



## Introduction

Argolight multidimensional slides are specifically designed for assessing and following the performances of fluorescence-based imaging systems.

Argo-SIM slides are specifically designed for structured illumination microscopes, as well as any system exploiting deconvolution algorithms. The slides consist in a special glass piece (ArgoGlass) set on a stainless steel carrier. Different fluorescent patterns are embedded inside the glass. They also exhibit a contrast in bright and dark fields, DIC (Differential Interference Contrast) and phase contrast. The patterns are accurately positioned and stable to light illumination. The analysis of pattern images can be simplified using Argolight software solutions.

## Fluorescent properties

Patterns are excitable from 300 nm to 550 nm, with a maximum emission efficiency at around 340 nm. The emission is a broad continuum and the efficiency decreases as the excitation wavelength shifts towards the red.

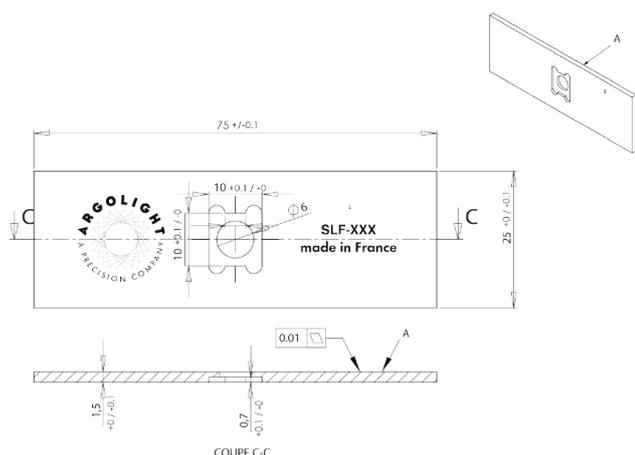
## Stability of the fluorescence

Under normal use (irradiances, either peak or average, no higher than 50 GW/cm<sup>2</sup>), the fluorescence spectrum for a given illumination setting is identical in shape and intensity to any similar part of the patterns.

Under specific illumination configurations, the intensity of the patterns may decrease. However, this decrease is transient. The fluorescence intensity recovers to its initial value after some time. The recovery time depends on the irradiation conditions (power density, wavelength, pixel size, exposure time).

This behaviour is reproducible: for a given intensity and exposure time, the rate of decrease and recovery time will always be the same.

## Schematics of the slide



## ArgoGlass Description

ArgoGlass is a special glass produced at the Argolight facility to insure its homogeneity and purity. Its refractive index is similar to the glass used in glass slides or glass coverslips.

## Slide compatibility

### Imaging compatibility

Compatible	Not compatible but not damaging	Not compatible and damaging
Widefield Microscopy Confocal Microscopy Structured Illumination Microscopy FLIM Spinning Disk Microscopy	PALM STORM FRAP FRET Any imaging technology using depletion or multiple dyes.	STED Multiphoton Microscopy Any imaging technology using ultrashort pulsed laser

### Objective compatibility

The slides are compatible with dry and oil immersion objectives. The slides are compatible with water as an immersion liquid, but continuous exposure higher than five minutes should be avoided.

## Patterns inside the slide

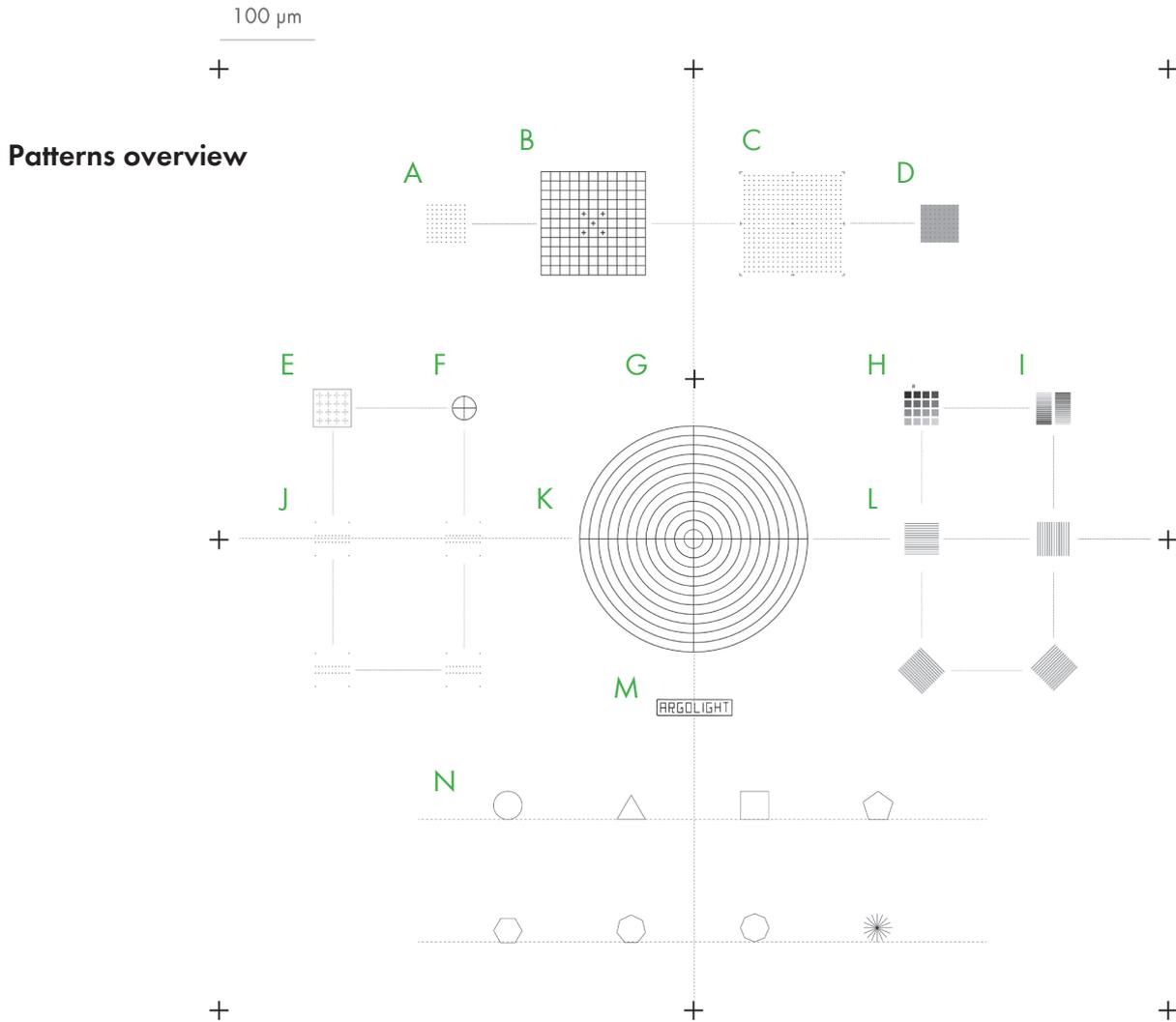
Patterns are positioned ( $170 \pm 5$ )  $\mu\text{m}$  below the top glass surface, on a horizontal plane which flatness is within  $\pm 5$  mrad.

This emulates the presence of a microscope cover-slip, having a thickness of ( $170 \pm 5$ )  $\mu\text{m}$  and a refractive index of ( $1.5255 \pm 0.0015$ ) at 546.1 nm.

The maximum relative positioning error is  $\pm 110$  nm in XY and  $\pm 110$  nm in Z within each individual pattern.

The thickness (in the Z direction) of these patterns is about ( $600 \pm 200$ ) nm FWHM (Full Width at Half Maximum).

Patterns description can be found in the back of this spec sheet.



**Patterns description**

The slide contains 14 types of patterns.

**Pattern A - 3D Matrix of rings.** A 3D matrix of  $9 \times 9 \times 9$  rings, separated by  $5 \mu\text{m}$ , on a total volume of  $40 \mu\text{m} \times 40 \mu\text{m} \times 40 \mu\text{m}$ .

**Pattern B - Grid.** A grid with a step of  $10 \mu\text{m}$ , with 5 crosses of  $5 \mu\text{m}$  length in some squares.

**Pattern C - 2D Matrix of rings.** A 2D matrix of  $21 \times 21$  rings, separated by  $5 \mu\text{m}$ , on a total field of  $110 \mu\text{m} \times 110 \mu\text{m}$ . The field of rings is surrounded by eight landmarks, and exhibits a  $3 \mu\text{m}$  long cross in its center.

**Pattern D - Matrix of rings on a background.** This pattern is identical to pattern A, with a background that is  $10 \mu\text{m}$  below.

**Pattern E - Matrix of crosses.** A matrix of  $4 \times 4$  crosses, having a length of  $5 \mu\text{m}$  and separated by  $10 \mu\text{m}$ , surrounded by a frame. The crosses are composed of vertical lines that are in the same plane, and by horizontal lines, going gradually deeper within the glass. The spacing between the vertical and horizontal lines gradually increases from 0 to  $1.5 \mu\text{m}$ , with a step of  $100 \text{nm}$ .

**Pattern F - Meridians of a sphere.** Three circles of diameter  $25 \mu\text{m}$  in different orthogonal planes, featuring the meridians of a sphere.

**Pattern G - Repositioning crosses.** The repositioning crosses are  $20 \mu\text{m}$  long and are positioned  $500 \mu\text{m}$  from one to another in

the X direction, the Y direction, or both.

**Pattern H - 4 x 4 Intensity.** Sixteen  $6 \mu\text{m}$ -wide squares having different fluorescence intensity levels following a linear evolution, organized in a  $4 \times 4$  matrix.

**Pattern I - 2x16 Intensity** Twice sixteen  $15 \mu\text{m} \times 0.7 \mu\text{m}$  rectangles having different fluorescence intensity levels following a linear evolution, organized in a  $2 \times 16$  matrix.

**Pattern J - 3D Crossing stairs.** Empty cylinders embedded at different depths, like two crossing stairs, surrounded by four pillars. There are four stairs in the slide, with varying steps:  $1, 0.5, 0.25$  and  $0.125 \mu\text{m}$ .

**Pattern K - Target.** Concentric circles with increasing radii from  $10 \mu\text{m}$  to  $120 \mu\text{m}$  with a step of  $10 \mu\text{m}$ , featuring a target.

**Pattern L - Gradually spaced lines.** Pairs of  $36 \mu\text{m}$ -long lines which spacing gradually increases, from  $80$  to  $340 \text{nm}$ , with a step of  $20 \text{nm}$ . Four sets of lines are present: one vertical, one horizontal, and two oriented at  $+ \text{ and } - 45^\circ$ .

**Pattern M - Logo.** Letters forming the name "Argolight", and surrounded by a  $80 \mu\text{m} \times 18 \mu\text{m}$  frame.

**Pattern N - Geometrical figures.** A circle, a triangle, a square, a pentagon, an hexagon, an heptagon, an octagon and a star with 16 arms.