INTERNATIONAL STANDARD

**ISO 8255-1**

Third edition
2017-06

# Microscopes — Cover glasses —

## Part 1: Dimensional tolerances, thickness and optical properties

*Microscopes — Lamelles couvre-objet —*

*Partie 1: Tolérances dimensionnelles, épaisseur et propriétés optiques*



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# Foreword

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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 documents 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).

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The committee responsible for this document is ISO/TC 172, *Optics and photonics*, Subcommittee SC 5, *Microscopes and endoscopes*.

This third edition cancels and replaces the second edition (ISO 8255-1:2011), of which it constitutes a minor revision to correct the designation of cover glass.

A list of all parts in the ISO 8255 series can be found on the ISO website.



 

# Introduction

This document defines dimensions and specifies optical quality requirements in order to guarantee the quality of observation.

The data given in this document are applicable to most products in use and have been adapted to take into account the relevant national standards in vigour.

This document contains requirements for dimensional tolerances, thickness and optical properties, whereas quality requirements and test methods related to the material are dealt with in ISO 8255-2.

# Microscopes — Cover glasses —

# Part 1:
# Dimensional tolerances, thickness and optical properties

## 1 Scope

This document specifies requirements for dimensional tolerances, thickness and optical properties for microscope cover glasses used for transmitted light microscopy in the visible spectral range (400 nm to 760 nm).

## 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 8036, *Microscopes — Immersion liquids for light microscopy*

## 3 Terms and definitions

No terms and definitions are listed in this document.

ISO and IEC maintain terminological databases for use in standardization at the following addresses:

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

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

## 4 Requirements

### 4.1 General

All media that are located between the specimen and the microscope objective are in their optical effect part of the objective. Such media are usually cover glasses and immersion media. Immersion media are defined in ISO 8036; their refractive index shall be taken into account for the selection of the cover glass.

Microscope objectives, unless equipped with correction collars, are designed for a specific immersion medium (e.g. air, oil or water) and cover glass thickness. The design thickness is $t$ = 0,17 mm, unless otherwise marked on the objective.

When using microscope objectives with high numerical aperture, deviations from the nominal cover glass thickness leads to severe optical aberrations, mainly spherical aberration.

The refractive index of the cover glass material needs to be specified for a broad spectral range to maintain good chromatic correction. This is achieved by specification of the refractive index, $n_e$, for a reference wavelength ($\lambda_e$ = 546,07 nm) near the maximum of the eye's spectral sensitivity and the corresponding Abbe number, $\nu_e$.

### 4.2 Dimensional tolerances for thickness

The thickness tolerances of cover glasses shall be in accordance with Table 1.



 

**Table 1 — Dimensional tolerances of types of cover glasses**

Dimensions in millimetres

| Designation/Type | Design thickness<br>$t$ | Tolerance |
|---|---|---|
| No. 1 ½ | 0,17 | $^{+0,02}_{-0,01}$ |
| No. 1 ½ H | 0,17 | $^{+0,005}_{-0,005}$ |
| No. 1 | 0,17 | $^{+0,00}_{-0,04}$ |

In addition to the thicknesses given in Table 1, cover glasses are available in other thicknesses. These can be used for certain applications or in combination with objectives specifically designed for such non-standard cover glasses. However, it shall be pointed out that the highest optical quality may not necessarily be obtained with such cover glasses, when using objectives of high numerical aperture.

Annex A provides guidelines for the selection of cover glass No. 1 ½ and No. 1 ½ H with objective numerical aperture values in combination with common immersion media.

## 4.3 Dimensional tolerances for length, width and diameter

The limiting tolerances for nominal length, $l$, and width, $w$, for rectangular cover glasses (Form A) as shown in Figure 1 or diameter, $d$, for round cover glasses (Form B) as shown in Figure 2 are ±0,5 mm.

Typical dimensions for rectangular and round cover glasses are given in Table B.1.

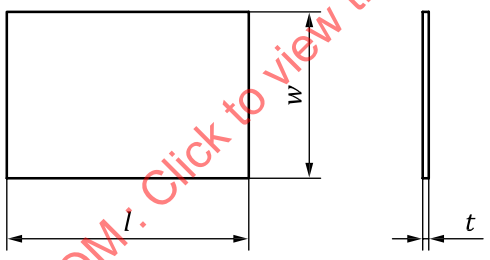


**Figure 1 — Form A: Rectangular cover glass**

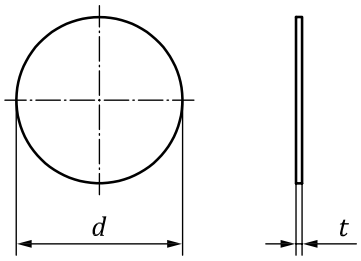


**Figure 2 — Form B: Round cover glass**

## 4.4 Optical properties

Cover glasses shall have the following optical properties:

Principal refractive index: $n_e = 1,525\ 5 \pm 0,001\ 5$

Abbe number: $\nu_e = 56 \pm 2$

NOTE The principal refractive index, $n_e$, is the index of refraction for light of wavelength $\lambda_e$ = 546,07 nm (green Mercury e-line). This wavelength is located close to the maximum spectral sensitivity of the human eye and is commonly used as a reference wavelength for optical computations (see ISO 7944).

The Abbe number, $v_e$, is computed according to Formula (1):

$$v_e = \frac{n_e - 1}{n_{F'} - n_{C'}} \quad (1)$$

where

$n_e$ is the principal refractive index;

$n_{F'}$ is the refractive index for light of wavelength $\lambda_{F'}$ = 479,99 nm (blue Cadmium F′-line);

$n_{C'}$ is the refractive index for light of wavelength $\lambda_{C'}$ = 643,85 nm (red Cadmium C′-line).

## 5 Marking and labelling

The packaging of microscope cover glasses that comply with this document shall display the following information:

a) the thickness, No. 1 ½, No. 1 ½ H, or No. 1;

b) the dimensions for length and width, or diameter;

c) the average number of cover glasses per package or mass;

d) the name of the manufacturer or supplier and country of origin;

e) a marking to show that the cover glasses comply with the requirements of this document.

# Annex A
(informative)

# Guidelines for selection of cover glass type

To achieve optimum results, using the cover glass types shown in Table A.1 is recommended. The proper selection of the cover glass type depends on the application and individual properties of the objective as shown in Table A.1. Users, including those with objectives with mechanisms to correct for cover glass thickness, should always use cover glass that conforms with the other optical properties specified in this document.

**Table A.1 — Guidelines for selection of cover glass type**

| Immersion medium | Min. numerical aperture | Max. numerical aperture | Type of cover glass |
|---|---|---|---|
| **Air** | 0,35 | 0,70 | No. 1 ½ |
| **Air** | 0,70 | — | No. 1 ½ H |
| **Water** | 0,60 | 0,90 | No. 1 ½ |
| **Water** | 0,90 | — | No. 1 ½ H |
| **Glycerol** | 0,80 | 1,10 | No. 1 ½ |
| **Glycerol** | 1,10 | — | No. 1 ½ H |
| **Immersion oil** | 0,90 | 1,30 | No. 1 ½ |
| **Immersion oil** | 1,30 | — | No. 1 ½ H |

# Annex B
(informative)

# Typical dimensions for rectangular and round cover glasses

**Table B.1 — Typical dimensions for rectangular and round cover glasses**

Dimensions in millimetres

| Form | Length<br>*l*<br>±0,5 | Width<br>*w*<br>±0,5 | Diameter<br>*d*<br>±0,5 |
|---|---|---|---|
| A | 18 | 18 | — |
| | 22 | 22 | — |
| | 24<br>32<br>40<br>50<br>60 | 24 | — |
| B | — | — | 18 |
| | — | — | 22 |