

**Mouse kidney section.**  
Maximum projection of a stack containing 65 planes.  
Green: glomeruli and convoluted tubules (wheat germ agglutinin Alexa Fluor® 488)  
Blue: Nuclei (DAPI).

Structured Illumination

# Leica Microsystems' Intelligent Structured Illumination Microscopy

The Integrated System Solution with OptiGrid®  
for Upright and Inverted Research Microscopes

Living up to Life

# Leica Microsystems' Intelligent Struct

## Leica DMI4000 B and DMI6000 B **Inverted** Research Microscopes

### Key Benefits

#### Excellent results

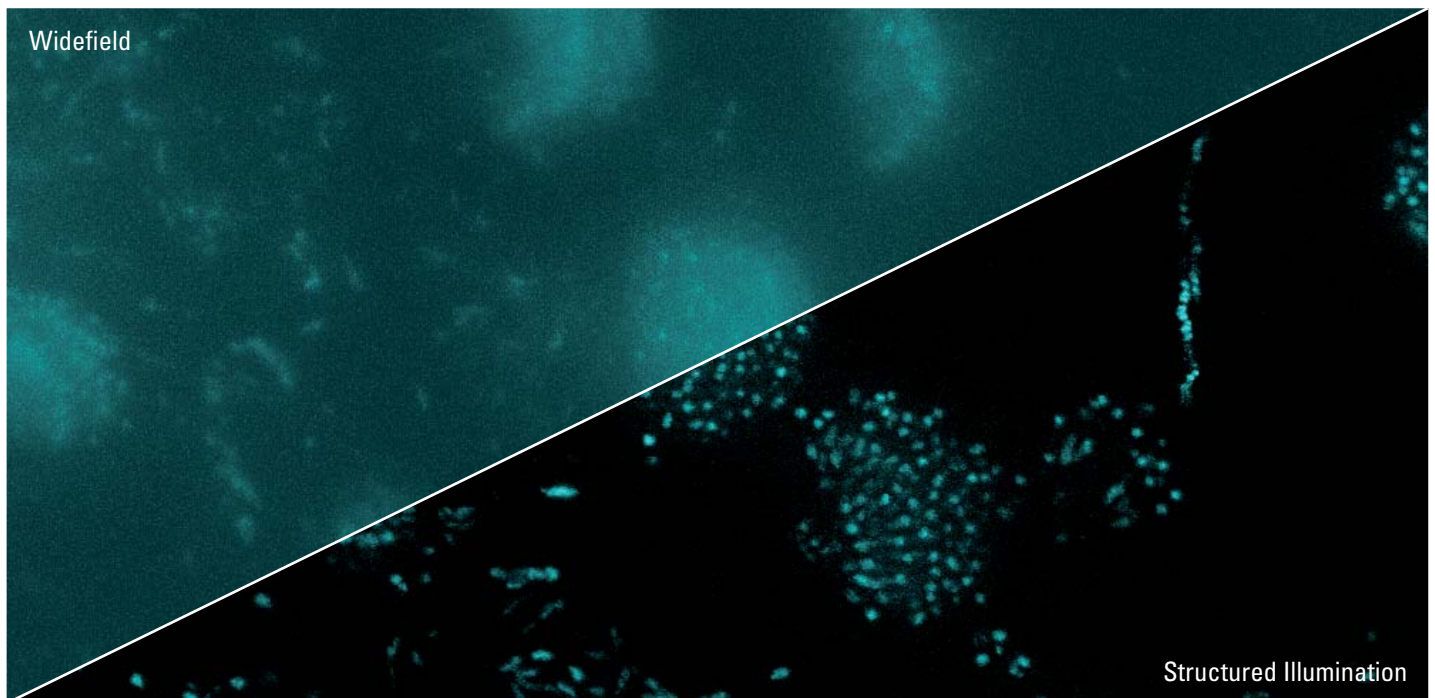
When the magnification or contrast method is changed, the Illumination Manager automatically sets the optimum aperture diaphragm, field diaphragm, and light intensity parameters. Prisms, modulators, analyzers or polarization filters are switched in or out of the light path accordingly. This provides reliable, comparable, and reproducible results.

#### Quality and speed

The perfect interplay of fluorescence-optimized optics in the fluorescence module is a unique Leica development: The fast internal filter wheel and the high-speed shutter in conjunction with the integrated Intensity Manager deliver the best fluorescence images in record time.



Perfect integration of the OptiGrid® module into Leica research microscopes



**Nuclei in spore carriers of *Aspergillus nidulans*.** Maximum projection of a stack containing 138 planes. Blue: Nuclei (DAPI).  
Courtesy of Prof. Dr. Reinhard Fischer and Nadine Zekert, KIT – Karlsruhe Institute of Technology, Germany.

# Structured Illumination Microscopy – Brilliant

## Superior Contrast for Ultra Fine Structures

- **Integrated system solution:** perfect integration of the OptiGrid® module with Leica research microscopes
- **Ultra high contrast:** smart combination of electronics and optics for high-resolution, high-contrast 2D and 3D fluorescence images
- **Compatible:** the same OptiGrid® module can be used in both upright and inverted Leica research microscopes
- **Convenient and time saving:** one optimized grid for all objectives
- **Automatic grid focusing:** high-resolution fluorescence images at the press of a button
- **Powerful software:** Leica MM AF makes setup and image acquisition easy

### Perfect harmony, maximum image quality

Intelligent Structured Illumination Microscopy from Leica Microsystems uses the proven OptiGrid® technique. Working in harmony with Leica's superior quality optics, the aperture diaphragm integrated with the fluorescence light path also enhances contrast. The aperture diaphragm can be controlled via the software or the intuitive Leica SmartTouch interface.

### Always in focus

For fast, reproducible results: the automatic focus function of the OptiGrid® keeps the grid structure in focus, from UV to IR light.

### The Leica solution

A unique feature of this development is that the **same OptiGrid® module** can be used **for both upright and inverted** Leica research microscopes. Furthermore, there's **no need to change the grid**, as one grid covers all magnifications from 10x to 100x, which offers convenience, avoids errors, and saves valuable time.

### The complete system

Intelligent Structured Illumination Microscopy from Leica Microsystems is a complete system from a single source: The OptiGrid® module integrated with Leica research microscopes is perfectly complemented by the intuitive Leica MM AF software and a wide selection of fluorescence cameras.



# nt Fluorescence Images

## Leica DM5000 B, DM5500 B and DM6000 B Upright Research Microscopes

### Key Benefits

#### Intelligent automation

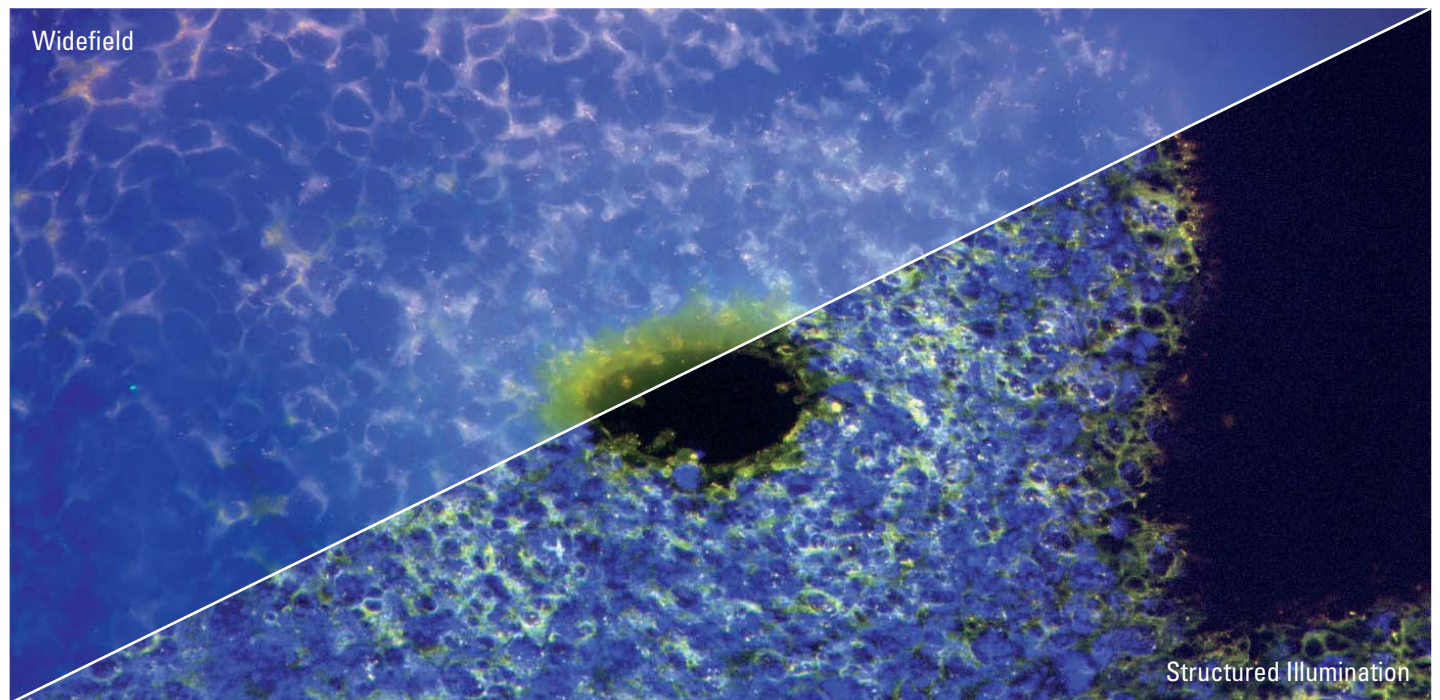
The optimal parameters are automatically set in transmitted light and fluorescence, depending on the selected contrast method. All settings, including the fine adjustments for objective prisms, are saved for each objective. The fully automated fluorescence axis with **apochromatic** fluorescence light path provides brilliant images with stunning contrast. Intelligent automation means: **absolute reproducibility and excellent image quality.**

#### Fluorescence Intensity Manager

Leica Microsystems' unique patented Fluorescence Intensity Manager (FIM) enables fast, precise, and reproducible setting of fluorescent light intensity. Leica's FIM eliminates photo-bleaching by instantly restoring your optimal settings.



The same OptiGrid® module can be used in both upright and inverted Leica research microscopes



**Eye anlage of E10 mouse embryo.** Maximum projection of 84 z-sections, 0.2  $\mu\text{m}$  z-distance, 40x/1.25. DNA: Hoechst 33342, beta-Tubulin: AlexaFluor®488, AlexaFluor®633  
Sample: Courtesy of Dr. Yves Lutz, Imaging Center IGBMC, Strasbourg, France

## Structured Illumination – the Smart Solution

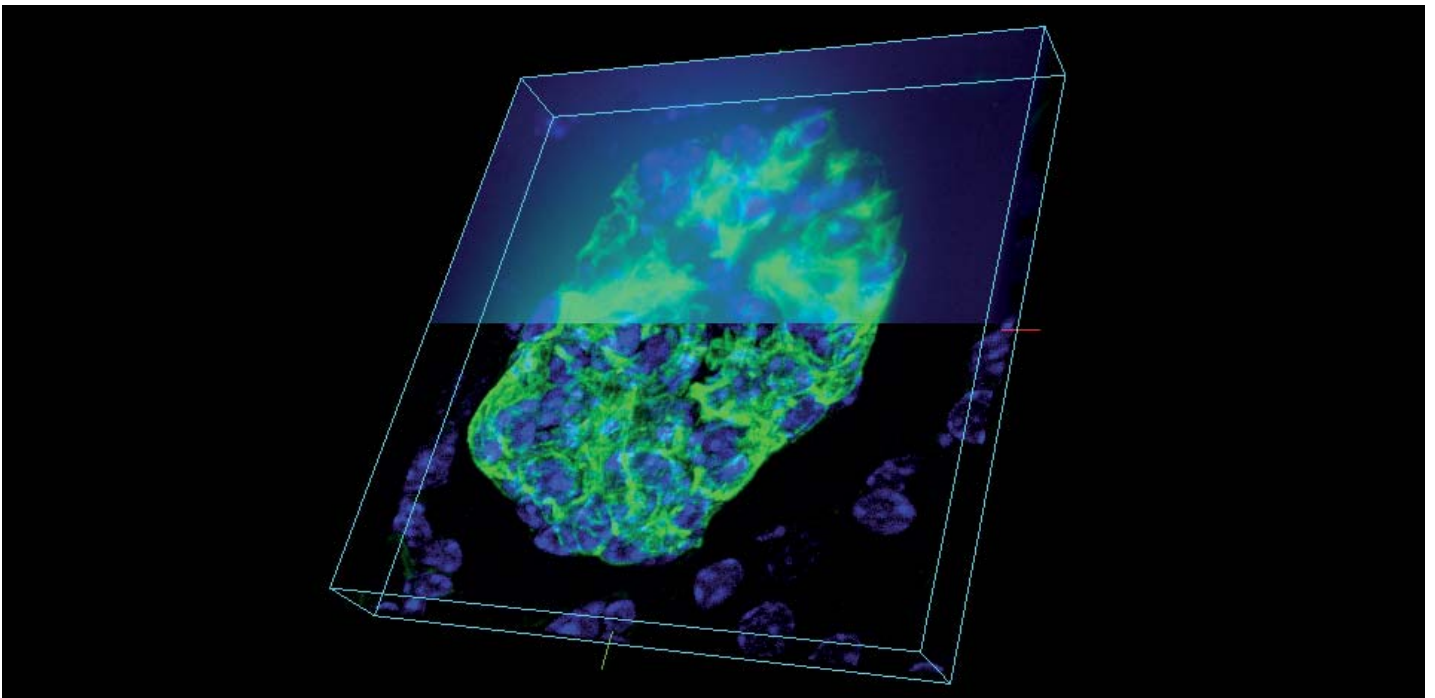
Resolution and contrast are crucially important for fluorescence applications in widefield microscopy. However, haze can be a problem when imaging thick specimens. Fluorescent light from different planes in the specimen reaches the focal plane, blurring the image and reducing contrast. There are various options for haze reduction, either confocal microscopy, time-consuming deconvolution – or the smart principle of structured illumination. The result: excellent contrast, superb axial resolution and ultra-sharp 2D sections of the specimen.

### The principle

A grid structure in the field diaphragm plane of the fluorescence axis is sharply imaged in the object plane. At least three raw images are captured, moving the grid structure each time, and used by the patented OptiGrid® algorithm to calculate a final image.

### The result

The clear image, haze-free and with enhanced contrast, can be immediately viewed on the PC monitor. Also, a sharply defined 3D reconstruction of the object can be generated by recording several z planes.



**3D reconstruction of a glomerulus (mouse kidney).** Maximum projection of a stack containing 139 planes. With the software package Leica MM AF a 3D reconstruction was performed. The better contrast as well as the better resolving power of small structures is clearly visible after the use of the OptiGrid®.

# Leica MM AF Powered by MetaMorph®

## The Integrated System for Bioimaging

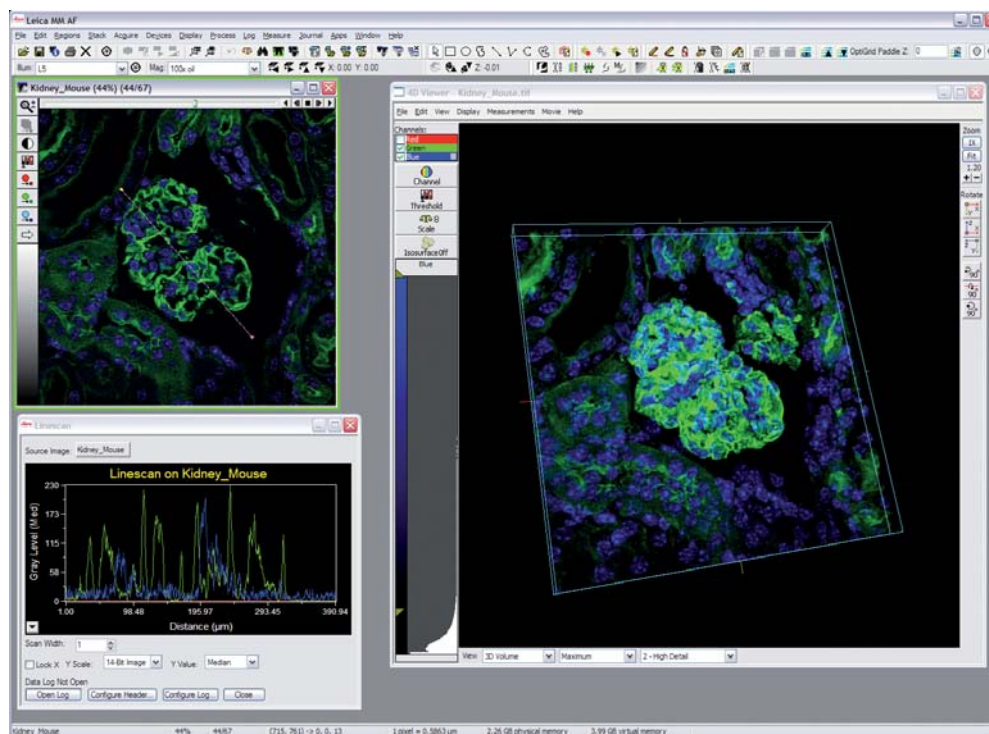
Leica MM AF Acquisition and Analysis is a software suite for the control and integration of Leica Microsystems research microscopes. Leica MM AF offers a large number of modules for sophisticated applications and analysis techniques.

### Easy tailoring

Intelligent Structured Illumination Microscopy is controlled with powerful Leica MM AF software. Leica MM AF guides you quickly and intuitively through the setup process and also makes it easy to acquire fluorescence images. You can use the Journal Editor to tailor your applications and automate the analysis routines to meet specific requirements: The functions you define are then executed at the press of a button.

**The Leica MM AF software suite also covers the following applications:**

- Multidimensional imaging
- Image overlay
- Colocalization
- Morphometric measurements
- Intensity quantifications
- Time-lapse imaging
- Morphological filters
- Manual tracking



Leica MM AF powered by MetaMorph®. Easy acquisition and analysis can be done within the Leica MM AF software package. The application module 4D viewer (optional) can display multidimensional datasets for the simultaneous view of multiple z section, wavelength, time points, stage position.