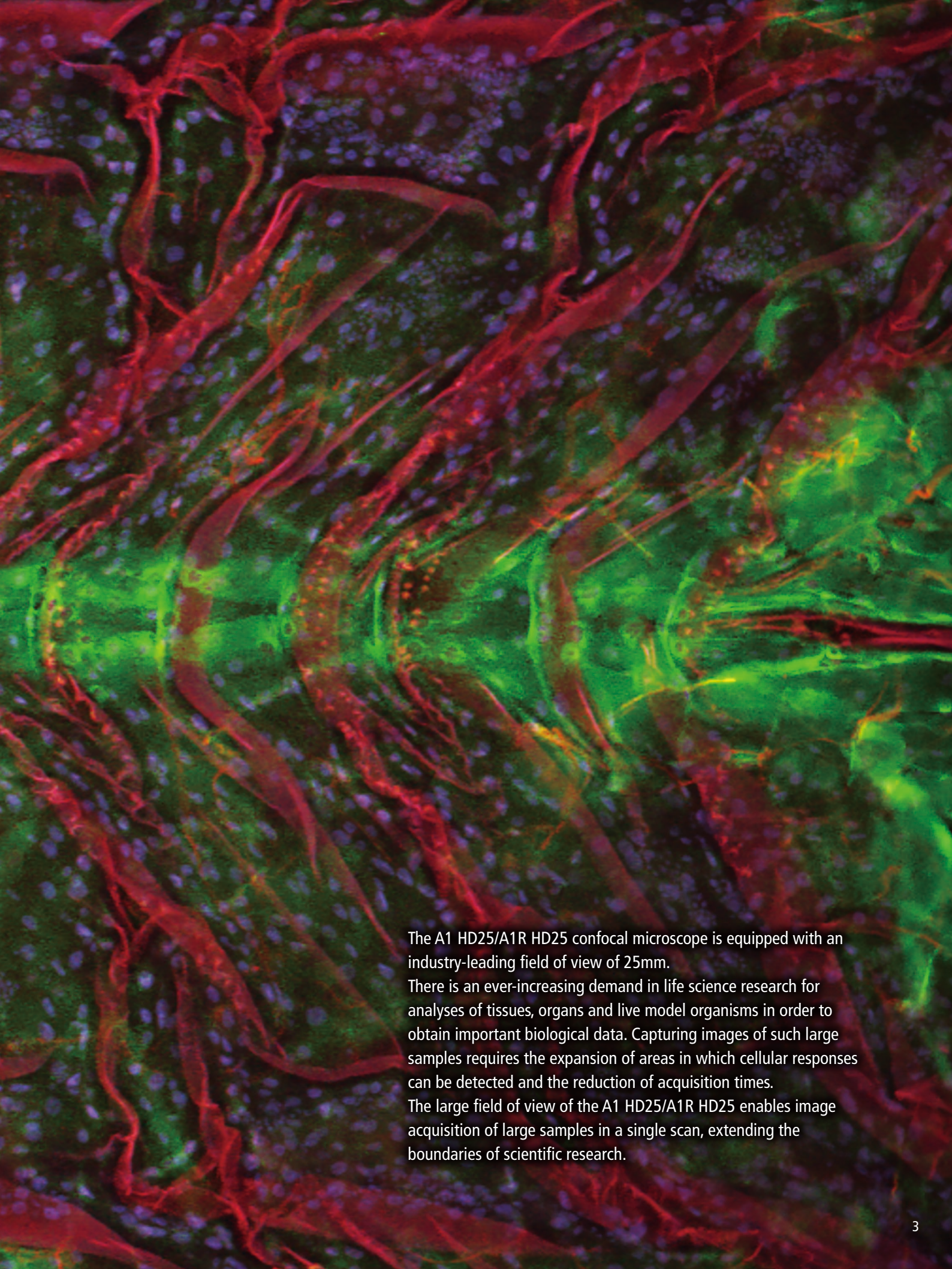
A1 HD25
A1 R HD25

Confocal Microscope

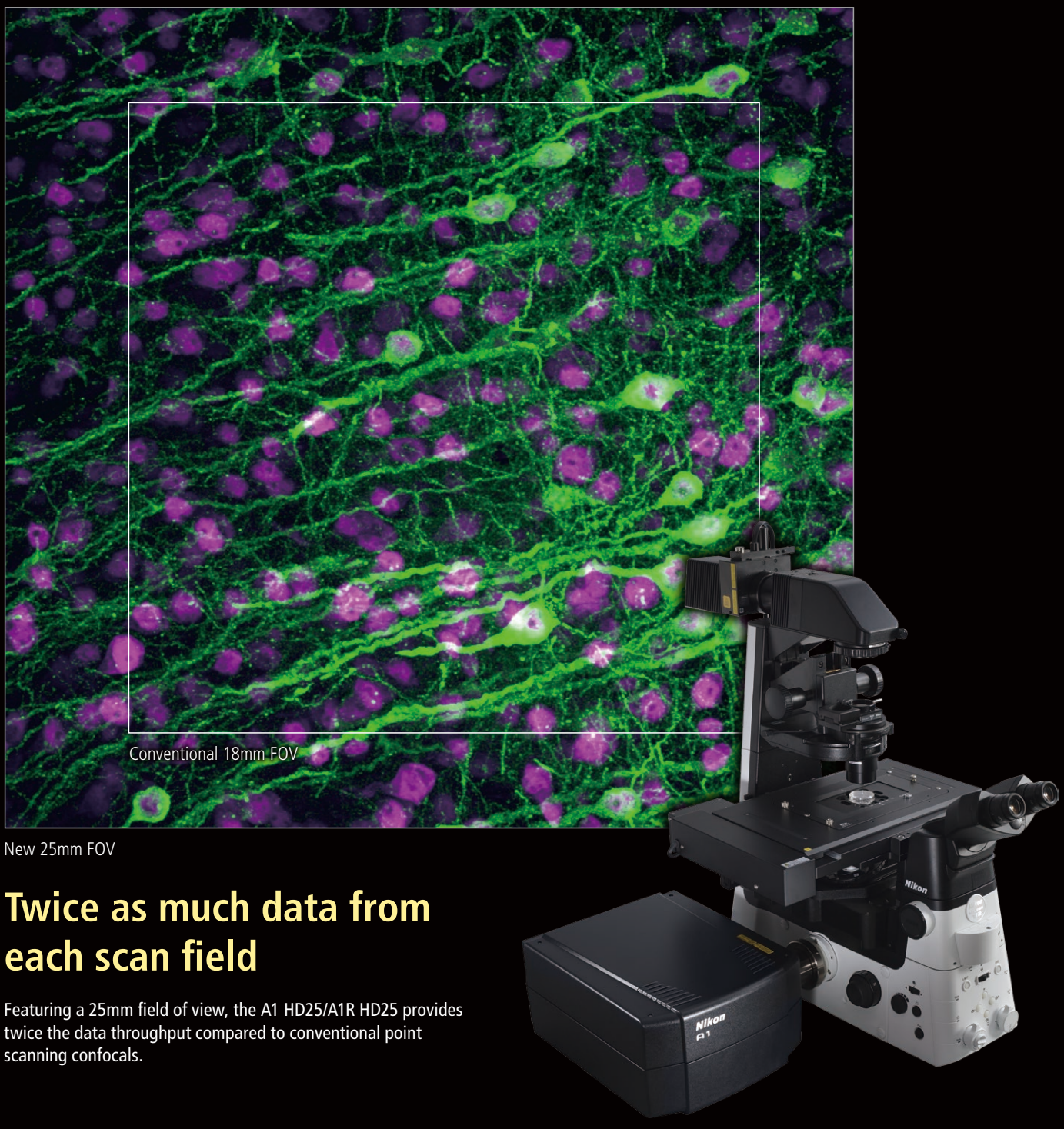
Shedding New Light On **MICROSCOPY**

See the bigger picture



The A1 HD25/A1R HD25 confocal microscope is equipped with an industry-leading field of view of 25mm.
There is an ever-increasing demand in life science research for analyses of tissues, organs and live model organisms in order to obtain important biological data. Capturing images of such large samples requires the expansion of areas in which cellular responses can be detected and the reduction of acquisition times.
The large field of view of the A1 HD25/A1R HD25 enables image acquisition of large samples in a single scan, extending the boundaries of scientific research.

Gain More Resolution

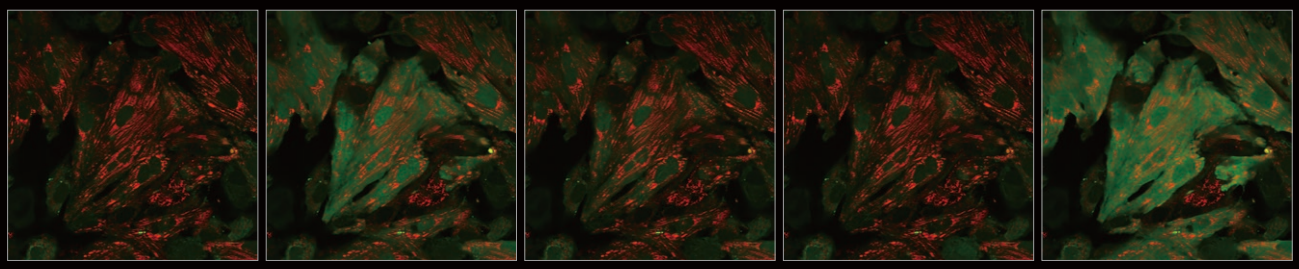


Twice as much data from each scan field

Featuring a 25mm field of view, the A1 HD25/A1R HD25 provides twice the data throughput compared to conventional point scanning confocals.

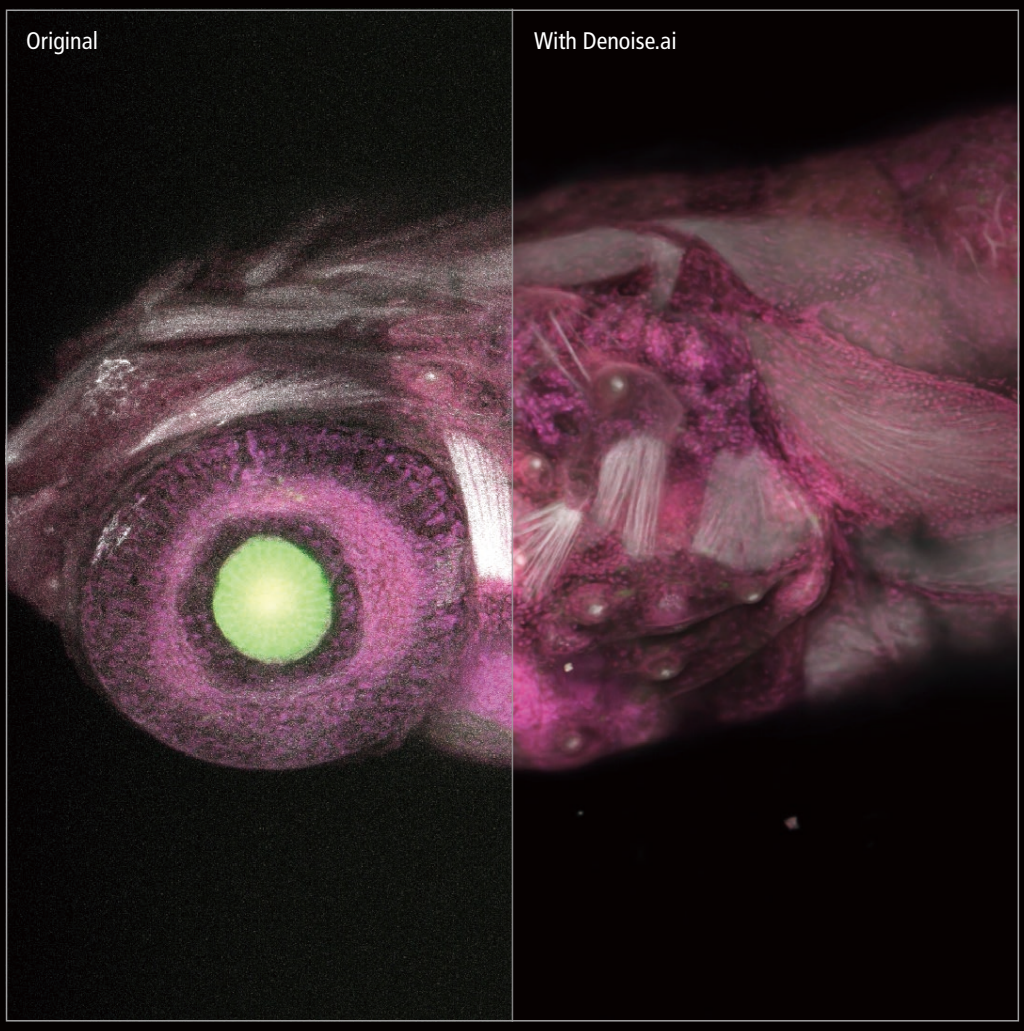
High-speed imaging with short exposure time

The A1R HD25 provides high speed imaging capability up to 720 fps, reducing phototoxicity even during extended periods of time-lapse imaging.



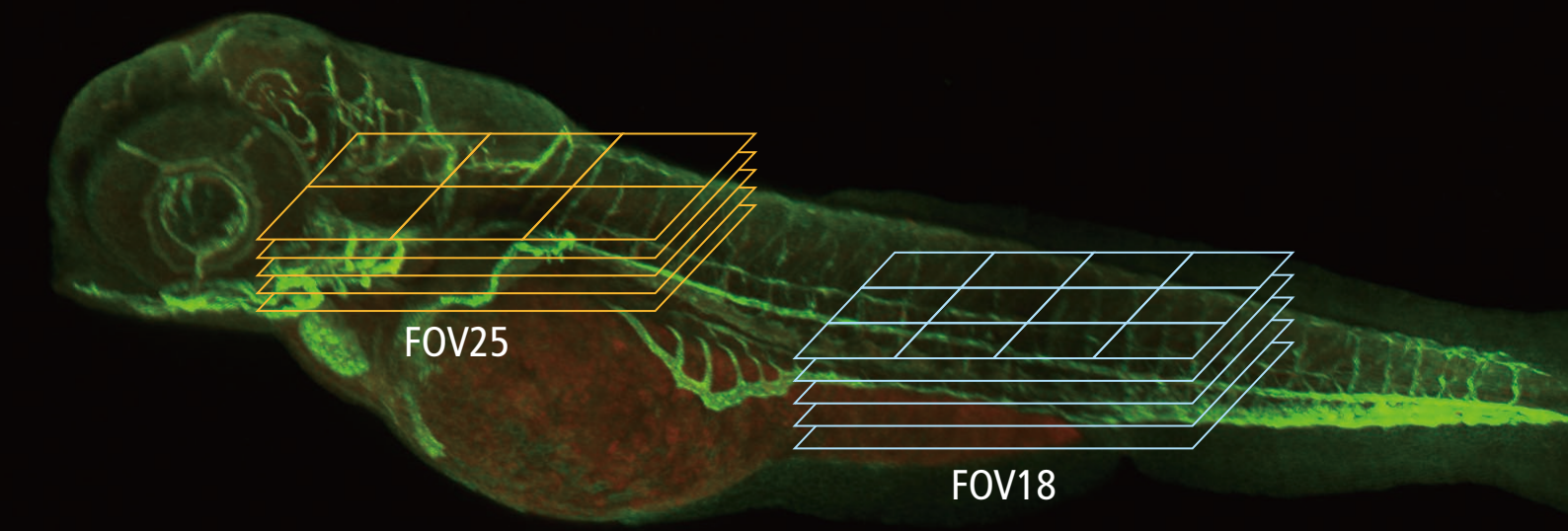
Noise-free imaging

Denoise.ai removes shot noise from resonant confocal images, enabling fast imaging speeds with high signal-to-noise ratios.



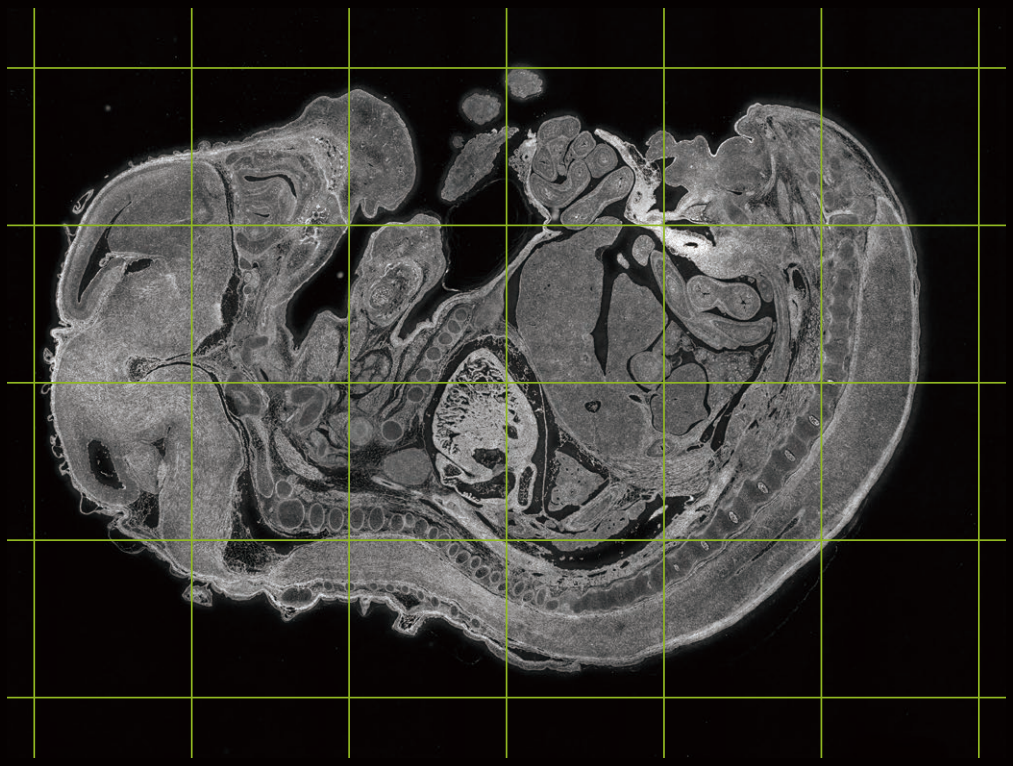
Fewer total images required for large high-resolution image stitching

Together with the Ti2-E inverted microscope, the A1 HD25/A1R HD25 is capable of high-quality 25 mm FOV images that capture nearly twice the imaging area of conventional point scanners in each stage position, enabling the acquisition of more spatial information in a single image than ever before.
The large FOV reduces both the required number of images for stitching large images and image acquisition time, enabling efficient and high-throughput imaging even with large samples such as live model organisms, tissues and organs.

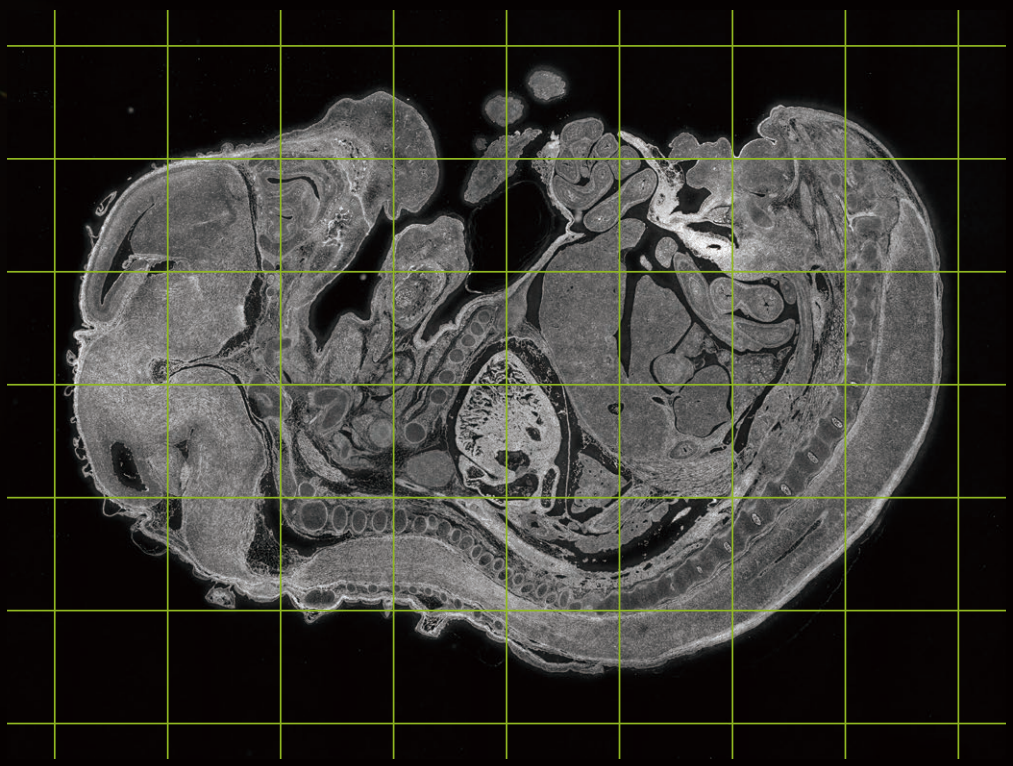


The large FOV can greatly reduce the number of images that are required, in particular for 3D (XYZ) large image stitching.
Total number of images with an FOV of 25: 6,600 (66 for XY stitching, 100 for Z stack)
Total number of images with an FOV of 18: 12,000 (120 for XY stitching, 100 for Z stack)

Doubled image area reduces the number of images by half



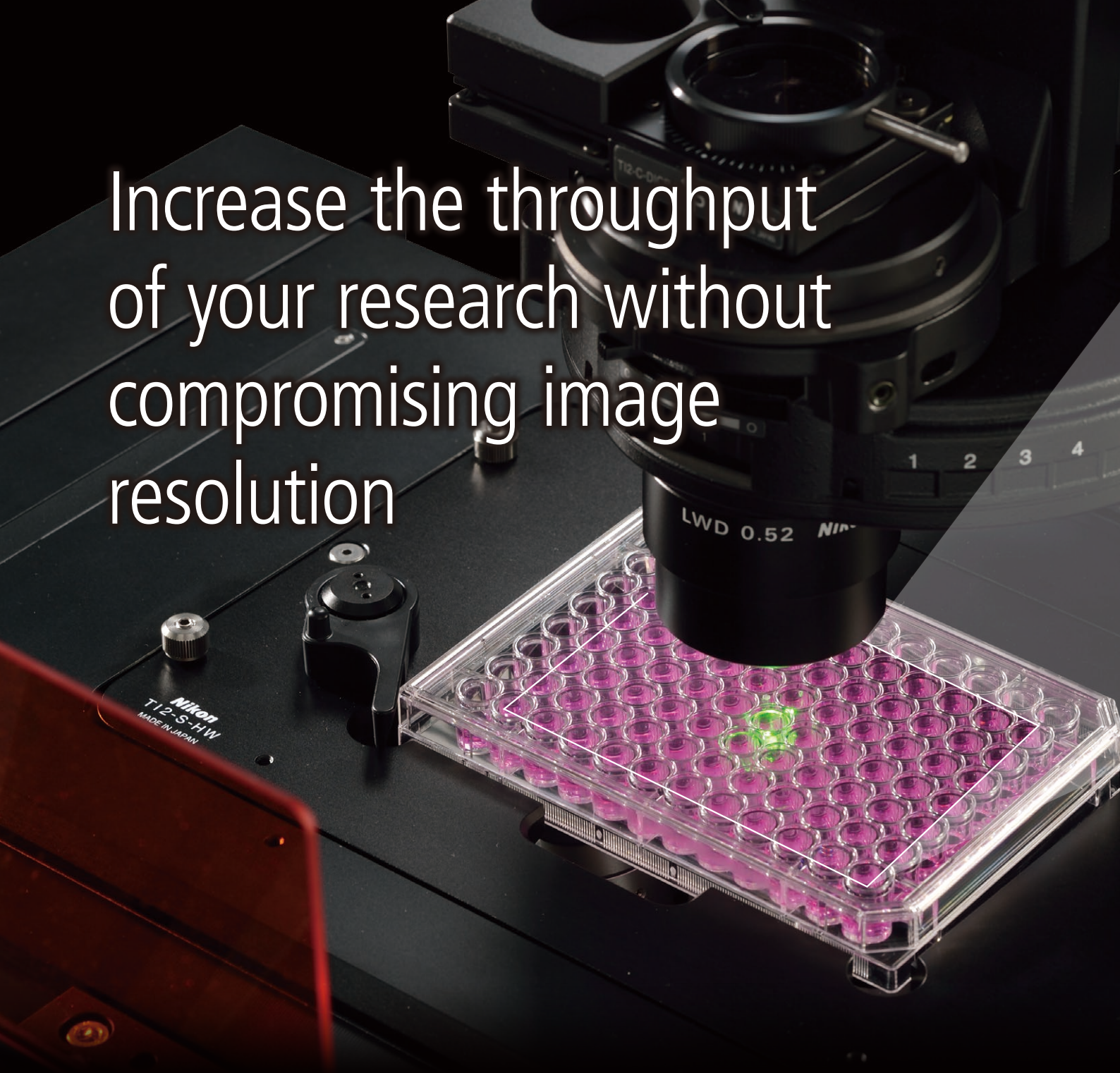
FOV 25 of A1 HD25/A1R HD25: a total of 24 frames



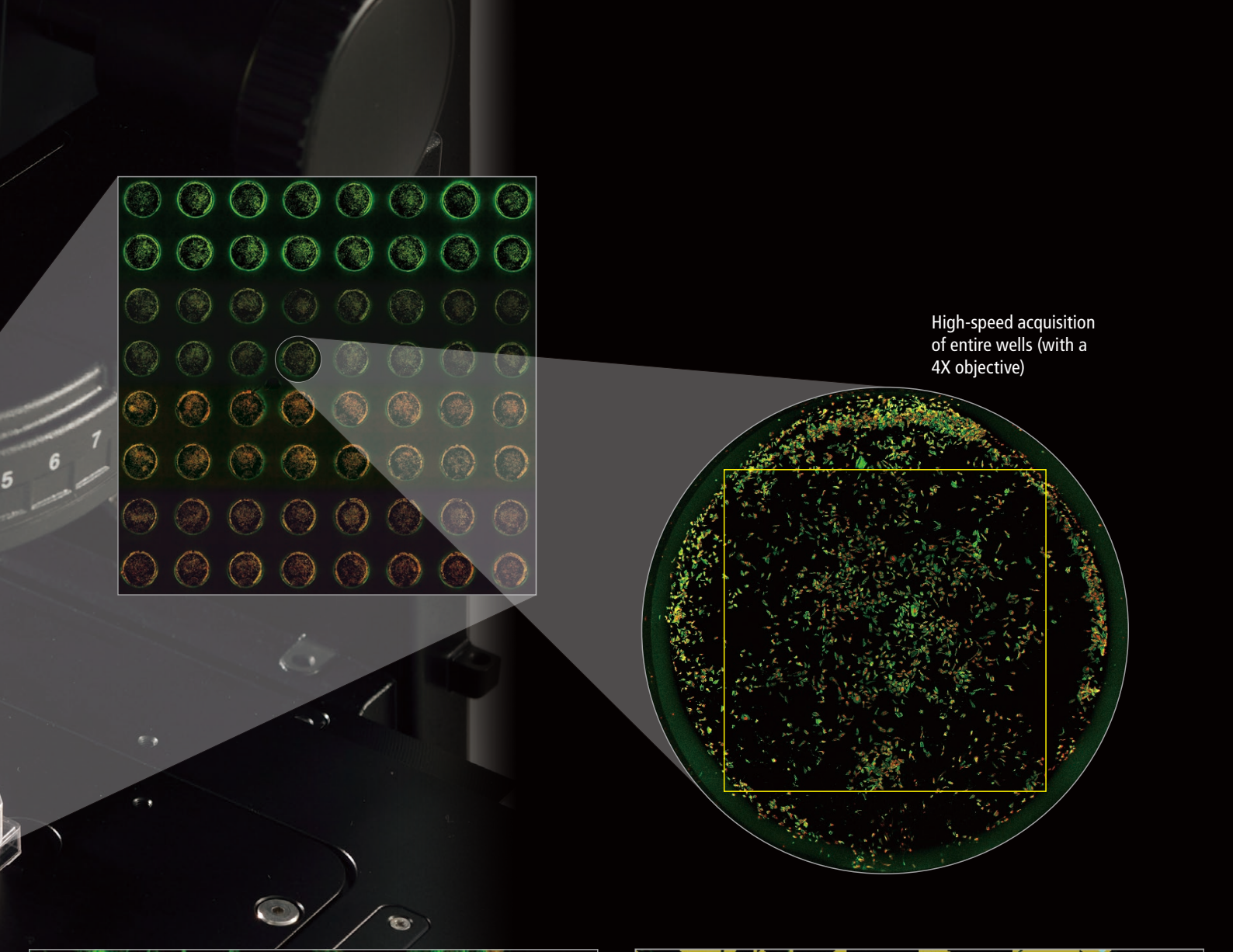
Conventional FOV 18: a total of 48 frames

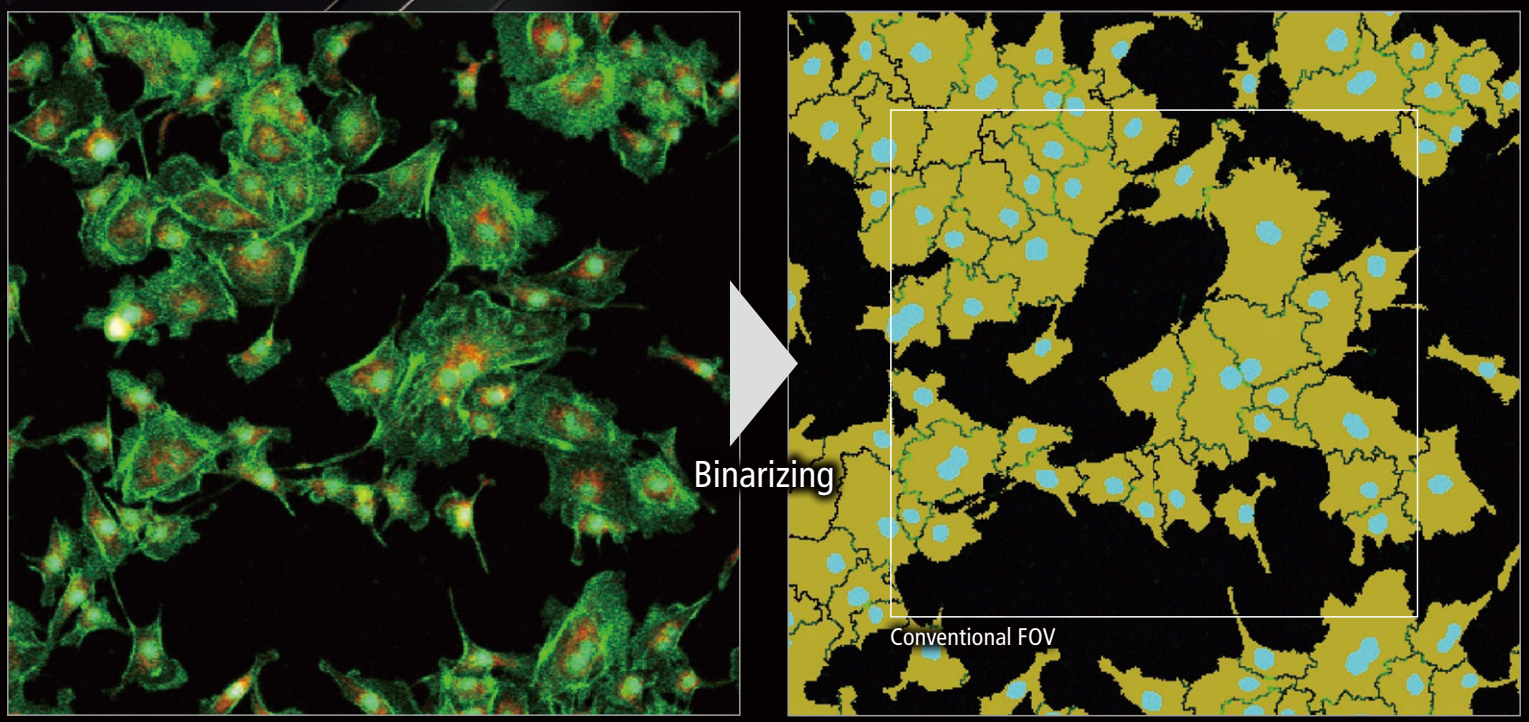
* Images are for illustrative purposes only

Increase the throughput of your research without compromising image resolution



The combination of a high-speed resonant scanner and large field of view is an ideal platform for high resolution screening assays. It dramatically reduces the time needed to analyze multiple samples and conditions.





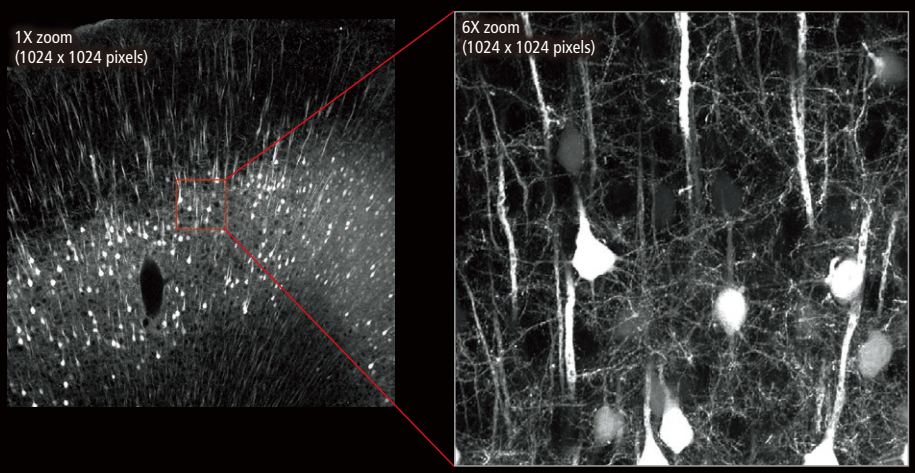
Large FOV enables measurement of larger areas and high-throughput analysis.

Ultrafast resonant scanner

The resonant scanner technologies incorporated in the A1R HD25 produce high-resolution, high-speed imaging at unparalleled levels. The A1R HD25 reduces photobleaching and can acquire the best images for high throughput live cell imaging at high resolutions or multi-dimensional dynamic imaging for applications such as time-lapse and multi-stage position time-lapse experiments.

High definition imaging up to 1K x 1K

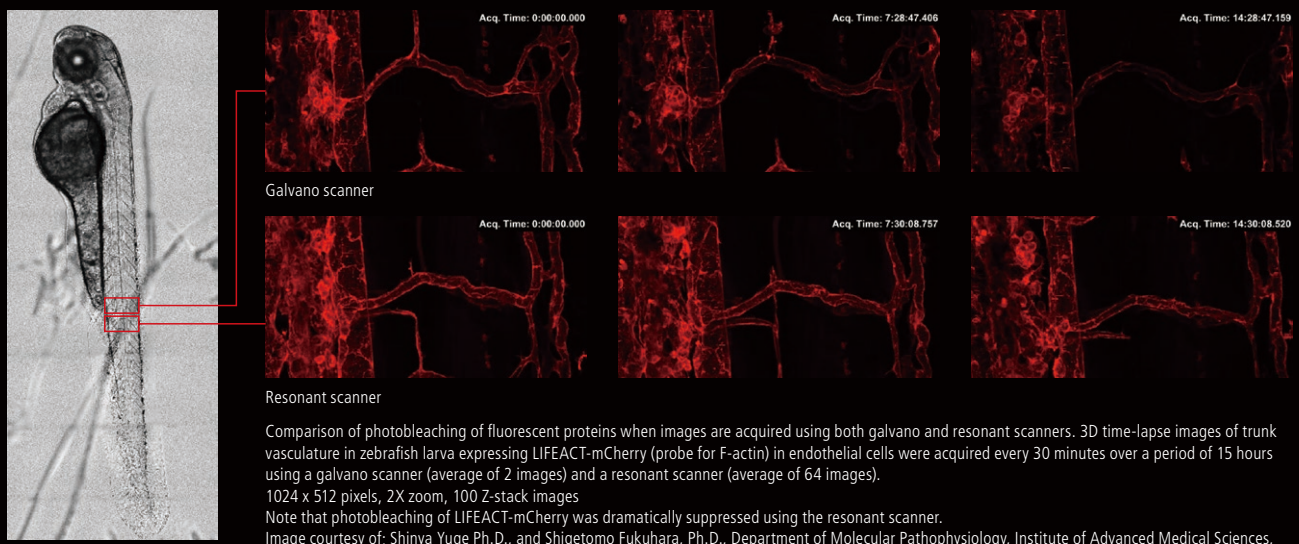
1024 x 1024 pixels enables acquisition of high-resolution, high-quality images at lower magnifications, enabling compatibility with a wide range of samples.



Comparison of a large FOV image and 6X zoomed image (1024 x 1024 pixels) of fine structures in a 2 mm brain slice of H-line mouse cleared with RapiClear1.52, SunJin Lab. Image courtesy of: Drs. Ryosuke Kawakami, Kohei Otomo, and Tomomi Nemoto, Research Institute for Electronic Science, Hokkaido University

Low phototoxicity for live cells

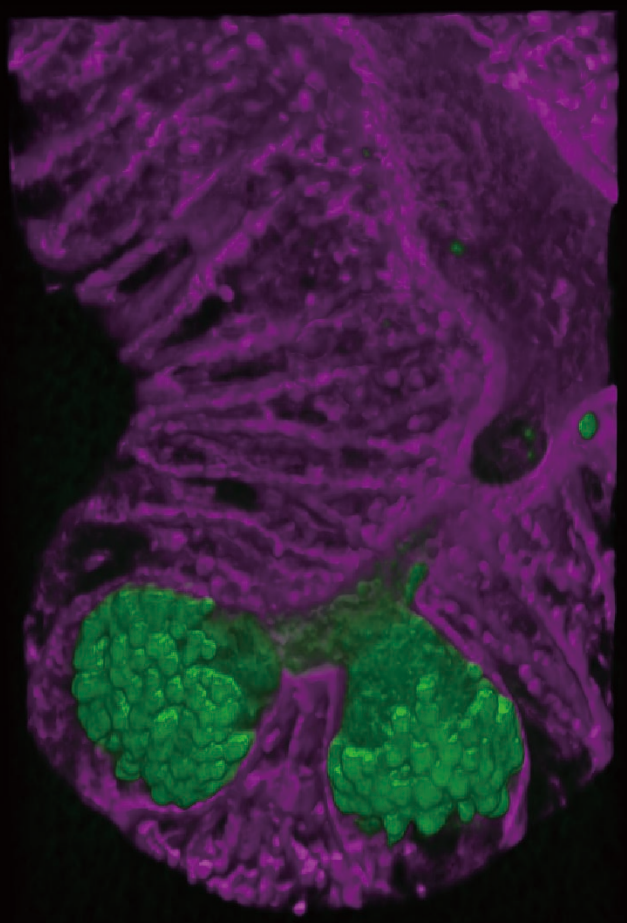
High speed imaging capability up to 720 fps, in combination with a large field of view, dramatically increases imaging throughput. This scanning method reduces the exposure time of the sample to excitation light, minimizing phototoxicity and photobleaching.

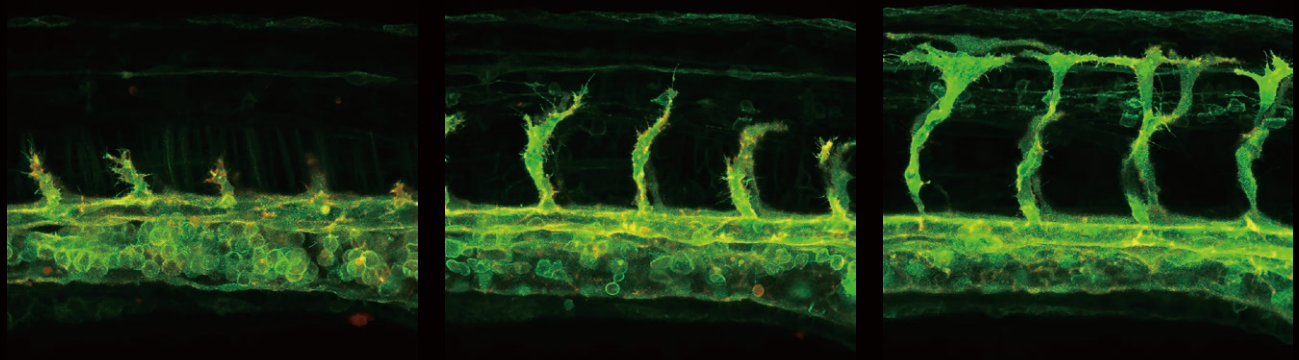


Comparison of photobleaching of fluorescent proteins when images are acquired using both galvano and resonant scanners. 3D time-lapse images of trunk vasculature in zebrafish larva expressing LIFEACT-mCherry (probe for F-actin) in endothelial cells were acquired every 30 minutes over a period of 15 hours using a galvano scanner (average of 2 images) and a resonant scanner (average of 64 images).
1024 x 512 pixels, 2X zoom, 100 Z-stack images
Note that photobleaching of LIFEACT-mCherry was dramatically suppressed using the resonant scanner.
Image courtesy of: Shinya Yuge Ph.D., and Shigetomo Fukuhara, Ph.D., Department of Molecular Pathophysiology, Institute of Advanced Medical Sciences, Nippon Medical School

Fast large-volume time-lapse imaging

Secretion of Paneth cell granule in response to carbachol was acquired by high-speed 4D live imaging (acquisition of 61 steps of Z-stack images at 1.98 s/volume using Piezo Z-stage and 1K resonant scanner) using enteroids, the three dimensional culture of intestinal epithelial cells. As the innate immune response, the secretion of Paneth cell granules (green) one by one into enteroid lumen is clearly observed with high-definition 3D time-lapse imaging.
Green: Zinpyr-1 (Paneth cell granules), Purple: CellMask™ Deep Red (plasma membrane)
Excitation wavelength: 488 nm, 638 nm Resolution: 1024 × 512 pixels
Image courtesy of: Dr. Yuki Yokoi, Dr. Kiminori Nakamura, Dr. Tokiyoshi Ayabe, Innate immunity Laboratory, Department of Cell Biological Science, Faculty of Advanced Life Science, Graduate School of Life Science, Hokkaido University

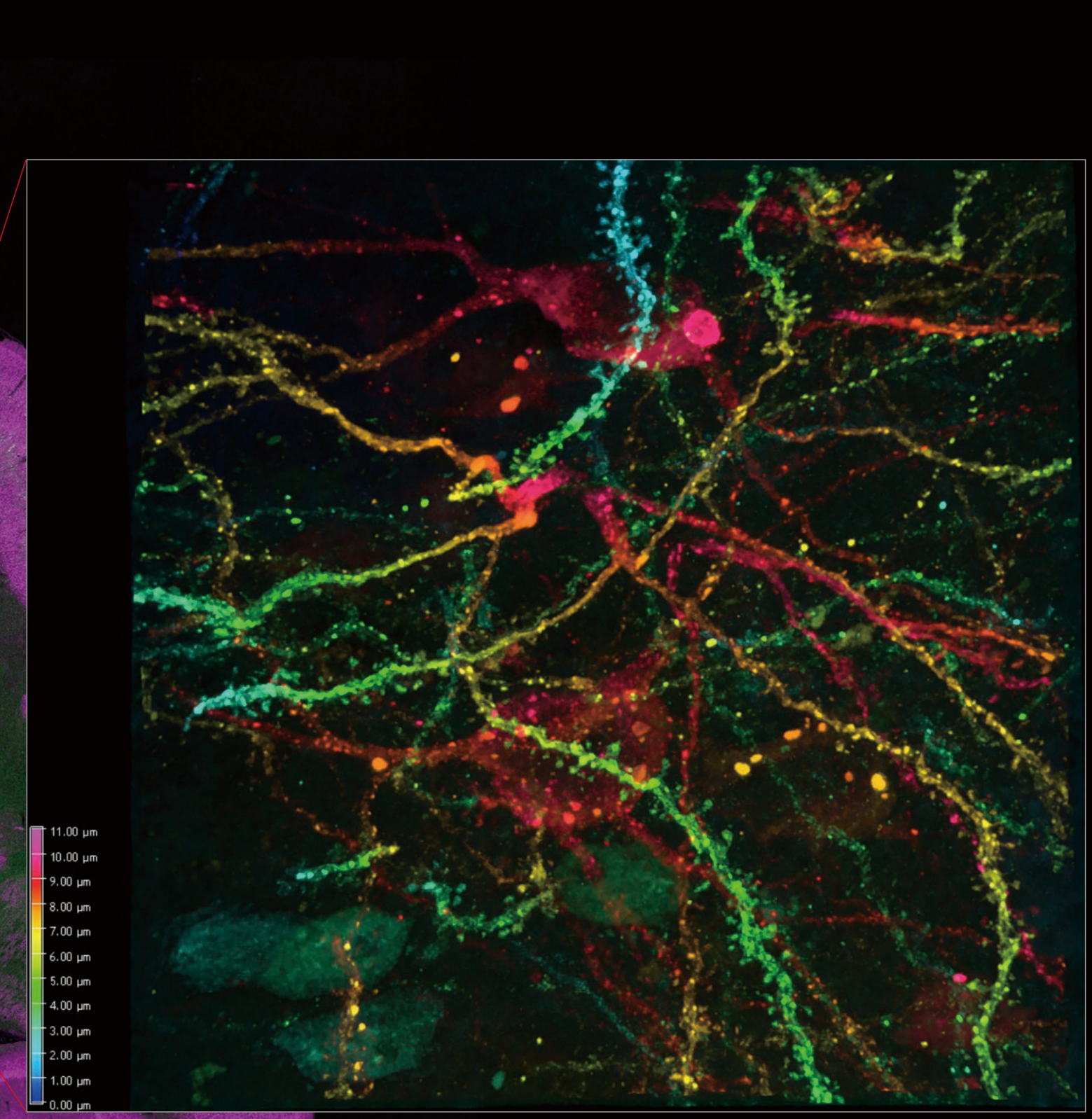




Time-lapse imaging of angiogenesis in zebrafish embryos expressing LIFEACT-mCherry (probe for F-actin) and MYR-GFP (probe for plasma membrane) in endothelial cells. 3D time-lapse images were acquired every 2.5 minutes for 14 hours starting from 22 hours post-fertilization using a resonant scanner (average of 64 images).
1024 x 1024 pixels, 2X zoom, 68 Z-stack images
Note that rapid formation and retraction of endothelial filopodia during angiogenesis has been clearly captured.
Image courtesy of: Shinya Yuge Ph.D., and Shigetomo Fukuhara, Ph.D., Department of Molecular Pathophysiology, Institute of Advanced Medical Sciences, Nippon Medical School

Superior images for both macro and micro imaging

Capture large-scale overview images as well as high magnification images with the same instrument. The 25mm FOV of the A1 HD25/A1R HD25 is effective for observation of large samples, while its 1Kx1K high-definition is ideal for the observation of minute structures.



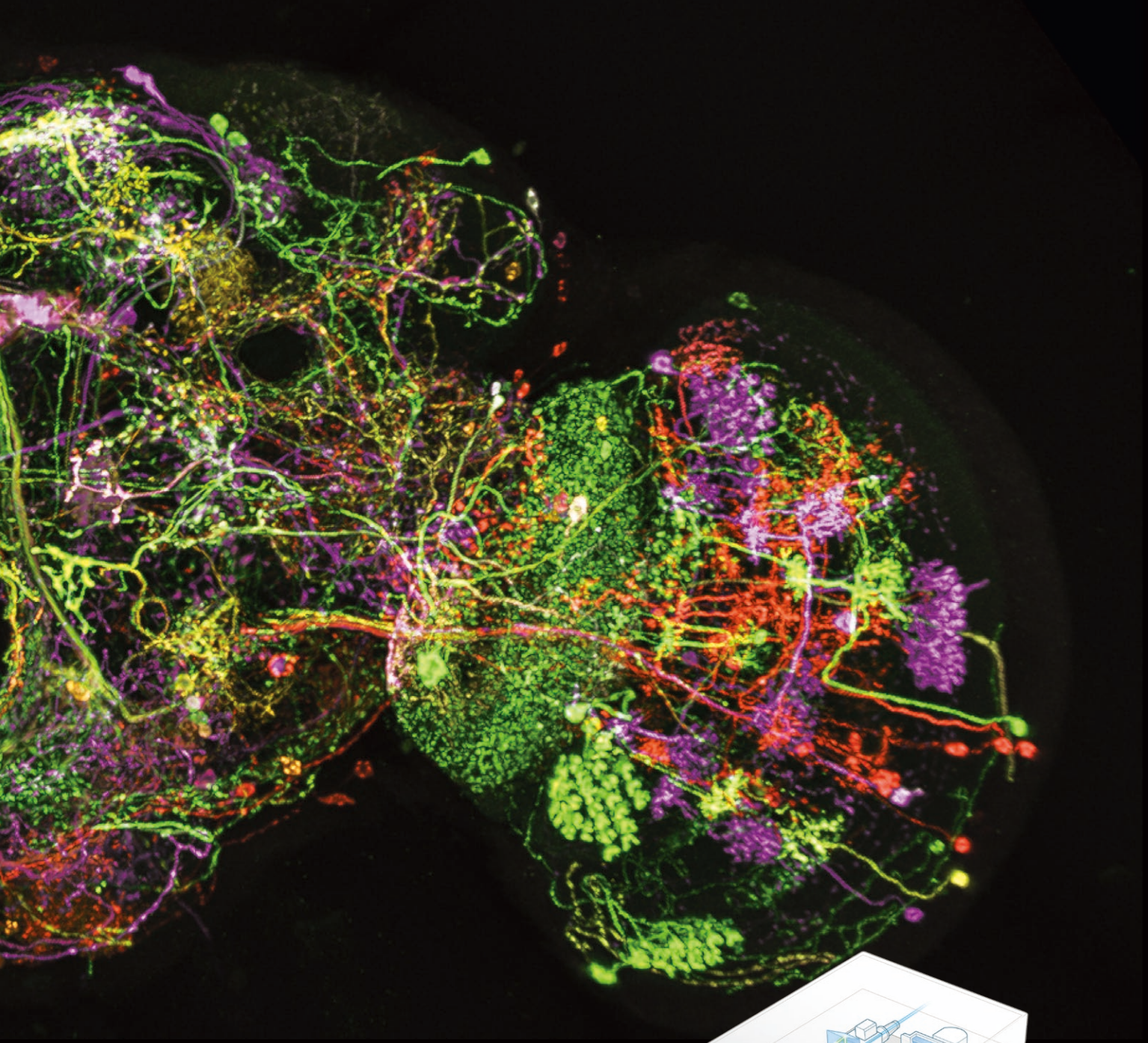
Stitched overview image of marmoset brain captured with CFI Plan Apochromat Lambda 10X objective and detailed image of dendritic spines captured with CFI SR HP Plan Apochromat Lambda S 100XC Sil objective

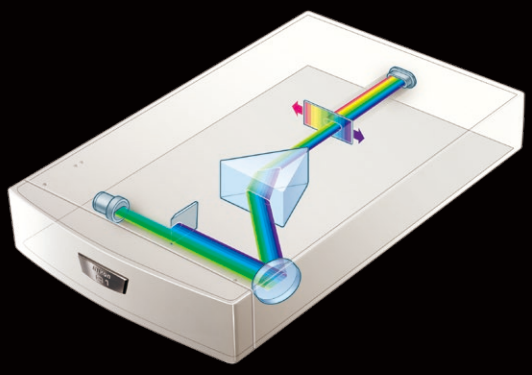
High sensitivity detector options for various types of fluorescence labels

Nikon provides highly sensitive detectors for even low intensity specimens. Various wavelength detection methods that support a wide range of imaging applications are available to suit your individual research needs.

A1-DUG-2 GaAsP Multi Detector Unit

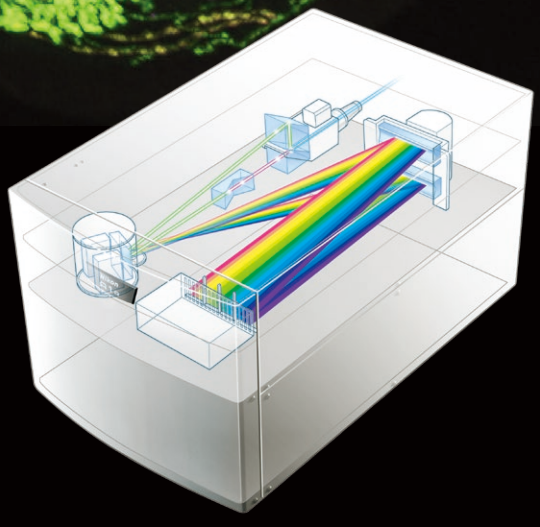
High-sensitivity 4-channel detection





A1-DUVB-2 GaAsP Detector Unit

Flexible detection with high sensitivity



A1-DUS Spectral Detector Unit

Accurate separation of closely overlapping fluorescence spectra and the elimination of autofluorescence

NIS.ai

Artificial Intelligence (AI) for Predictive Imaging, Image segmentation and Processing

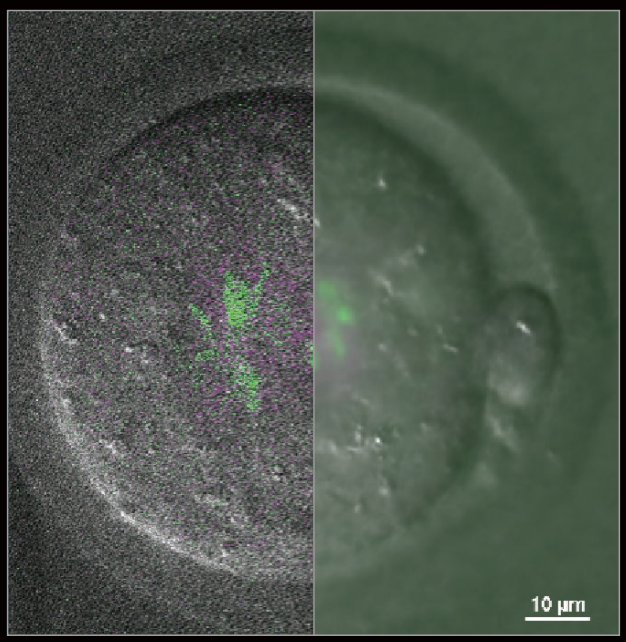
NIS.ai Artificial Intelligence Imaging Software module enables easy and accurate extraction of unbiased data from vast amounts of datasets

Denoise.ai

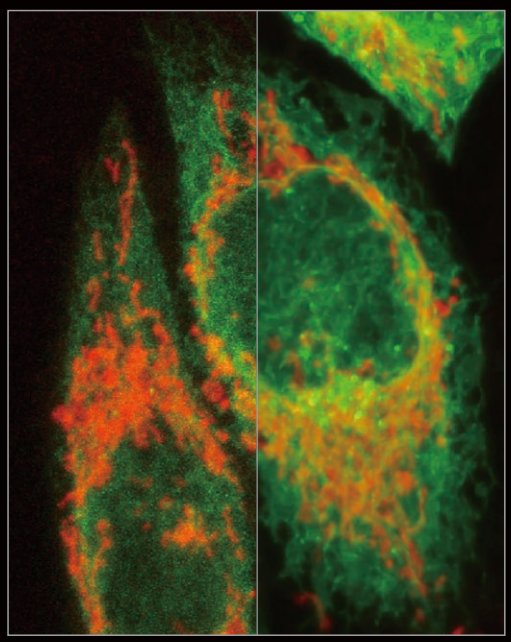
Utilizing *Deep Learning* to remove Poisson shot noise from resonant confocal images

Resonant scanning results in ultra short (tens of nanoseconds) dwell times that are extremely favorable to preventing photo damage and to increasing specimen viability for long term imaging.
While resonant scanning at very short exposure times usually requires line averaging to reduce Poisson shot noise contributions, users can now employ Denoise.ai instead to eliminate the noise contribution. Denoise.ai can recognize and remove the shot noise component of images, increasing clarity and allowing for shorter exposure times and longer time-lapse experiments while maintaining viability.

* Denoise.ai is a standard module of the NIS-Elements C software.



Original With Denoise.ai



Original With Denoise.ai

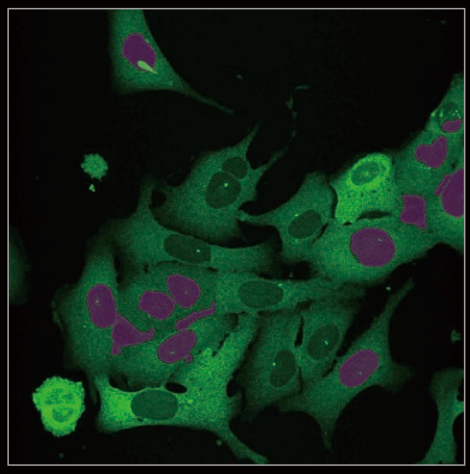
Histone H2B of fertilized mouse egg is labeled in green and MAP4 is labeled in magenta
Excitation wavelength: 488 nm (Laser power 0.1%), 561 nm (Laser power 0.2%)
Resolution: 1024 x 1024 pixels (A1R HD25 resonant scanner)
Objective: CFI Plan Apochromat Lambda S 40XC Sil
Image courtesy of: Dr. Yoshiteru Kai, Reproductive Medicine Research Center, Yamashita Shonan Yume Clinic

Segment.ai

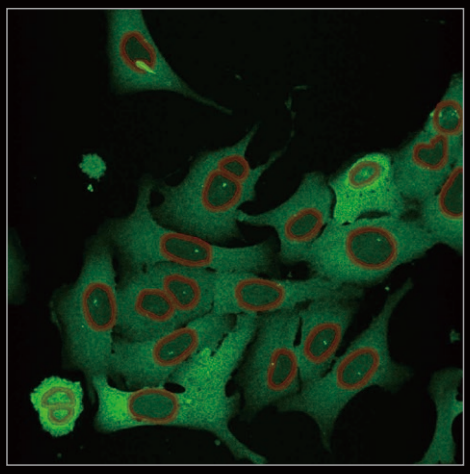
Leveraging AI to automate complex object segmentations that normally require manual tracing methods

Some images are nearly impossible to segment by traditional intensity thresholding methods. Segment.ai trains the neural network to automatically identify and segment complex structures based on a small training dataset that includes manually segmented images. Once trained, Segment.ai can be used to automatically segment large numbers of datasets in an unbiased fashion.

* Segment.ai is an optional module of the NIS-Elements Ar and C softwares.



Conventional Thresholding

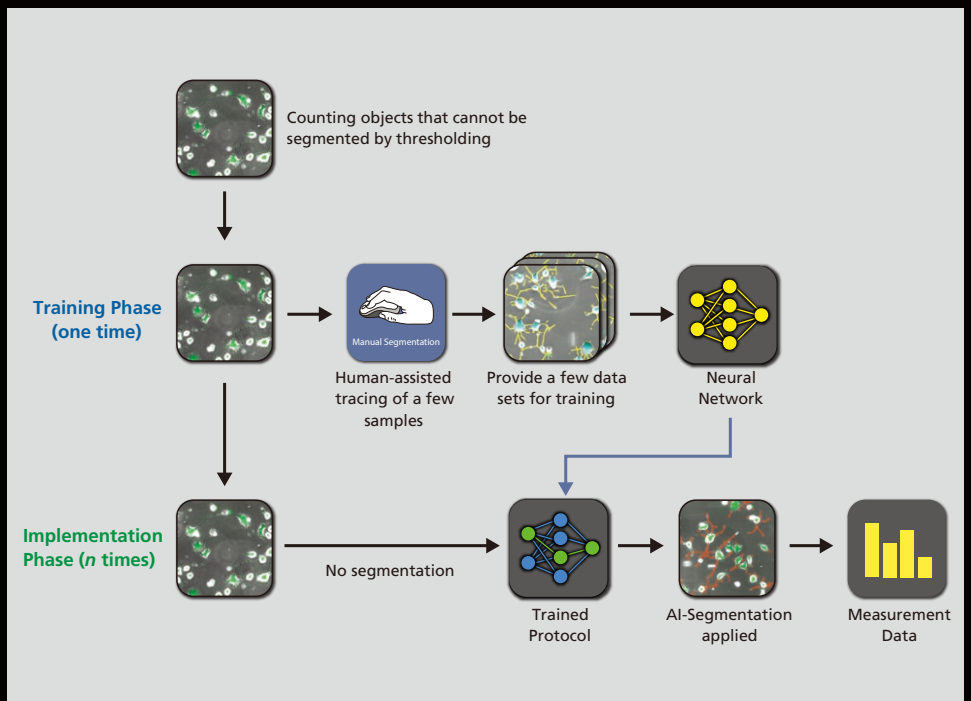


AI Segmentation

The goal was to make intensity measurements along the nuclear envelope of cells. Conventional segmentation could not differentiate the cellular structures and misses several cells. AI-trained segmentation recognizes and identifies the nuclear envelope successfully.

No programming skills required

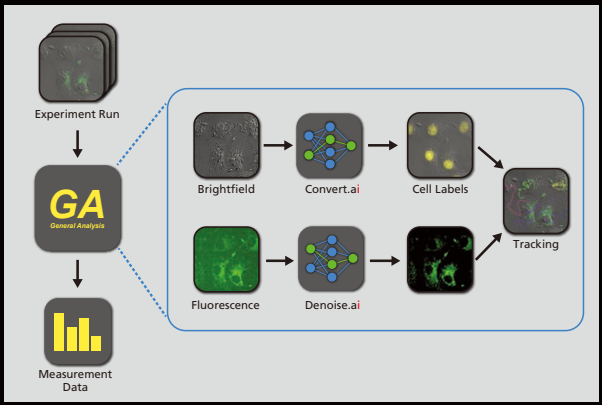
NIS.ai employs convolutional neural networks (CNNs) to learn from labeled training data created by either conventional segmentation or human-assisted tracing of a small subset of representative samples. When using the module, the software interface makes it easy to apply complex deep learning to sample data, eliminating the need to design a complex neural network and apply training data to it. Automated tools take this training data and apply the neural network to recognize patterns. The result training recipe can then be applied repeatedly and reliably to similar samples to process or analyze huge volumes of data at significantly faster speed than traditional techniques.



GA3

An analysis pipeline with AI capabilities

Using NIS-Elements General Analysis (GA3), multiple conventional segmentation and AI tools can be combined to create data measurement routines customized for a specific experiment. These can be applied across multiple images, experiment runs, or high content data.
Because GA3 is freely customizable, it can be adapted to new experiment routines easily. Routines can be embedded as well during experiment acquisition runs.

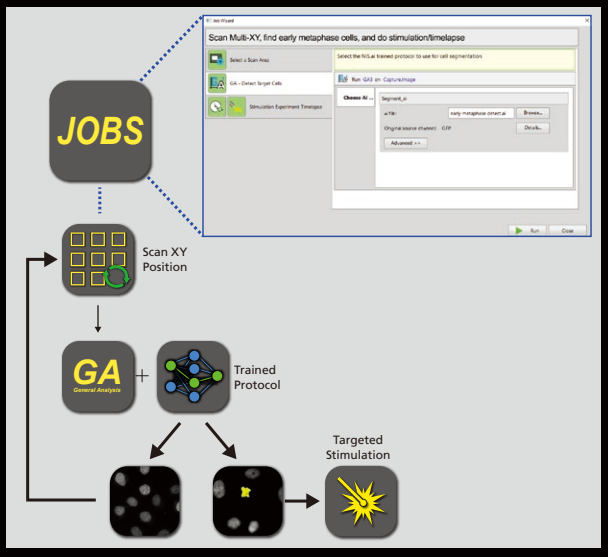


General Analysis can be used to integrate multiple AI tools into the analysis pipeline. For example, Convert.ai can be used to virtually identify nuclei from brightfield images for cell tracking while Denoise.ai can be used to reduce noise from the fluorescence channel.

JOBS

Use NIS.ai as part of an imaging pipeline

NIS.ai tools can be combined with all other features of the NIS-Elements platform to develop imaging protocols and targeted analysis from basic counting through rare event or selective phenotype detection and analysis.
This can be incorporated post-acquisition or, more impactfully, as an integral part of an experimental protocol so that NIS-Elements Intelligent Acquisition analysis results obtained during the experiment run can guide the experimental parameters in different directions. Using the JOBS experiment wizard, customized experiments with embedded analysis tasks and branches based on analysis results can be created, allowing for higher throughput and more targeted acquisitions.



Example of utilizing Segment.ai in an experiment run to analyze XY positions as they are captured, and to search for specific phenotypes. When a target cell is found, a stimulation experiment is performed. If no target cell is found, the experiment proceeds to the next XY position.

NIS-Elements Imaging Software Modules

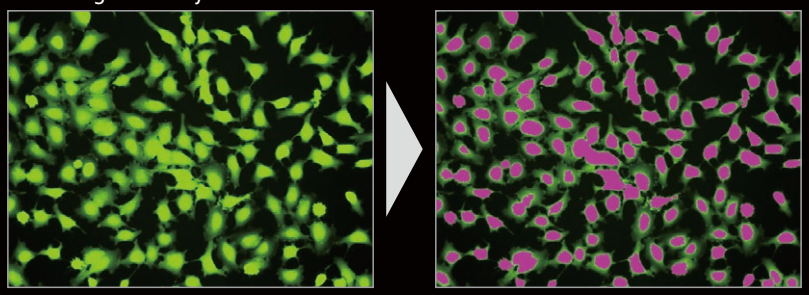
HC
High Content Analysis

High Content Analysis

HC offers fully automated acquisition and analysis of a large number of high-content, multi-dimensional images following an easy stepwise workflow. This enables quick experimental setups and provides an immediate view of measurement data per well during image acquisition via a heat map for trend observation and further analysis.

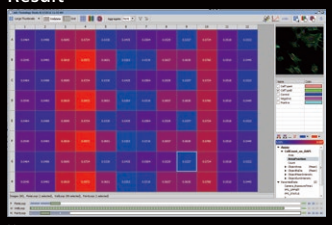
Acquisition

Processing and Analysis

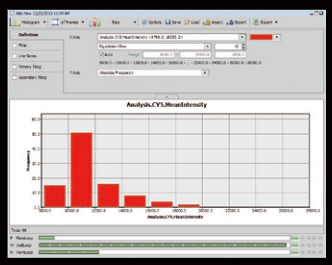


Light intensity analysis

Result



Heat map



Graph

ER
Enhanced Resolution

Confocal resolution enhancement

The NIS-Elements ER module increases conventional confocal resolution by up to two-fold, providing up to 120nm resolution in XY and 300nm in Z. Higher resolution confocal images can be easily generated with a single click. The software assesses the captured image and automatically determines processing parameters to achieve enhanced resolution.

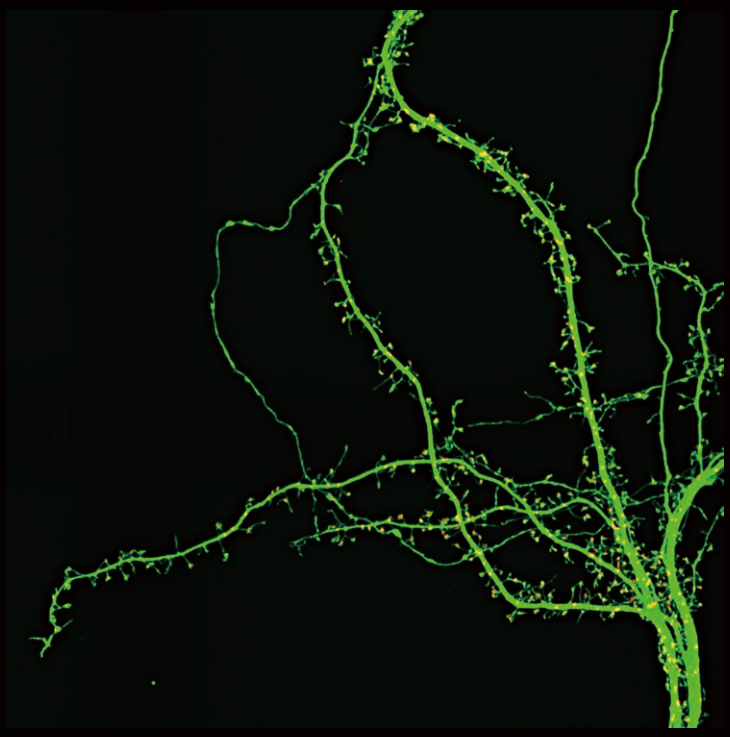


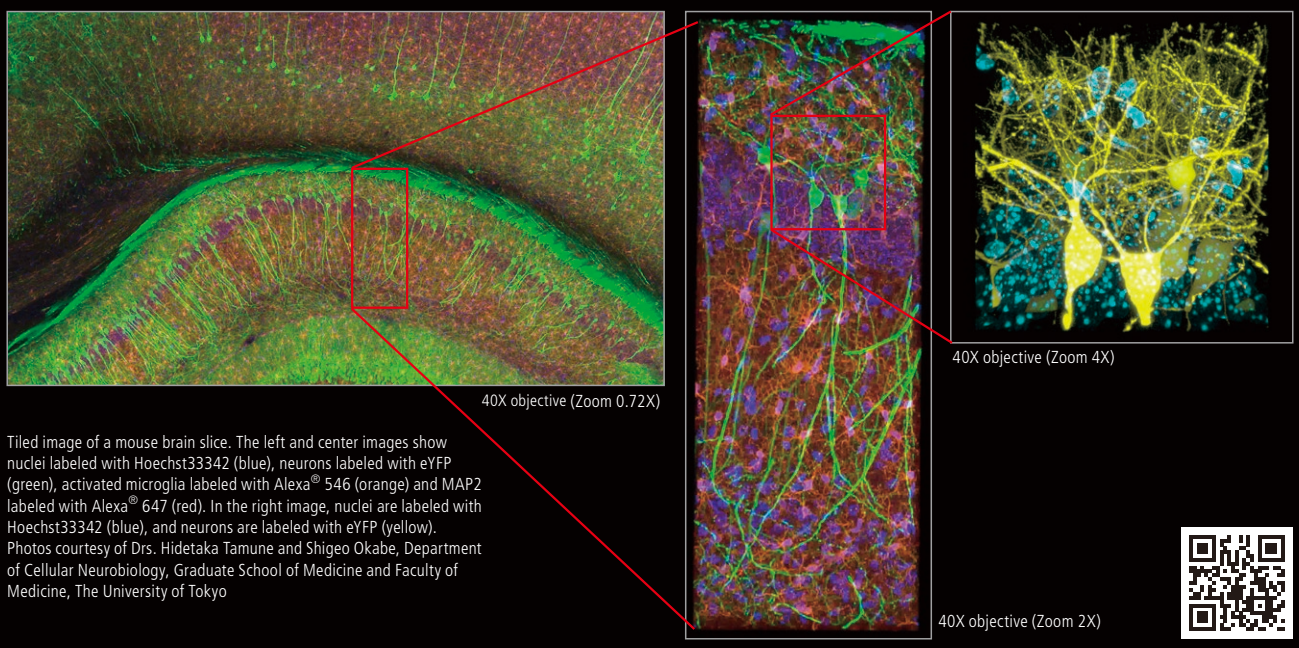
Image courtesy of: Drs. Yutaro Kashiwagi and Shigeo Okabe, Department of Cellular Neurobiology, Graduate School of Medicine and Faculty of Medicine, The University of Tokyo.

Superior optical technologies to support all confocal applications

Nikon provides a broad range of high-NA objectives with unrivaled optical quality to redefine the boundaries of confocal imaging. Options include silicone oil immersion objectives for thick live cell imaging, large-FOV low-magnification objectives and easy-to-use dry objectives. Chromatic aberrations are corrected from ultraviolet to near infrared range, enabling excellent multicolor imaging.

CFI Plan Apochromat Lambda S 40XC Sil

This objective is designed to be used with silicone oil as the immersion medium. Silicone oil has a refractive index that closely matches that of the interior of live cells, thus minimizing spherical aberration when imaging live cells or thicker specimen. Furthermore, silicones oil demonstrates minimal evaporation, an ideal feature for long-term time-lapse imaging.

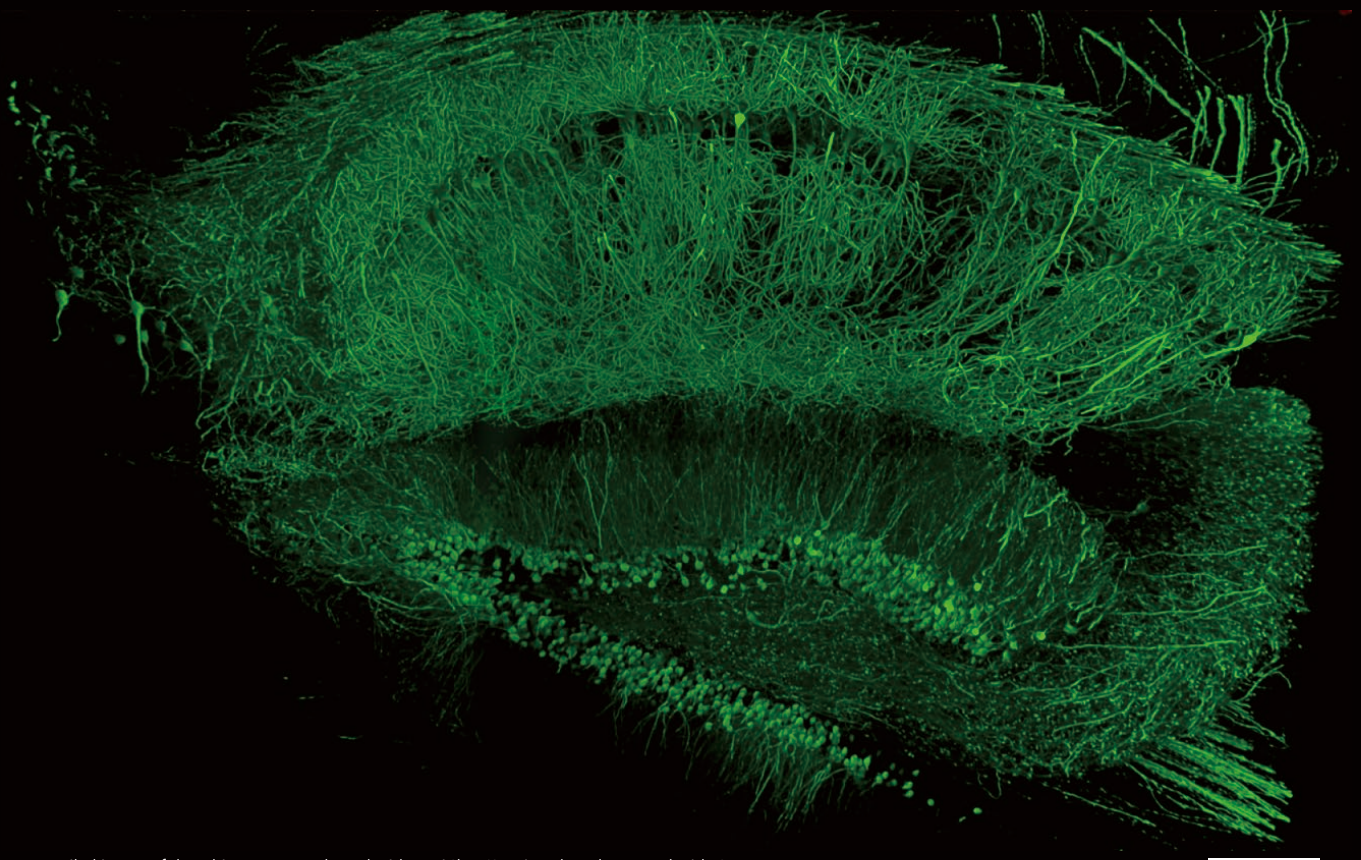


40X objective (Zoom 0.72X)

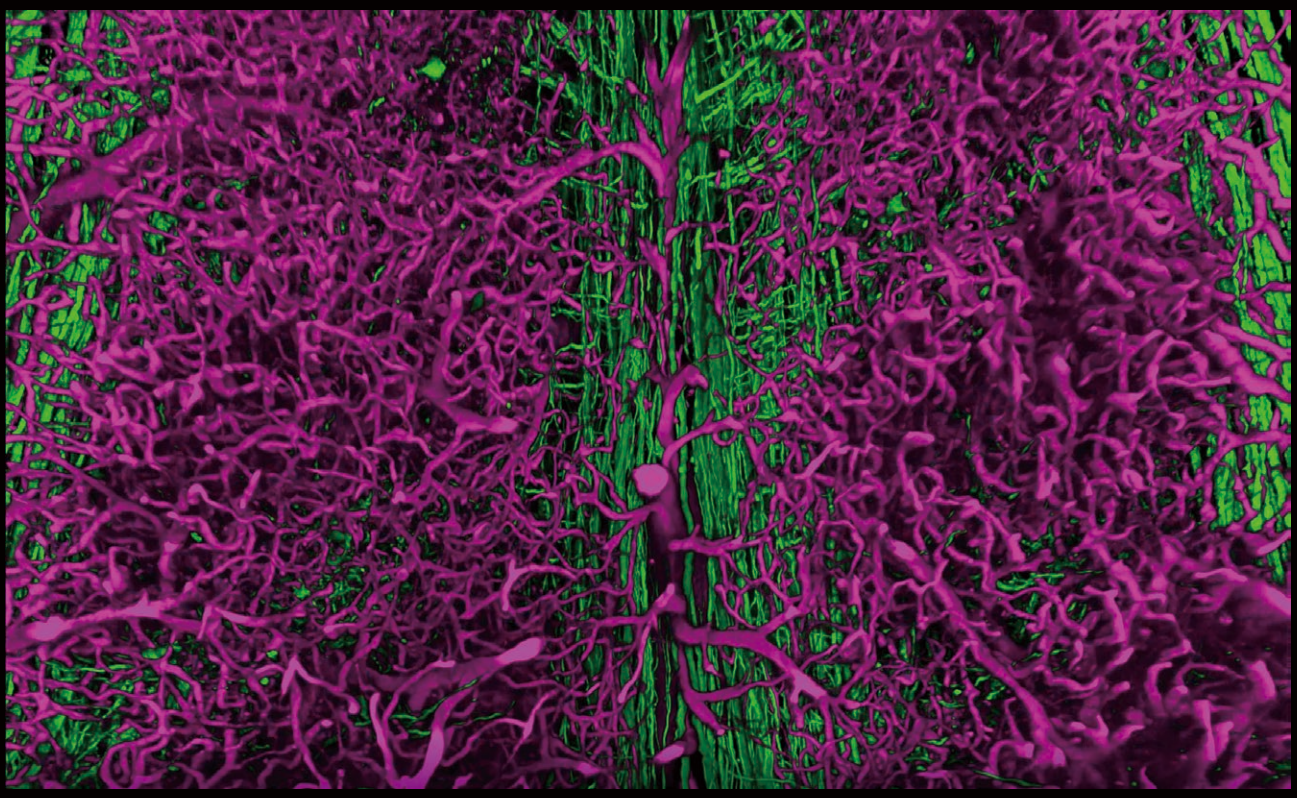
40X objective (Zoom 4X)

40X objective (Zoom 2X)

Tiled image of a mouse brain slice. The left and center images show nuclei labeled with Hoechst33342 (blue), neurons labeled with eYFP (green), activated microglia labeled with Alexa® 546 (orange) and MAP2 labeled with Alexa® 647 (red). In the right image, nuclei are labeled with Hoechst33342 (blue), and neurons are labeled with eYFP (yellow). Photos courtesy of Drs. Hidetaka Tamune and Shigeo Okabe, Department of Cellular Neurobiology, Graduate School of Medicine and Faculty of Medicine, The University of Tokyo



Detailed image of deep hippocampus cleared with RapiClear/SunJin Lab and captured with CFI Apochromat LWD Lambda S 20XC WI



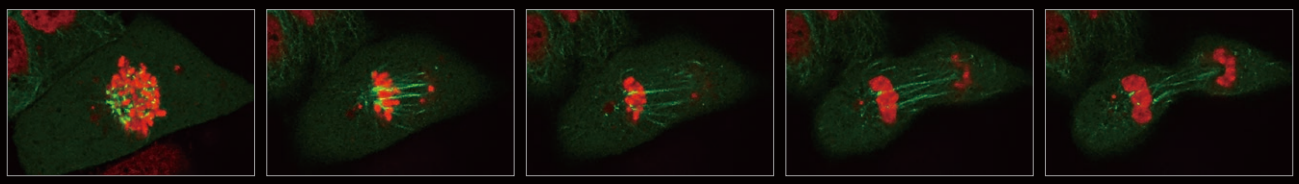
Neural circuit (green) and blood vessel (magenta) of spinal cord cleared with RapiClear/SunJin Lab captured with CFI Apochromat LWD Lambda S 20XC WI

Optional accessories for live-cell imaging

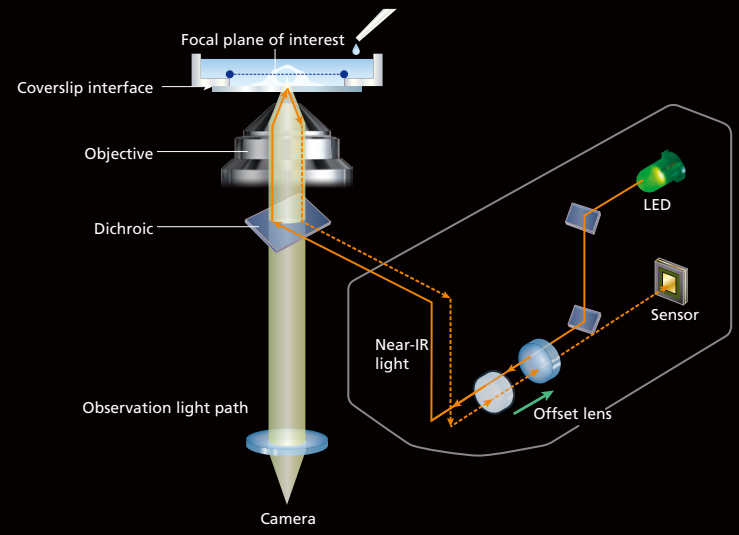
For long-time observation

Perfect Focus System

With the Ti2-E inverted microscopes, the Perfect Focus System (PFS) automatic focus maintaining mechanism can be used. It continuously corrects focus drift during long time-lapse observations or when reagents are added.
*Use with glass bottom dish is recommended.



Consistent focus is maintained during time-lapse imaging over thirty minutes.



Water immersion dispenser

Automatically applies immersion water to the tip of an objective, preventing water from evaporating or overflowing during experiments.

For high-speed 3D image acquisition

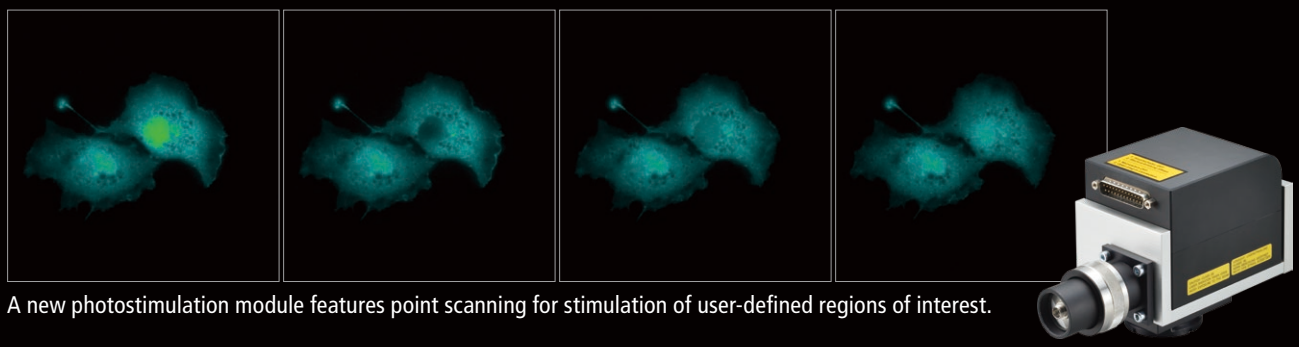
Motorized piezo Z stage

Allows high-speed line Z scanning in combination with a motorized microscope stage.

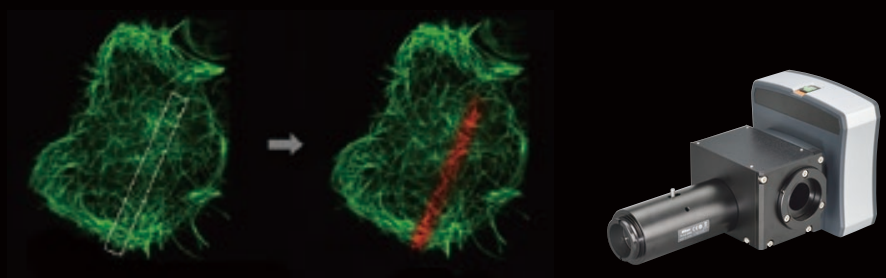
For high-speed live cell imaging during photostimulation

A new point photostimulation module, available for the Ti2-LAPP modular illumination system, allows the A1 HD25/A1R HD25 to acquire confocal images while simultaneously stimulating the desired area of a sample. TIRF module, DMD module, and epi-fluorescence module are also available for the LAPP system.

Simultaneous A1 HD25/A1R HD25 imaging and photostimulation



A new photostimulation module features point scanning for stimulation of user-defined regions of interest.

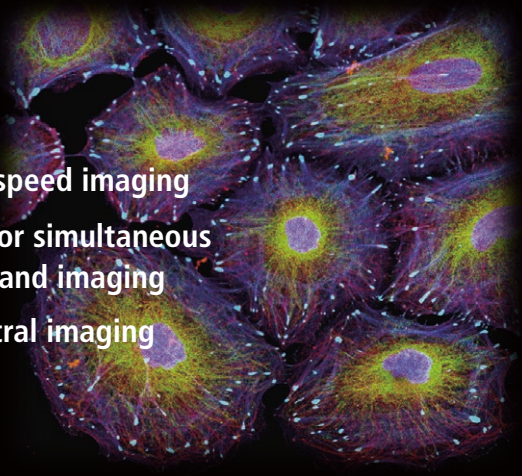


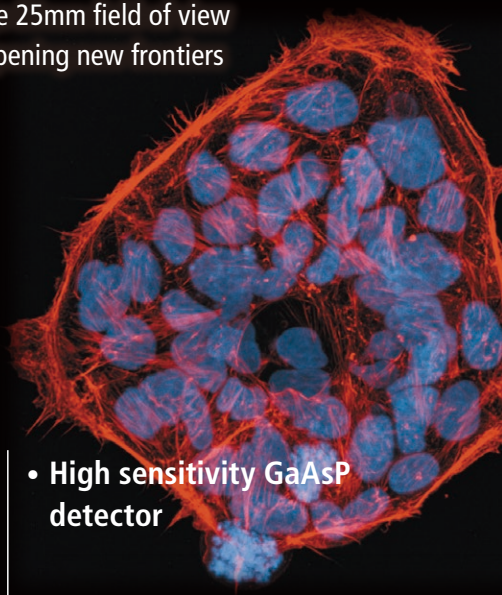
DMD module simultaneously stimulates multiple regions of interest of any user-defined shape.

Various other accessories are available. Please contact Nikon or its distributors in your region

A1ways Evolving

Nikon's capacity to respond to customer requirements has resulted in a continuously updated confocal system that exhibits outstanding performance. A1's latest innovation includes a large 25mm field of view and AI-based tools for enhancing confocal image quality and analysis capabilities, opening new frontiers in research.

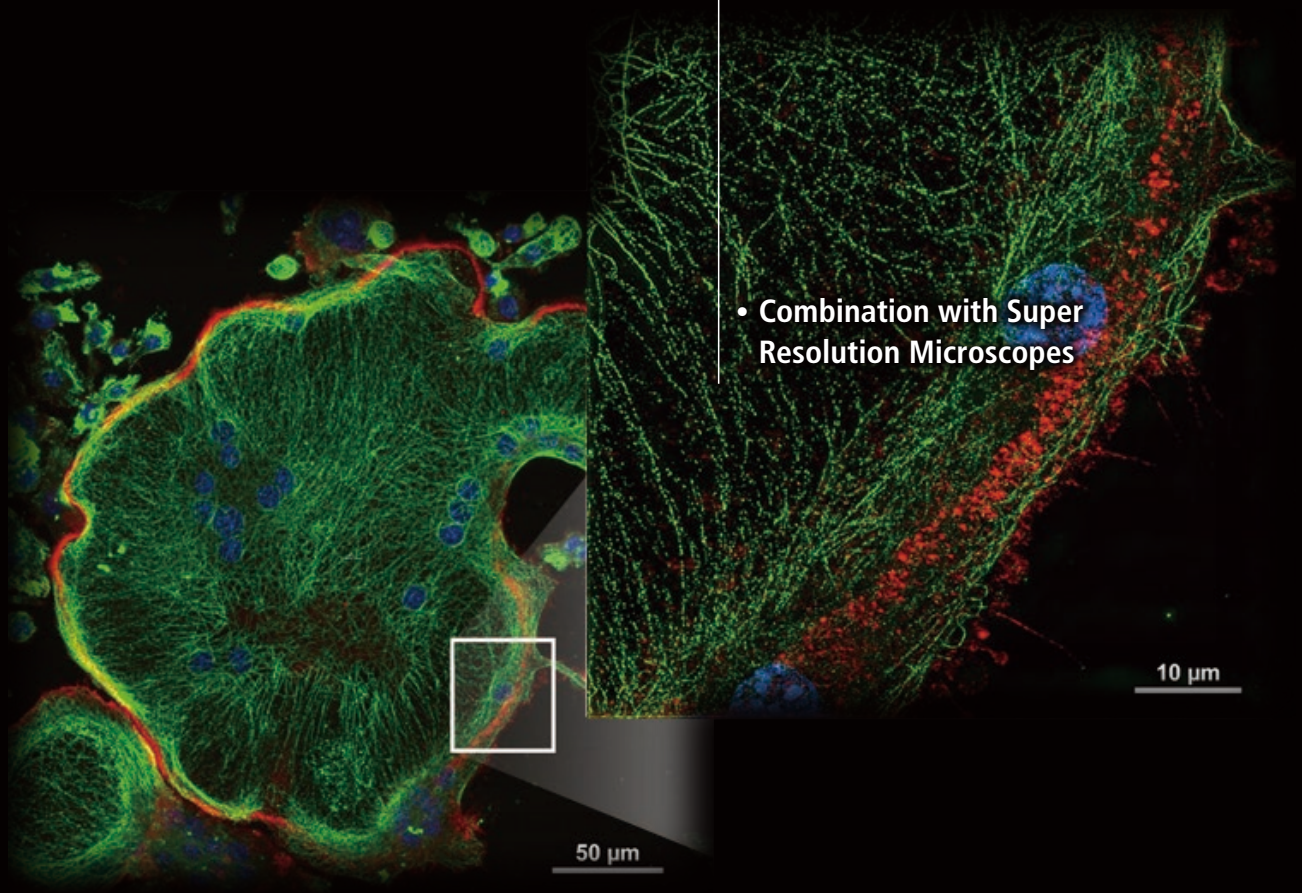




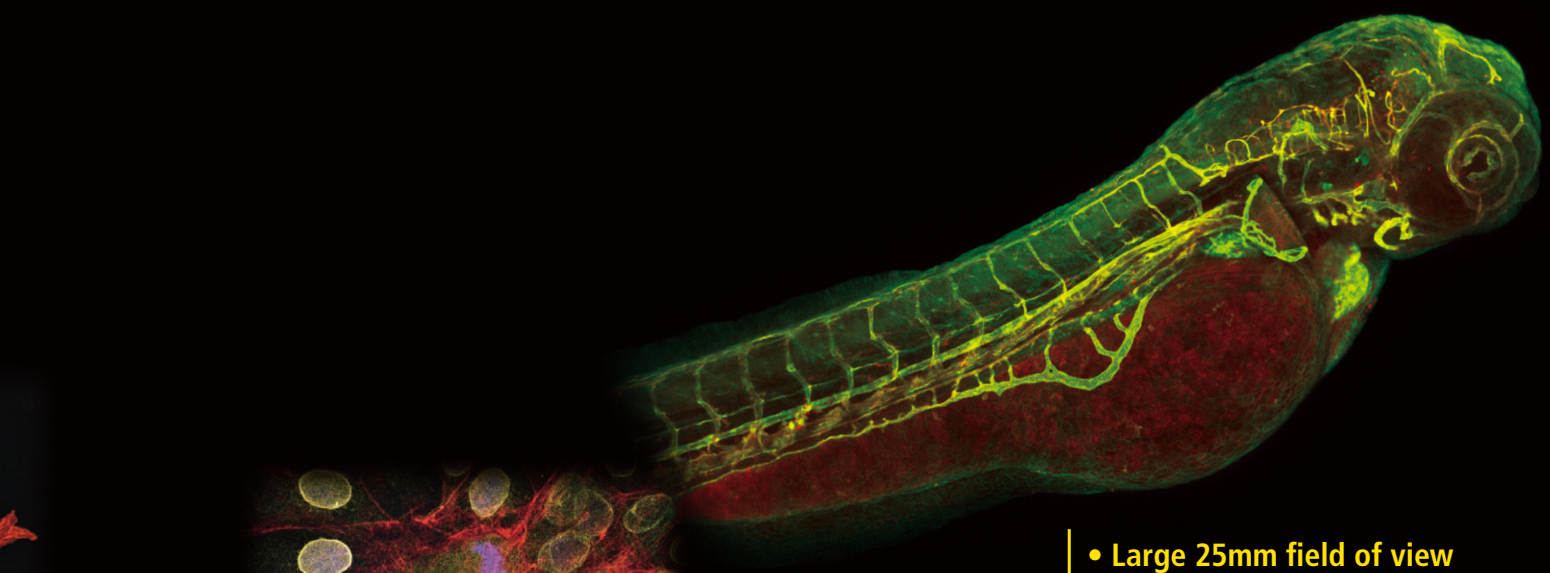
- Video rate high-speed imaging
- Hybrid scanner for simultaneous photoactivation and imaging
- 32-channel spectral imaging

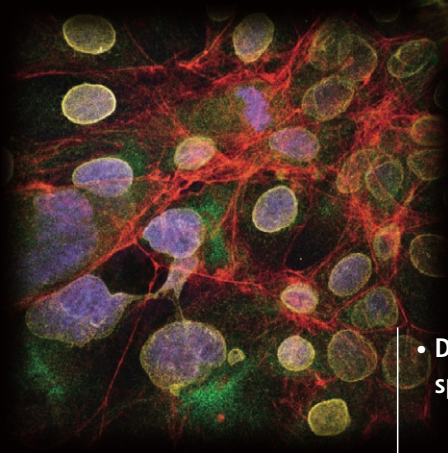
- High sensitivity GaAsP detector

2008 | **2009** | **2010** | **2011** | **2012** | **2013**



- Combination with Super Resolution Microscopes

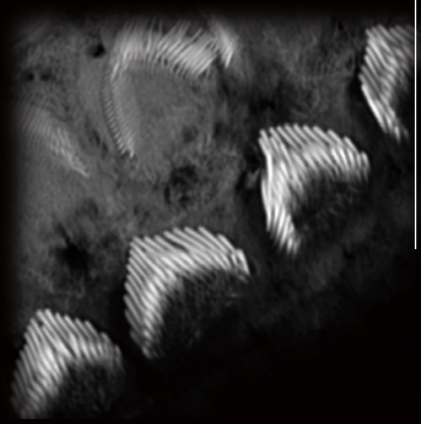




• Large 25mm field of view

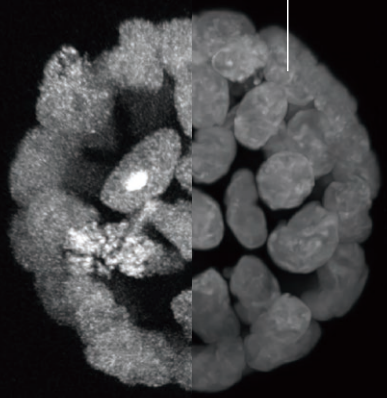
• DUVB filterless compact spectral detector

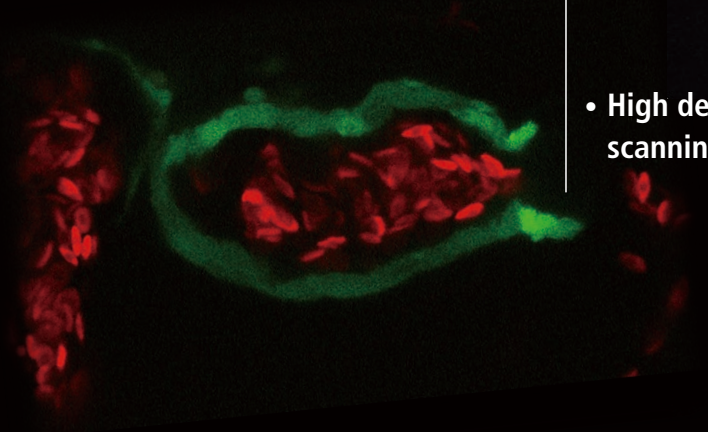
2014 2015 2016 2017 2018 2019



• NIS-Elements ER resolution enhancing software

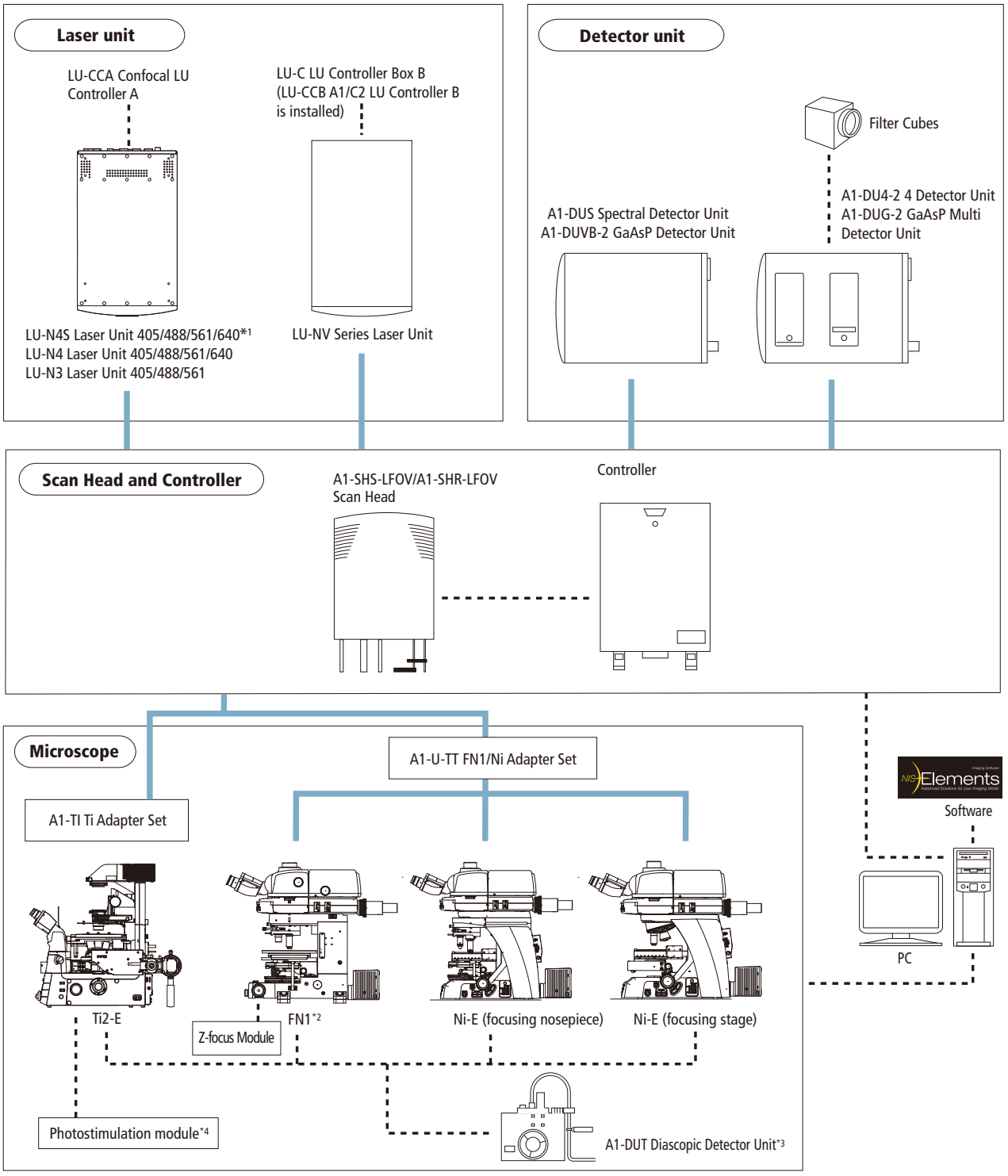
• Denoise.ai for shot noise removal





• High definition resonant scanning

System diagram



*1 When using Spectral Detector Unit. *2 NI-TT Quadrocular Tilting Tube can be used. *3 Dedicated adapter may be required, depending on microscope model.
*4 Adapter for Ti2-LAPP system, light source, hybrid dichroic mirror for photostimulation and imaging, and control board are all required.

Laser units with great flexibility and efficiency

LU-NV series

- Supports up to eight wavelengths and switching between seven fiber outputs.
- Lasers available for this series are: 405 nm, 445 nm, 458 nm, 488 nm, 514 nm, 532 nm, 561 nm, 594 nm, 640 nm and 647 nm.
- High-power lasers for the N-SIM/N-STORM super resolution microscope are available.

LU-N4/N4S 4-laser unit/ LU-N3 3-laser unit

The LU-N4/LU-N4S is equipped with four lasers (405 nm, 488 nm, 561 nm, and 640 nm), while the LU-N3 has three lasers (405 nm, 488 nm, and 561 nm). The LU-N4S is compatible with spectral imaging.

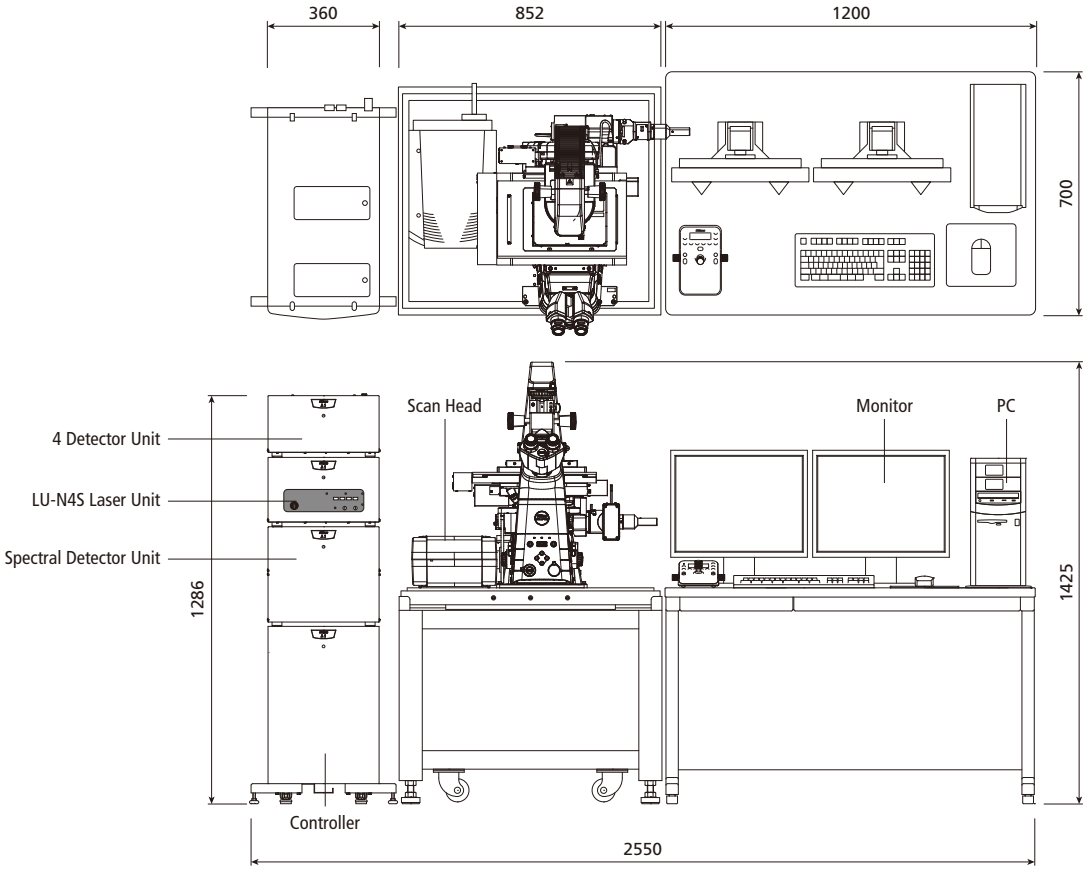
Specifications

		A1 HD25	A1R HD25
Scan head input/output port		1 laser input port 2 signal output ports for standard, spectral and optional detector*1	
Laser	LU-N3 3-laser unit	405 nm, 488 nm, 561nm lasers are installed; built-in AOTF *Cannot be used with A1-DUS spectral detector	
	LU-N4/N4S 4-laser unit	405 nm, 488 nm, 561 nm,640 nm lasers are installed; built-in AOTF *LU-N4 cannot be used with A1-DUS spectral detector	
	LU-NV series laser unit	Compatible lasers : 405 nm, 445 nm, 458 nm, 488 nm, 514 nm, 532 nm, 561 nm, 594 nm, 640 nm, 647 nm ; built-in AOTF	
Standard fluorescence detector	Wavelength	400-750 nm	
	Detector	A1-DU4-2 4 Detector Unit: 4 Multi-Alkali PMTs A1-DUG-2 GaAsP Multi Detector Unit: 2 GaAsP PMTs + 2 Multi-Alkali PMTs	
	Filter cube	6 filter cubes commonly used for a microscope mountable on each of three filter wheels Recommended wavelengths: 450/50, 482/35, 515/30, 525/50, 540/30, 550/49, 585/65, 595/50, 700/75	
Diascopic detector (option)	Wavelength	485-650 nm	
	Detector	Multi-Alkali PMT	
FOV		Ti2-E: Square inscribed in a ø25 mm circle Ni-E/FN1: Square inscribed in a ø18 mm circle	
Image bit depth		4096 gray intensity levels (12 bit)	
Scan head	Standard image acquisition	Scanner: galvano scanner x2 Pixel size: max. 4096 x 4096 pixels Scanning speed: Standard mode: 1.4 fps (512 x 512 pixels, bi-direction, 0.72x zoom) 2 fps (512 x 512 pixels, bi-direction, 1x zoom) Fast mode: 10 fps (512 x 512 pixels, bi-direction, 8x zoom) 200 fps (512 x 16 pixels, bi-direction, 8x zoom)*2 Zoom: 0.72-1000x continuously variable Scan mode: X-Y, X-T, X-Z, XY rotation, Free line, Line-Z	
	High-speed image acquisition	—	Scanner: resonant scanner (X-axis, resonance frequency 7.8 kHz), galvano scanner (Y-axis) Pixel size: max. 1024 x 1024 pixels Scanning speed: 15 fps (1024 x 1024 pixels), 30 fps (512 x 512 pixels), 60 fps (256 x 256 pixels) to 720 fps (512 x 16 pixels), 7,800 lines/sec (line speed) Zoom:0.72x, 0.82x, 0.9x, 1x, 1.2x, 1.5x, 1.75x, 2x, 2.4x, 3x, 4x, 5x, 6x, 7x, 8x Scan mode: X-Y, X-T, X-Z Acquisition method: High-speed image acquisition,
	Dichroic mirror	Low-angle incidence method, Number of positions: 8 Standard filter: 405/488/561/640, BS20/80 Optional filter:405/488, 405/488/561, 405/488/543/640, 457/514	
	Pinhole	12-256 µm variable (1st image plane)	
Spectral detector (option)	A1-DUS spectral detector unit	Number of channels: 32 Wavelength detection range: 400 - 750 nm Spectral image acquisition speed: 4 fps (256 x 256 pixels) Maximum pixel size: 2048 x 2048 (Spectral mode/Virtual filter mode) Wavelength resolution: 2.5/6.0/10.0 nm, wavelength range variable in 0.25 nm steps Compatible with galvano scanner only	
	A1-DUVB-2 GaAsP detector unit	Number of channels: 1 GaAsP PMT with variable emission plus 1 optional GaAsP PMT (A1-DUVB-OP) with a user-defined dichroic mirror and barrier filter Wavelength detection range: 400 - 720 nm, narrowest: 10 nm, broadest:320 nm Maximum pixel size: 4096 x 4096 (CB mode/VB mode) Wavelength resolution: 10 nm, wavelength range variable in 1 nm steps Compatible with galvano and resonant scanners	
Z step		Ti2-E: 0.01 µm, 0.02 µm (with encoder control), FN1 stepping motor: 0.05 µm, Ni-E: 0.025 µm	
Compatible microscopes		ECLIPSE Ti2-E inverted microscope, ECLIPSE FN1 fixed stage microscope, ECLIPSE Ni-E upright microscope (focusing nosepiece type and focusing stage type)	
Option	Motorized XYZ	Motorized XY stage (for Ti2-E/Ni-E), High-speed Z stage (for Ti2-E), High-speed piezo objective-positioning system (for FN1/Ni-E)	
	Photostimulation module*3 (for Ti2-E)	XY galvano scanning unit (Light source: LU-N3/N4 Laser Unit) DMD module (Light source: C-LEDFI Epi-FL LED illuminator, LU-N3/N4 laser unit) Stimulation form: ROI/line/point Stimulation mode: Sequential, Simultaneous	
Software	Acquisition/analysis	Basic software: NIS-Elements C Optional software for high-resolution acquisition: NIS-Elements ER	
	Display/image generation	2D analysis, 3D volume rendering/orthogonal, 4D analysis, spectral unmixing	
	Image format	JP2, JPG, TIFF, BMP, GIF, PNG, ND2, JFF, JTF, AVI, ICS/IDS	
	Application	FRAP, FLIP, FRET(option), photoactivation, three-dimensional time-lapse imaging, multipoint time-lapse imaging, colocalization	
Control computer	OS	Windows 10 Pro 64bit, English version or Japanese version OS Version 1709 Windows 7 Professional, 64bit, SP1 English version or Japanese version, Windows Update KB3118401 or later	
	CPU	Intel Xeon W-2125 (4.0GHz, 4 cores, 8.25 MB, 2666 MHz) or higher	
	RAM	32GB or 64GB	
	HDD	1st HP Z Turbo G2 512GB PCIe M.2 SSD 2nd SATA HDD 2TB	
	Optical Drive	Super Multi drive, up to x 16 speed or higher	
	Graphics	NVIDIA Quadro P600 or higher (NIS-Elements ER: NVDIA Quadro P4000) (PCI Express / two-screen split display supported)	
	Extension slot	Two PCI Express 3.0 (x16) slots (one slot to be used for graphics) One PCI Express 3.0 (x8) slot Two PCI Express 2.0 (x4) slot	
	LAN port	10/100/1000 Network/Interface x 2 (for connection to controller, for connection to external LAN)	
	Monitor	1600 x 1200 or higher resolution, dual monitor configuration recommended	
Recommended installation conditions		Temperature 23 ± 5 ºC, humidity 70 % (RH) or less (non-condensing)	

*1 FCS/FCCS/FLIM is possible in combination with third-party systems
*2 Fast mode is compatible with zoom 8-1000x and scanning modes X-Y and X-T. It is not compatible with Rotation, Free line, CROP, ROI, Spectral imaging, Stimulation and FLIM.
*3 Adapter for Ti2-LAPP system, light source, hybrid dichroic mirror for photoactivation and imaging, and control board are all required.

Layout

Unit: mm



* Layout sample

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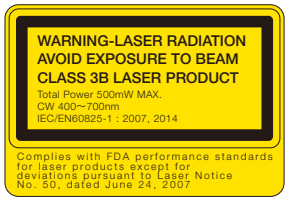

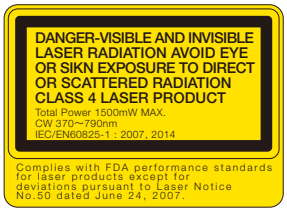

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Printed in Japan (2004)T

Code No.2CE-SCNH-7

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