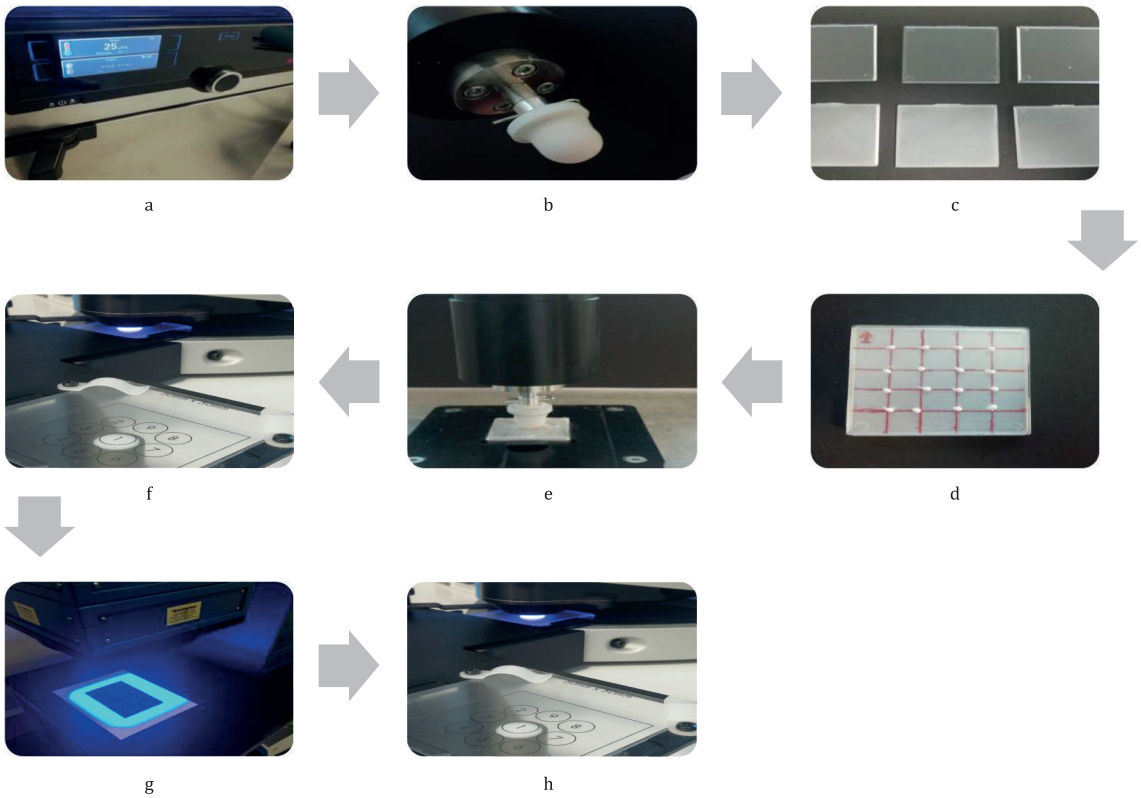
I_{err} is $I(\lambda) \times s_{er}(\lambda)$ measured by the radiometer.

7 Procedure

7.1 Outline of the test procedure

1) Preparation of reagents and materials.
2) Product application on plates and robot automatic spreading.
3) Measurement of initial absorbance using two plate types (290 nm to 400 nm).
4) Calculation of initial in vitro SPF.
5) Calculation of irradiation dose (based on initial in vitro SPF).
6) Irradiation with calculated dose.
7) Measurement of final post-irradiation absorbance using two plate types (290 nm to 400 nm).
8) Calculation of final in vitro SPF.

See Figure 1.



a Incubation at (27 ± 2) °C, at least 12 h before.

b Finger tool and finger cot installation.

c At least three moulded PMMA plates and three sandblasted plates.

d Product deposit.

e Spreading and drying step.

f First absorbance measurements.

g UV exposure.

h Second absorbance measurements.

Figure 1 — Key steps of the method

7.2 Preparation of reagents and materials

7.2.1 Plate preparation and handling

Three (at least) moulded PMMA plates and three (at least) sandblasted plates shall be used, each in accordance with the specifications in Annex B.

Plates and product shall be stored, in the dark, at (27 ± 2) °C for at least twelve hours before the start of the test.

The surface of sandblasted plates should be cleaned with a dry, antistatic microfibre cloth.

The plates should be handled carefully by holding them by the edges, avoiding finger contact with the surface.

A reference mark should be made on edge of each plate, outside of spreading area, to ensure the whole measurement process proceeds in the same order with plate placed in same orientation each time.

The plates shall be used without additional treatment on surface (chemical and/or physical). The plates shall be used only once.