

**OLYMPUS**<sup>®</sup>

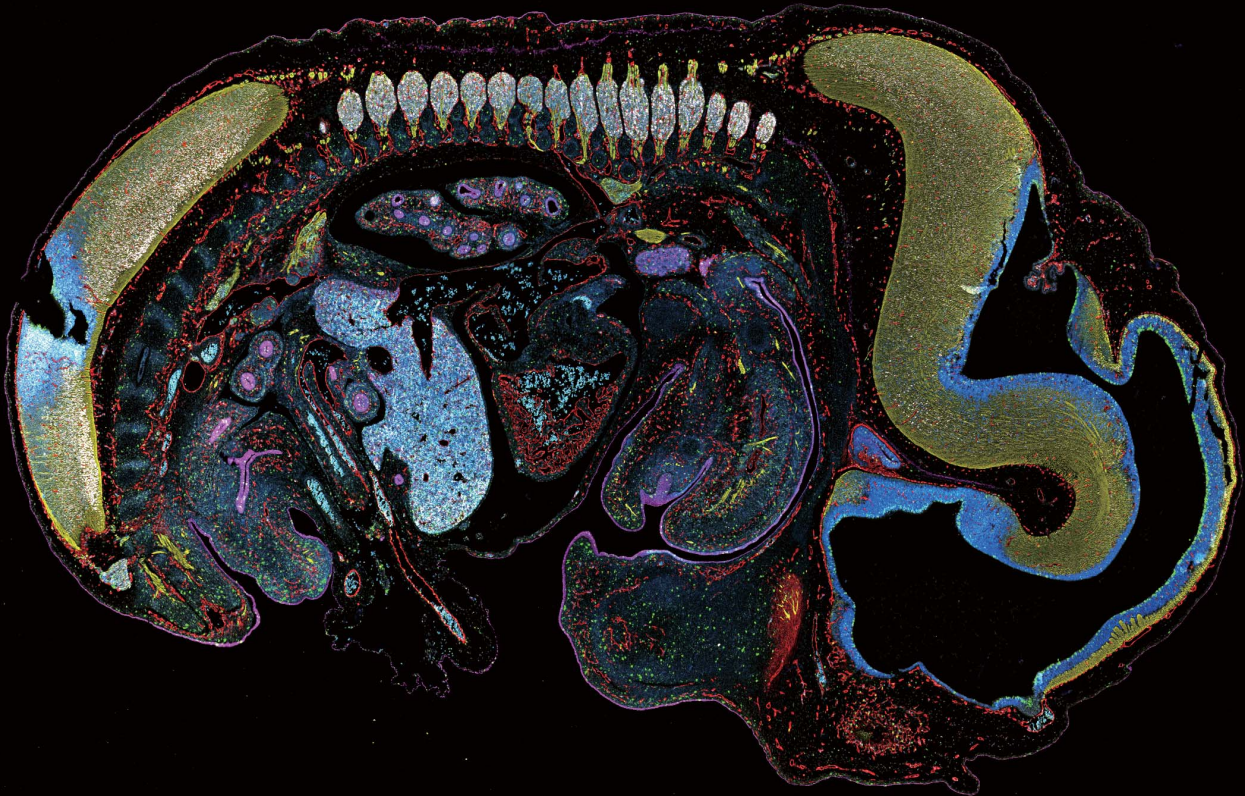
Your Vision, Our Future

Confocal Laser Scanning Microscope

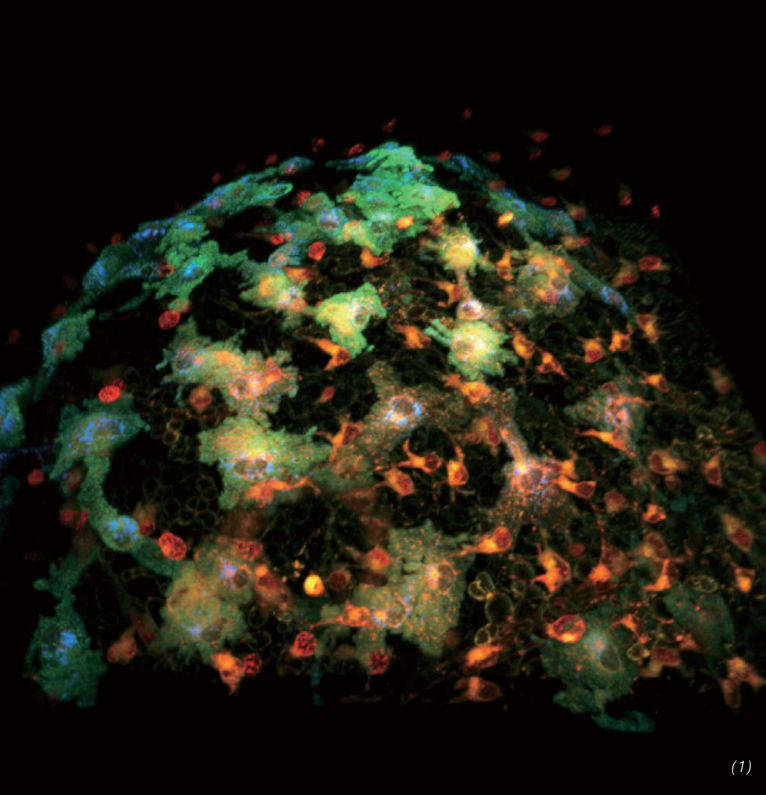
**FV3000**

FLUOVIEW

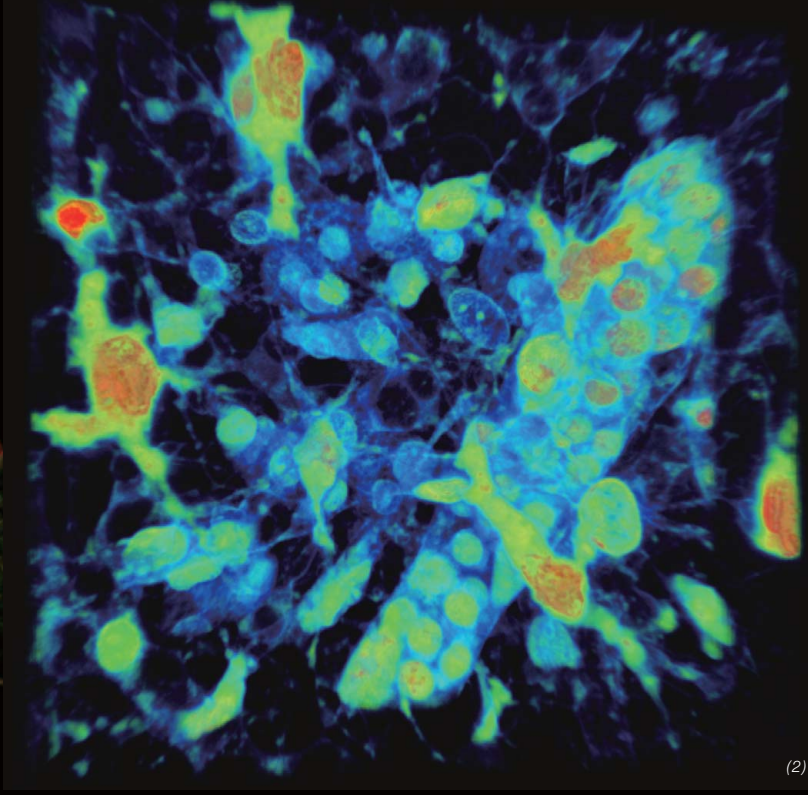
Next Generation FLUOVIEW for the Next Revolutions in Science





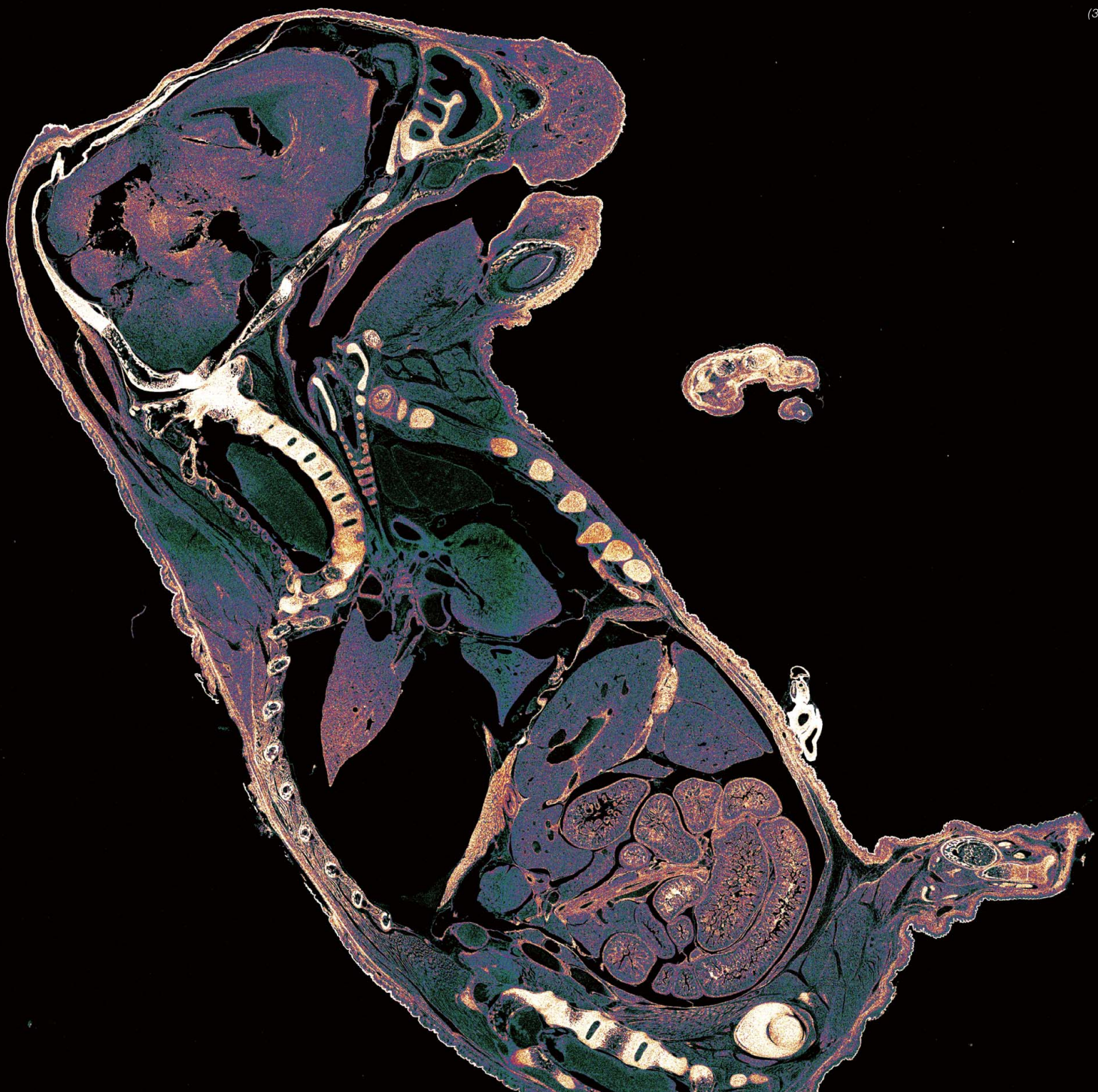


(1)



(2)

(3)



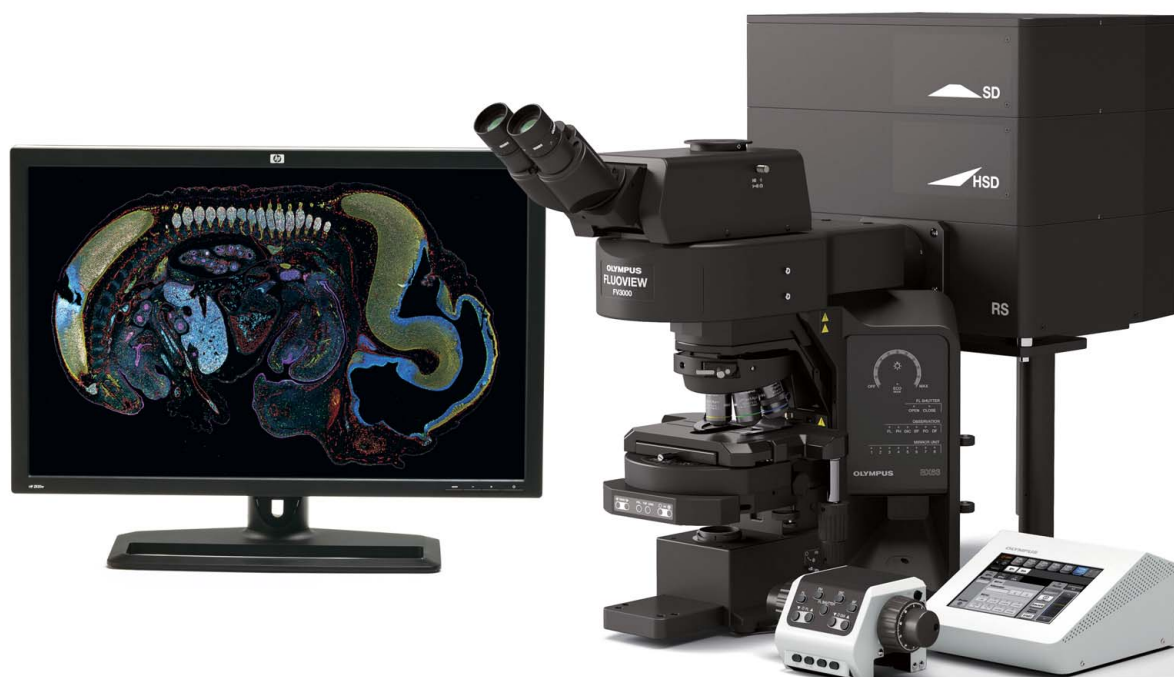


# FLUOVIEW FV3000 Laser Scanning Microscope

The FLUOVIEW FV3000 Series is designed to meet some of the most difficult challenges in modern science. With the high sensitivity and speed required for live cell and tissue imaging and the ease of use and flexibility required for microplate imaging and complex screening protocols, the FV3000 series supports complete workflows from live cell 2D–6D (x,y,λ,z,t,p) imaging through image processing, like deconvolution, and analysis. Built for fast, stable, and accurate measurements of biological reactions within living cells or tissues, even novice users can generate high-quality data and images.

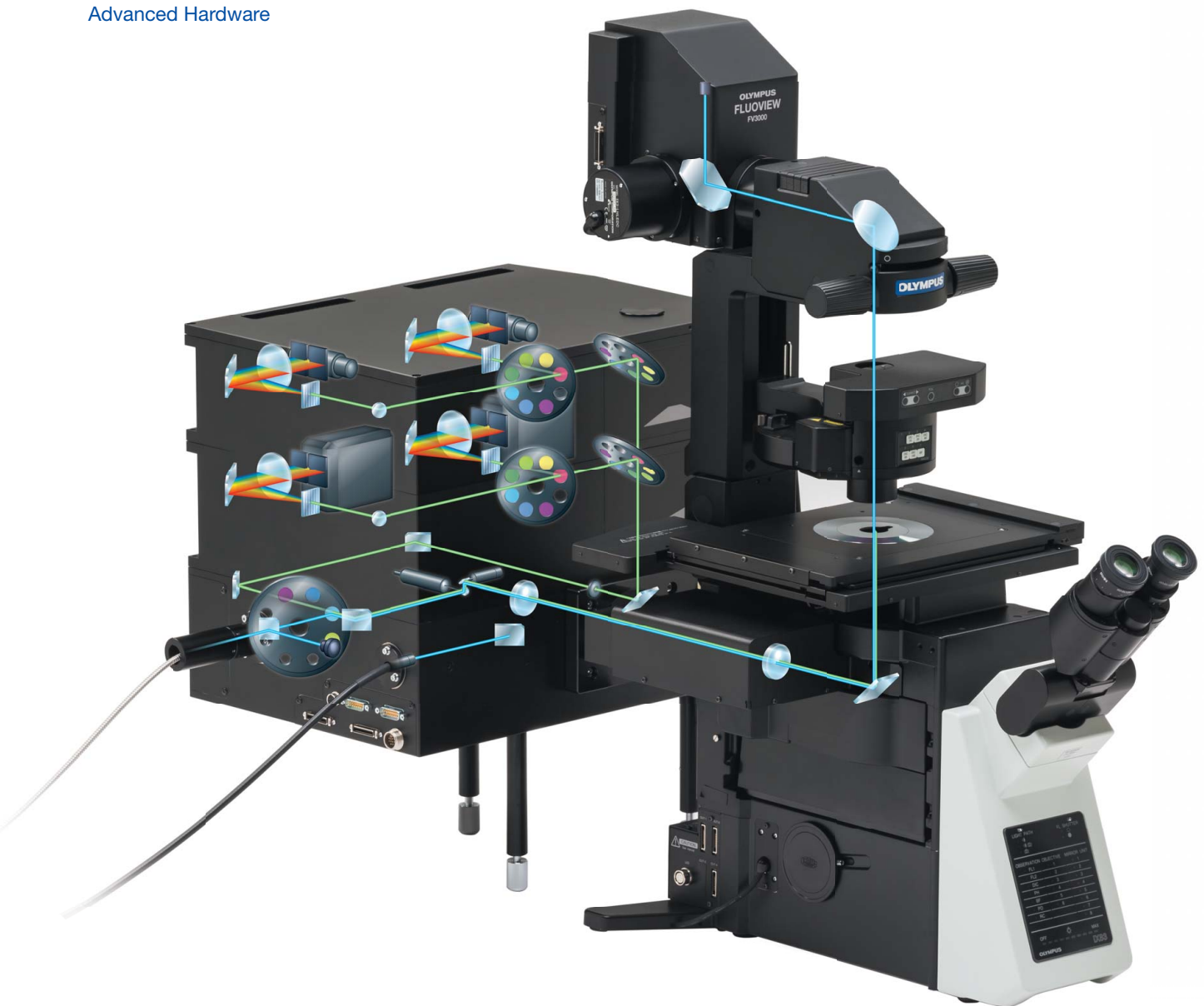
With Olympus' renowned optics at the heart of the system, the FV3000 features a new spectral detection concept for true multichannel spectral imaging with high sensitivity detection in multiple dynamic ranges, so even dim signals can be separated. The precision of galvanometer scanning is combined with the speed of resonant scanning in the FV3000RS hybrid scanner so users can combine precision and high-speed imaging in one experiment. The optical path enables macro to micro imaging from 1.25X to 150X magnification, enabling users to easily observe fine detail within the overall context of the tissue. Simplify complex experiments with robust, intuitive automation, such as one-click cellSens macro analysis for cell counting and segmentation analysis.

Built for long service life and low operating costs, the FV3000 uses long-lasting all diode lasers and LED illumination. The system features a modular, upgradeable design that includes 2-tier detection options, easily upgradeable laser configurations, and the stable and flexible IX83 or BX63LF microscopes. With user-savable and selectable software workflows, the system adjusts to individual needs. The facility manager tracking software makes it easy to track system usage by user, making the FV3000 the ideal confocal system for years of productive science in single and multi-user environments.



# Meeting the Application Challenges

Advanced Hardware



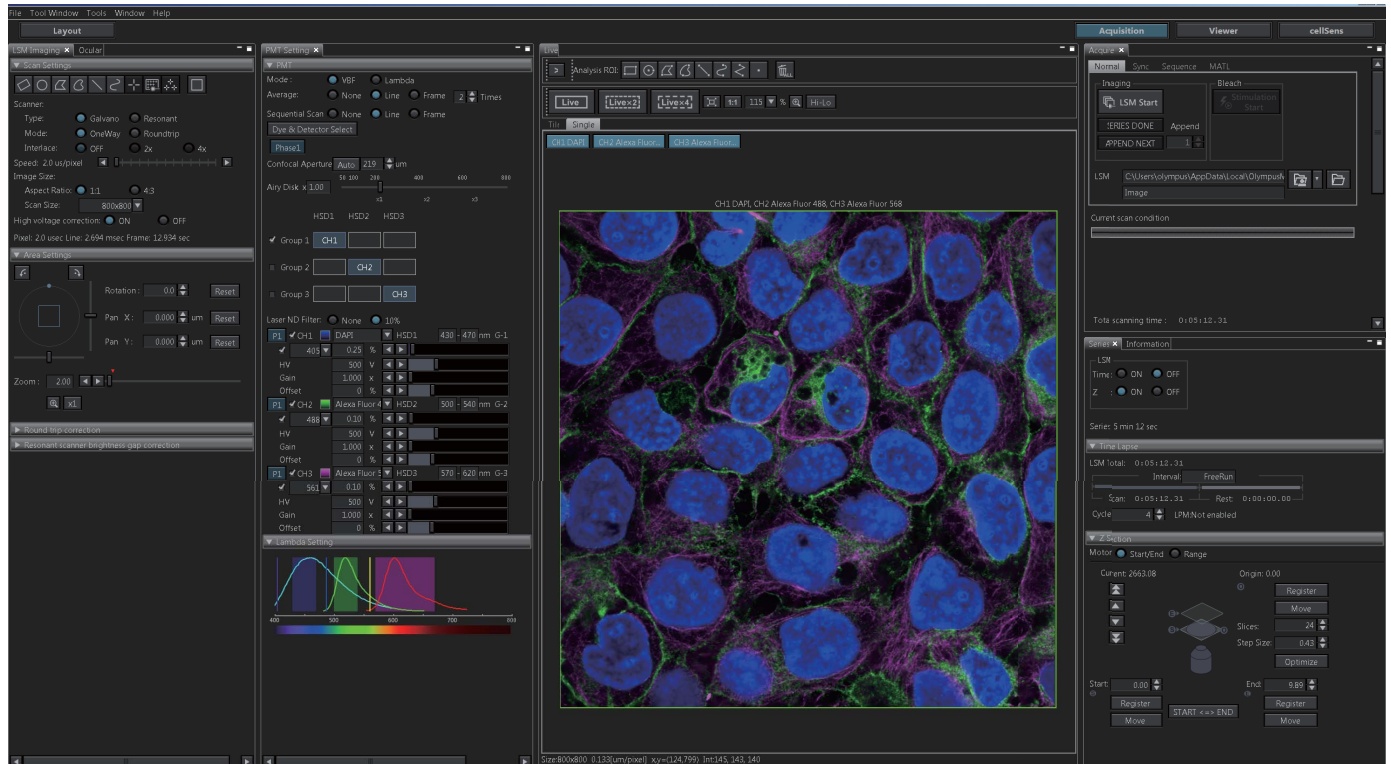
High Sensitivity  
Multi-Channel  
Imaging

Macro to Micro  
Imaging and  
Super Resolution

Increase Productivity  
with High-Speed  
Imaging



## Software User Interface Adaptable for Your Workflows



### 1. Layout

- Select the layout appropriate for your experiment

### 2. Acquisition Condition

- Auto-setting by dyes
- Reload the settings from previous experiments

### 3. Acquisition

- Acquire multi-dimensional images
- Acquire multi-area images
- Acquire images using a complex protocol

### 4. Viewer

- Review images during or after acquisition using various display methods

### 5. Analysis

- Various image analysis menus are available
- One-click macro analysis

Accurate Time-Lapse Imaging

Superior Objectives

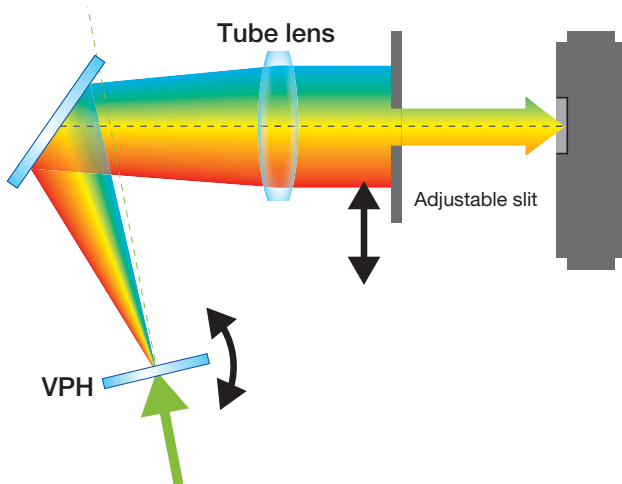
- High Sensitivity
- Multi-Channel Imaging ..... p. 5
- Macro to Micro Imaging and Super Resolution ..... p. 7
- Increase Productivity with High-Speed Imaging ..... p.11
- Accurate Time-Lapse Imaging ..... p.13
- Superior Objectives ..... p.17

# High Sensitivity Multi-Channel Imaging

## A Fully Spectral System with Sensitivity and Accuracy

The FV3000 series employs Olympus' TruSpectral detection technology. Based on patented\* Volume Phase Hologram (VPH) transmission and an adjustable slit to control light, the spectral detection is highly efficient, enabling users to select the detection wavelength of each individual channel to 2 nm.

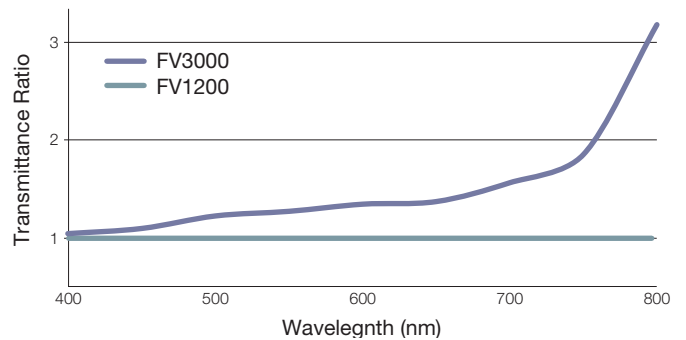
\* US8530824B/JP5541972B/EP2395380A



## Efficient TruSpectral Detection System

The FV3000 is a fully spectral series of confocal microscope powered by TruSpectral detection technology. TruSpectral detection delivers up to three-fold improved overall transmission and sensitivity, with high signal-to-noise ratio, resulting in excellent multi-color confocal imaging capabilities.

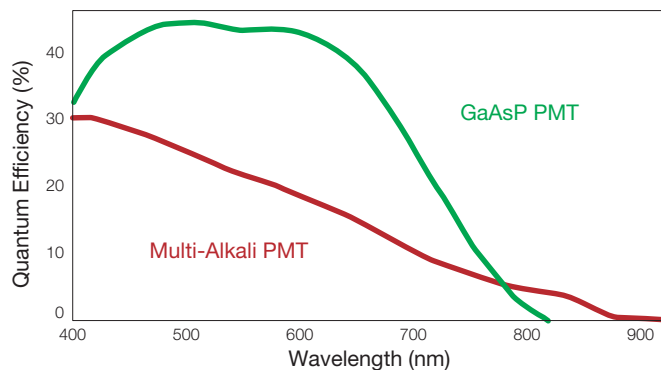
### Transmittance Ratio of FV3000 to FV1200 in Detection Path



## High-Sensitivity Spectral Detector (HSD) with GaAsP Photomultiplier Tubes Enhances Quantum Efficiency

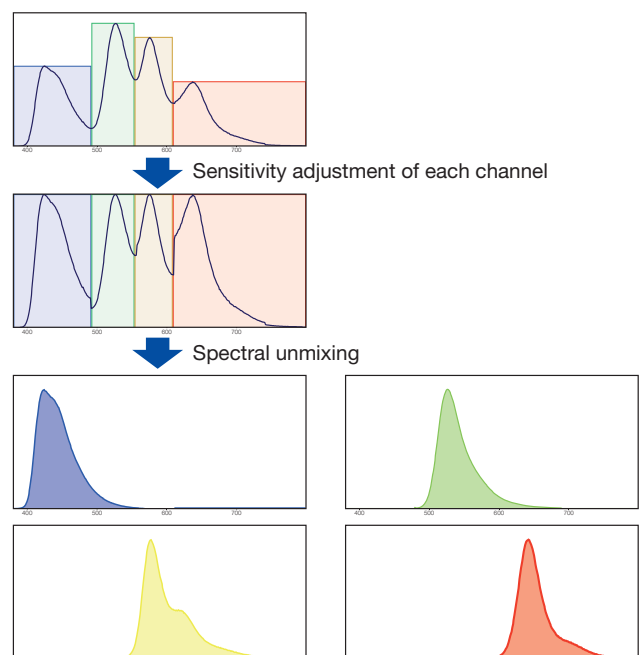
The GaAsP PMTs in the FV3000's high sensitivity detector enable users to view samples whose emission is too weak to view with conventional detection methods. The GaAsP PMT unit incorporates two channels with a maximum quantum efficiency of 45%, and Peltier cooling that reduces background noise by 20% for high S/N ratio images under very low excitation light.

### The Standard Quantum Efficiencies of Detector Technologies



## Multichannel TruSpectral Detection with Sixteen-Channel Unmixing

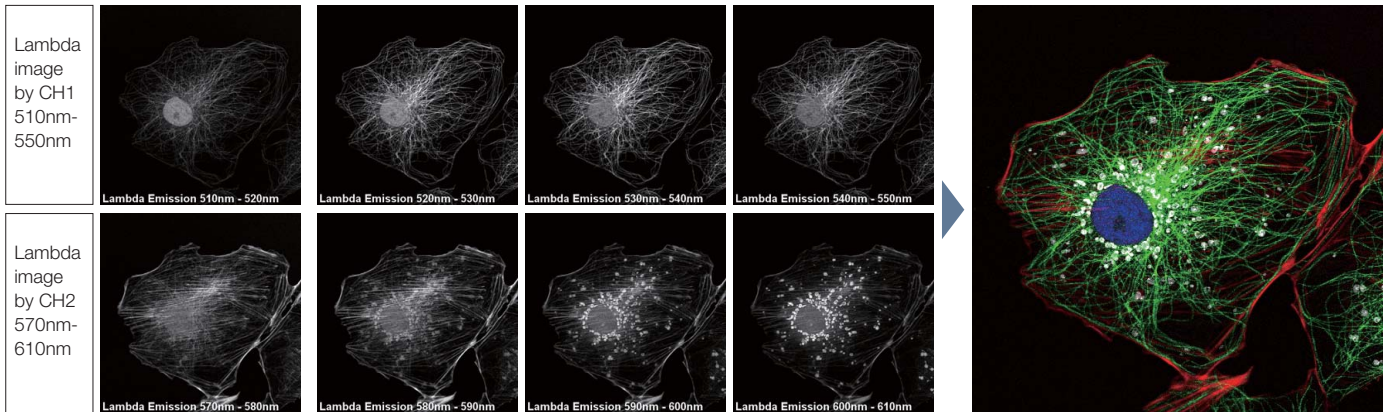
TruSpectral technology's efficient design and software enable spectral detectors to run in multichannel mode for both live and post-processing spectral unmixing with a multichannel lambda mode. The multichannel mode facilitates constant spectral unmixing during live cell experiments, separating complex fluorescence during acquisition. With up to four different dynamic ranges from the four different detection channels, bright and dim spectral signals can be separated by independently adjusting the sensitivity of each detector.





## Spectral Unmixing

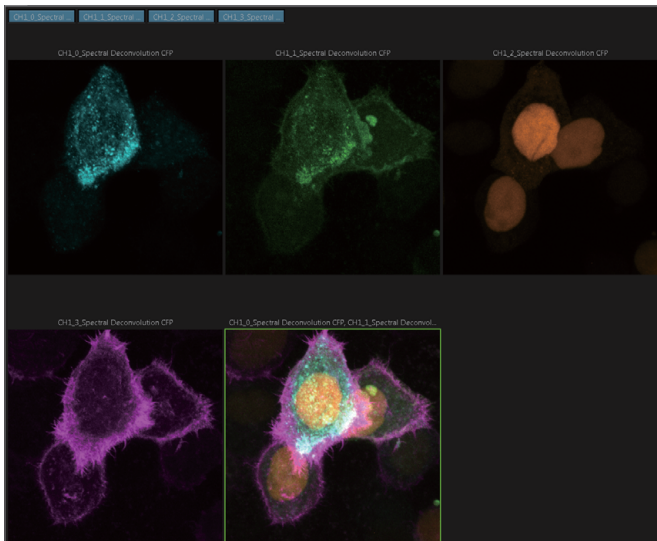
The deconvolution algorithm enables overlapping spectra to be separated based on the spectral information from lambda stack images. The fluorescence cross-talk between the channels can be eliminated by the unmixing algorithm during both image acquisition and post acquisition processing.



Spectral unmixing of a PTK2 cell labeled with YOYO-1, Alexa Fluor 488, Rhodamine-phalloidin and MitoTracker Red using multi-channel lambda stack image.

## Live Spectral Unmixing with TruSpectral Detection and Real-Time Processing

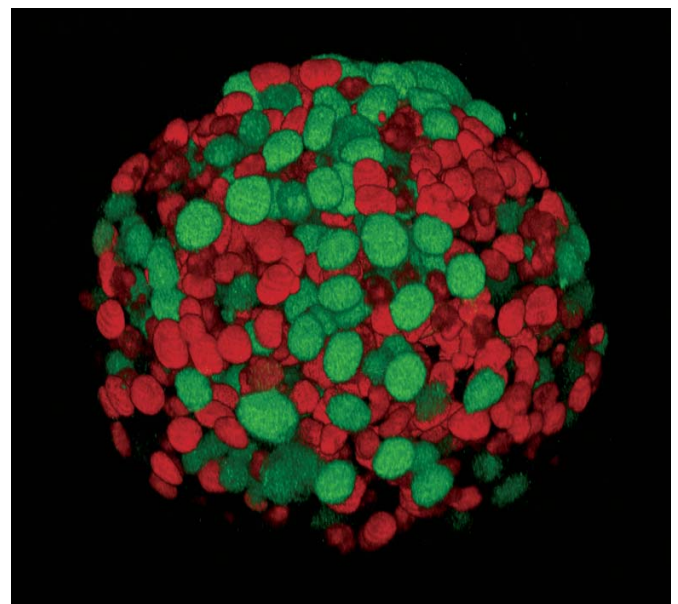
The power of TruSpectral detection plus multichannel mode means live spectral unmixing can be performed during image acquisition. Complex, overlapping spectra can be processed in real time.



Live blind unmixing of CFP (endosomes, blue), mAmetrine (plasma membrane, green), mKO (nucleus, orange) and mKeima (F-actin, purple) during time-lapse imaging. Image data courtesy of Dr. Kazuhiro Aoki, Dr. Michiyuki Matsuda, Graduate School of Medicine, Kyoto University.

## Live 3D Rendering

See your data unfold in real time with the live 3D image display function of the FV3000 software. 3D images can be constructed during image acquisition and shown as live images.

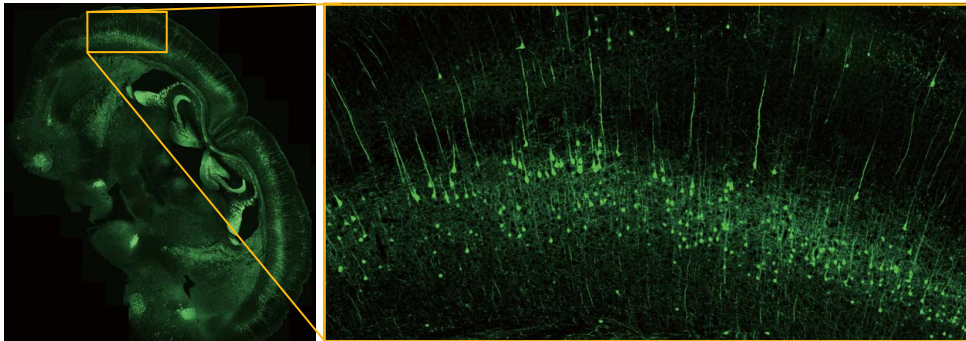


Fucci induced Spheroid of HT29 cell line  
Yuji Mishima, Ph.D., Kiyohiko Hatake M.D., Ph.D. Clinical Chemotherapy, Cancer Chemotherapy Center, Japanese Foundation for Cancer Research.

# Macro to Micro Imaging and Super Resolution

## Macro to Micro Observation

Finding areas of interest in samples can be challenging. The confocal optical design of the FV3000 series supports macro to micro imaging from 1.25X up to 150X, so users can quickly switch from low magnification overview observation to high-magnification, detailed observation of regions of interest. Users can employ image stitching at both macro and micro levels to generate overview images that show samples in context.



A stitched image of a coronal section (30  $\mu\text{m}$  thickness) from an adult YFP-H mouse cerebrum acquired with 20X objective (UPLSAPO20X).  
Image data courtesy of Takako Kogure and Atsushi Miyawaki, Cell Function Dynamics, Brain Science Institute of RIKEN.

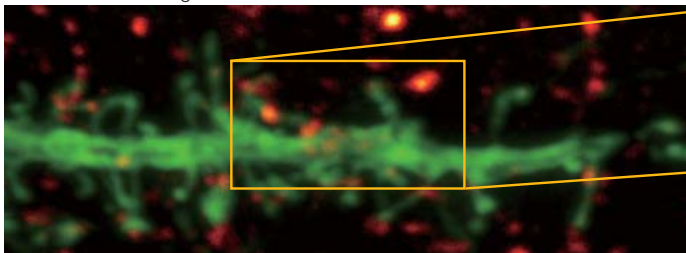
## Olympus Super Resolution (FV-OSR) Technology with Up to Four Simultaneous Channels

Olympus' widely applicable super resolution method requires no special fluorophores and works for a wide range of samples. Ideal for colocalization analysis, the Olympus Super Resolution imaging module can acquire four fluorescent signals either sequentially or simultaneously with a resolution of approximately 120 nm\*, nearly doubling the resolution of typical confocal microscopy. The imaging module is easy to use with minimal user training and can be added to any confocal system, making it a truly accessible method for achieving super resolution.

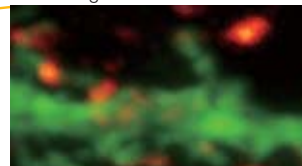
\* Subject to objective magnification, numerical aperture, excitation and emission wavelength, and experiment conditions.

## Beyond Deconvolving Confocal: Comparison of Confocal, Deconvolved Confocal and Deconvolved FV-OSR Images

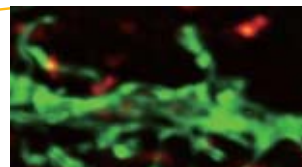
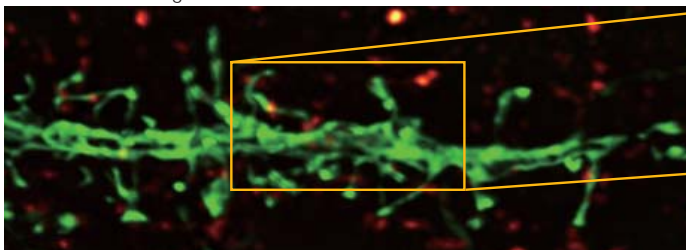
0.5 AU Confocal Image



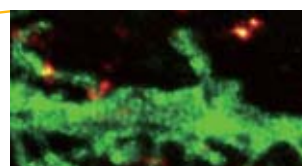
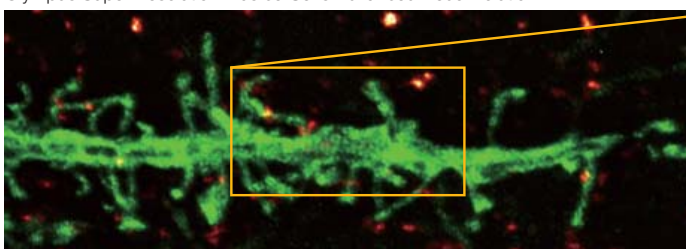
Enlargement



0.5 AU Confocal Image Deconvolved with cellSens Advanced Deconvolution



Olympus Super Resolution Plus cellSens Advanced Deconvolution

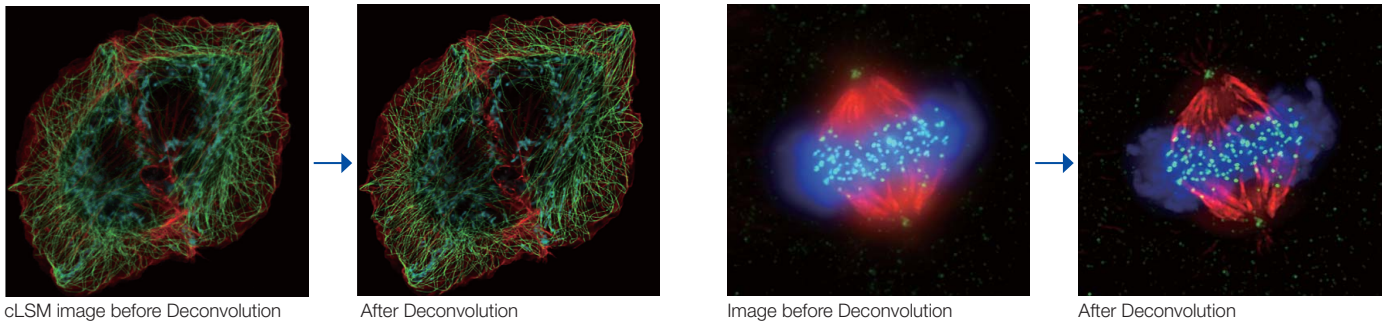


Secondary antibody labels against GFP (Alexa Fluor 488, neurons) and SV2 (Alexa Fluor 565, red).  
Sample courtesy of Dr. Ed Boyden and Dr. Fei Chen, MIT.



## Deconvolution

The optional constrained iterative deconvolution function improves the resolution, contrast, and dynamic ranges of confocal images obtained by the FV3000. The deconvolution function can be combined with Olympus Super Resolution (FV-OSR) to improve the z-axis resolution of the deconvolved images.

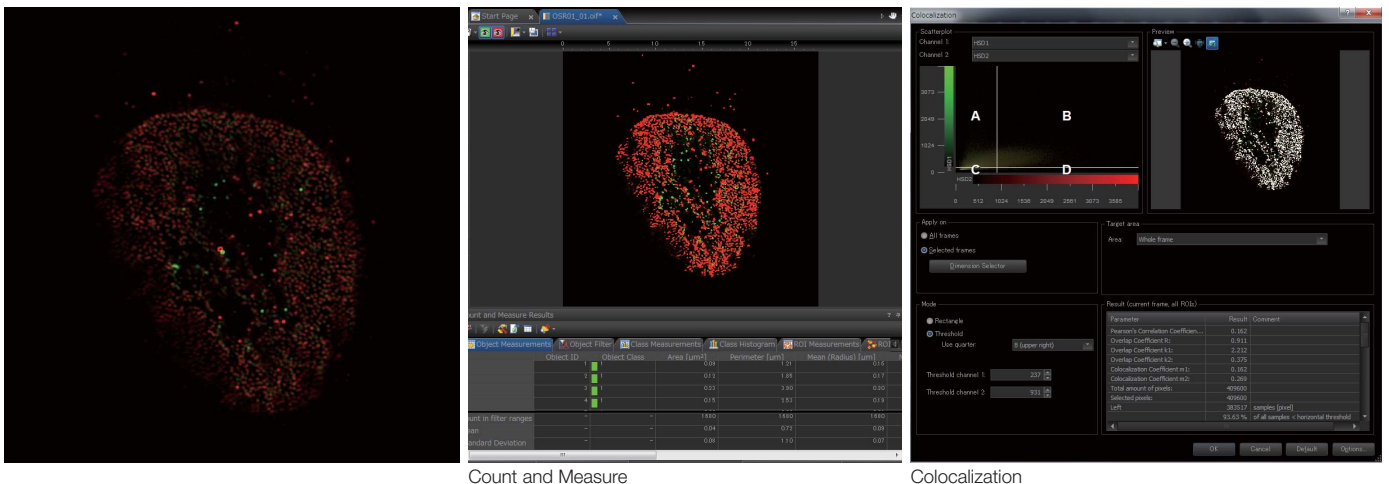


COS7 cells, triple staining,  
DAPI (cyan), Actin Bodipy-FL(green), Tubulin AF568 (red)  
Sample courtesy  
J. Doehner and U. Ziegler, Center for Image Analysis and Microscopy, University of Zurich

Cell line: HeLa (human cervical cancer cell line)  
Immunostaining: Hec1 staining (green, Alexa Fluor 488),  $\alpha$ -tubulin staining (red, Alexa Fluor 568), DAPI staining (blue)  
Mitotic spindle and kinetochores are stained with anti- $\alpha$ -tubulin (red) and anti-Hec1 (green) antibodies, respectively. Chromosomes interact with microtubules of the mitotic spindle via kinetochores (protein structures assembled on the centromere region of chromosomes)  
Image data courtesy of Masanori Ikeda and Kozo Tanaka, Department of Molecular Oncology, Institute of Development, Aging and Cancer, Tohoku University.

## Image Analysis

The FV3000 incorporates various optional analysis functions to complete the workflow from image acquisition through data analysis. The Count and Measure solution enables the measurement of the number, size, luminosity, and morphology of the segments. Colocalization enables the analysis of overlapping fluorescent spectra.

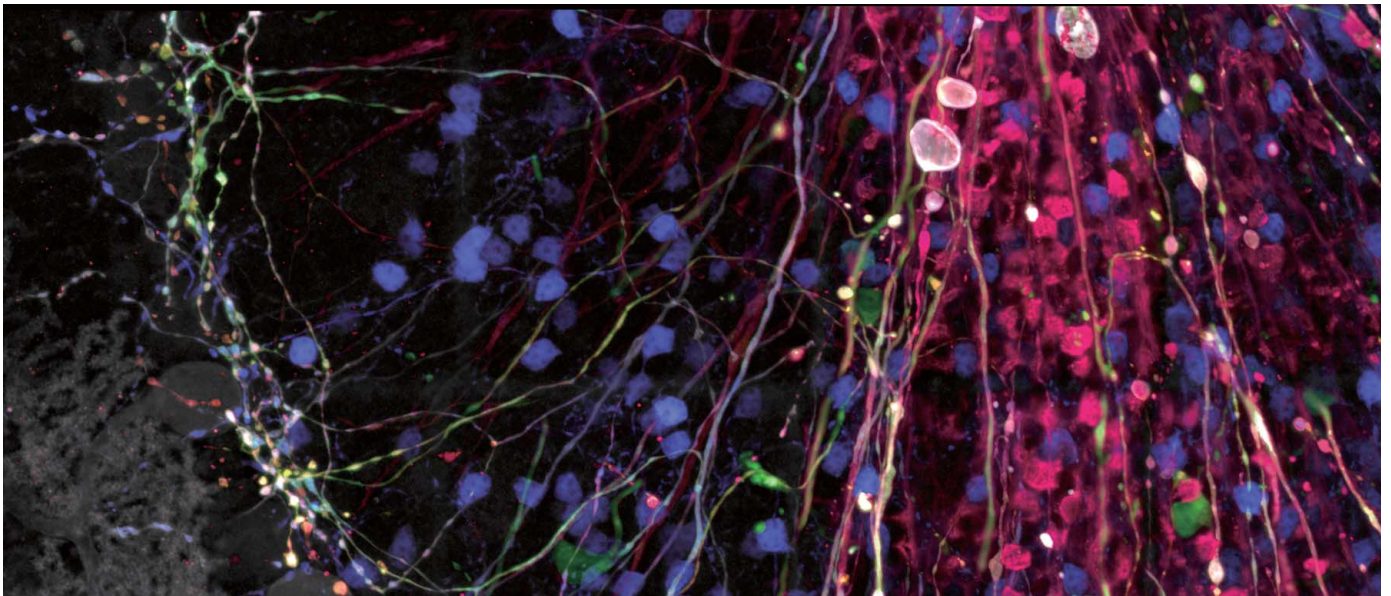


FV-OSR image of nuclear pores ( Nup153: Alexa Fluor 488, Nup62: Alexa Fluor 555)  
Sample courtesy of Hidetaka Kosako, Fujii Memorial Institute of Medical Sciences, Tokushima University

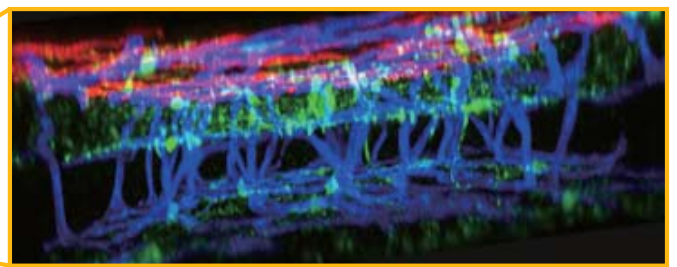
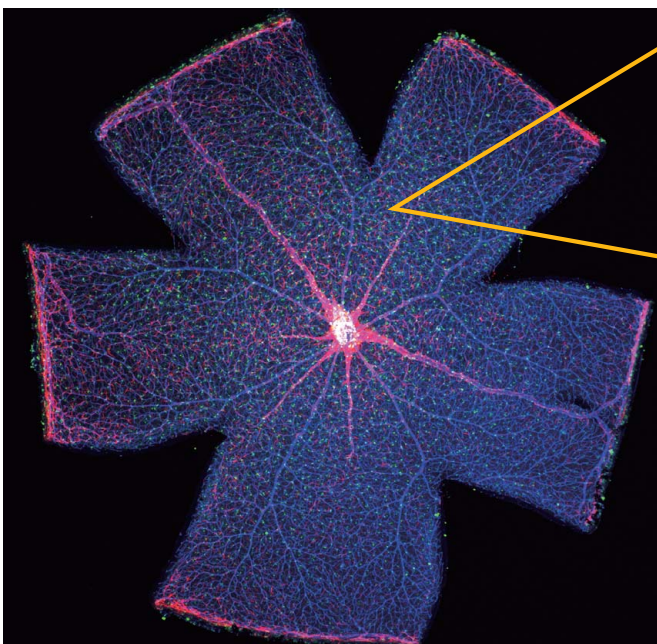
## Application: High Sensitivity Multi-Channel Imaging

Acquiring multi-color fluorescent images is important for analyzing the fine internal structure of cells and tissues and confirming protein expression. The FV3000's TruSpectral detectors enable users to select the detection wavelength on each channel to optimize signal detection for each individual fluorophore. The variable barrier filter mode provides simultaneous four-channel image acquisition, and the virtual channel module enables up to sixteen-channel image acquisition. The system's lambda scanning mode facilitates the separation of complex fluorescence signals by defining characteristic spectral emission profiles for each fluorophore, enabling accurate spectral unmixing of complex overlapping fluorescent signals.

### Spectral Unmixing



Brainbow AAV transfection of Purkinje cells, amplified with antibodies as described in Cai et al 2013. Visible are Purkinje cell somata, dendrites and axons, as well as some aspecific stainings of granule cells.



### Mouse retina

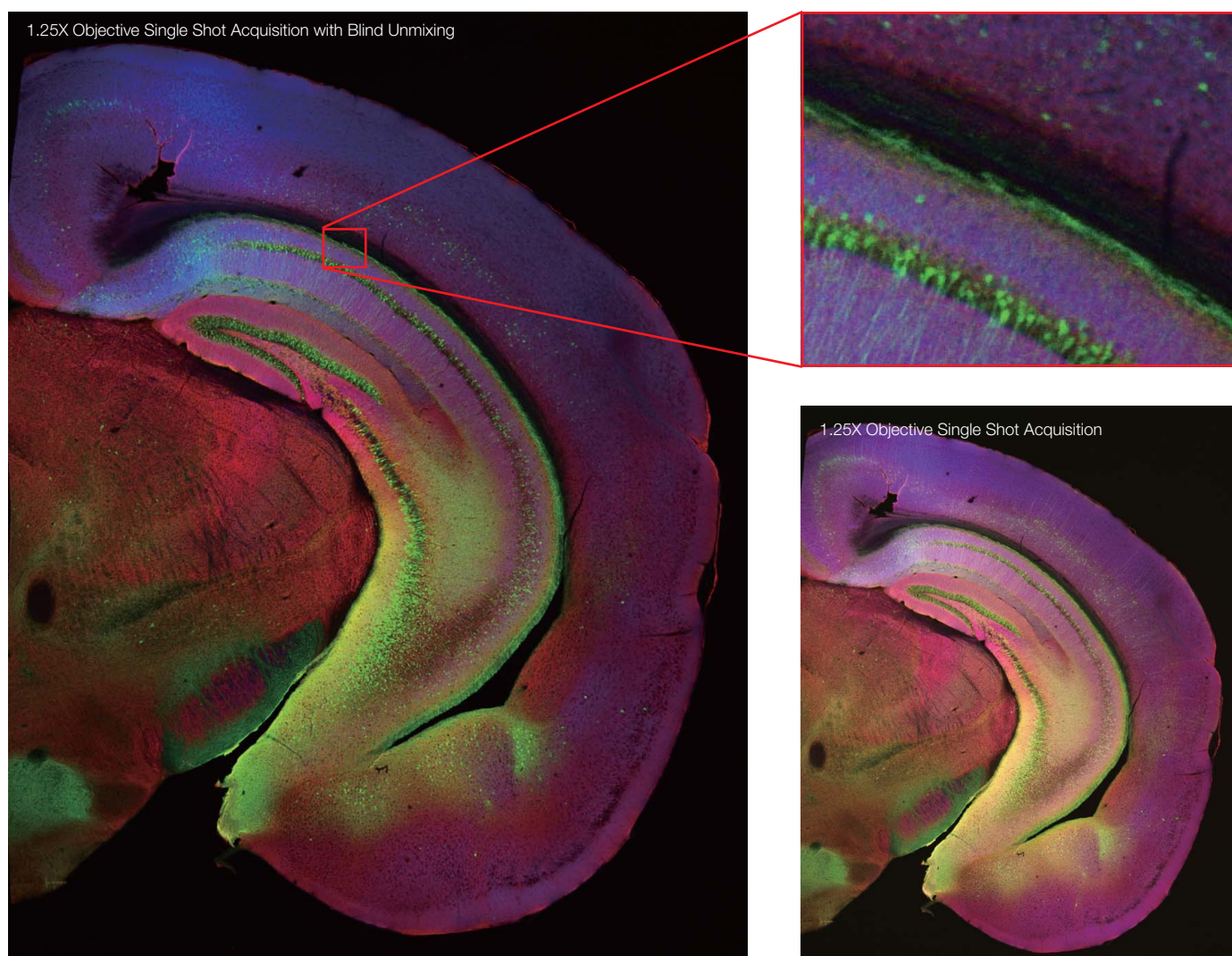
One shot overview image acquired by PLAPON2X and 3D image acquired by UPLSAPO40XS silicone immersion objective.

Image data courtesy of Dr. Yoshiaki Kubota, The Laboratory of Vascular Biology Center for Integrated Medical Research, School of Medicine, Keio University.

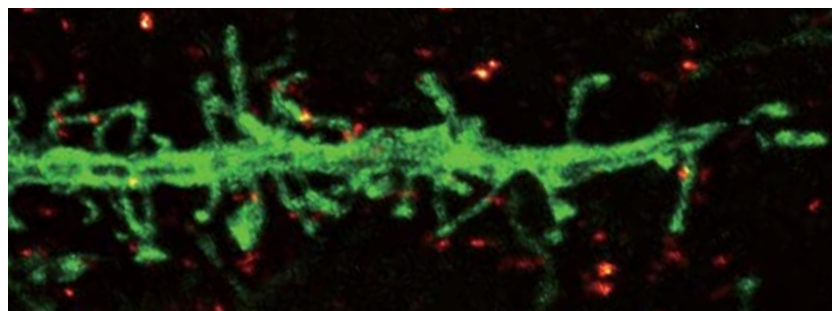


## Application: Macro to Micro Imaging and Super Resolution

Life science research applications require users to observe regions of interest within the context of the larger tissue structure. A large field of view is critical to being able to see a cell's components in relation to the entire tissue sample. The FV3000's optical lightpath facilitates macro to micro observation from 1.25X to 150X. Image stitching enhances these applications even more by enabling users to quickly locate target cells on the macro image, and follow up with higher resolution imaging of the cell's fine structures. For even greater resolution, Olympus Super Resolution technology (FV-OSR) can be coupled with this approach to provide optimal macro to micro imaging performance.



Mouse brain hemisection embedded for Expansion Microscopy (pre-expansion), labeled with secondary antibodies against GFP (Alexa Fluor 488, green), SV2 (Alexa Fluor 565, red) Homer (Alexa Fluor 647, blue). Sample courtesy of Dr. Ed Boyden and Dr. Fei Chen, MIT.

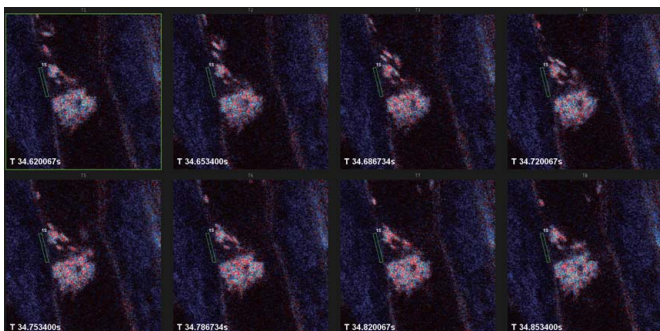


Dendrite (anti-GFP Alexa Fluor 488, green) and synaptic marker (SV2, Alexa Fluor 565, red) Olympus Super Resolution image processed with cellSens advanced constrained iterative deconvolution. Average full width half maximum measurements ~135 nm. Image acquired with 100X 1.35 NA silicone objective. Sample courtesy of Dr. Ed Boyden and Dr. Fei Chen, MIT.

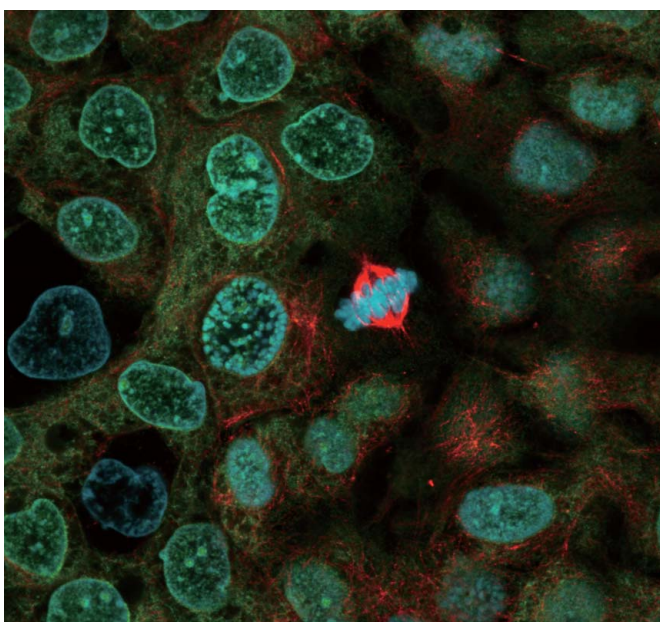
# Increase Productivity with High-Speed Imaging

## Galvanometer and Hybrid Galvo/Resonant Scanners

Users have their choice of two different types of scan units: galvanometer only with the FV3000 or hybrid galvanometer/resonant with the FV3000RS. The hybrid scan unit has a galvanometer scanner for high-precision scanning, as well as a resonant scanner that is ideal for high-speed imaging. With the galvanometer scanner and Olympus super resolution technology (FV-OSR), users can obtain resolutions down to 120nm with a high signal-to-noise ratio. The galvanometer scanner also features flexible scanning options, including precise tornado scanning as well as multipoint stimulation with 100ms switching time. The galvanometer scanner can image up to 16 frames per second. By switching to the resonant scanner, users can capture 30 frames per second with a full field of view at 512 x 512 pixels. By clipping down to 512 x 32 pixels, the resonant scanner can capture up to 438 frames per second to capture critical live physiological events such as calcium ion flux.



Platelets bound to a thrombosis in the blood vessel of a mouse. Images taken at 30 fps in full frame by resonant scanner with 2 CH GaAsP PMTs. Image data courtesy of Dr. Takuya Hiratsuka, Dr. Michiyuki Matsuda, Graduate School of Biostudies, Kyoto University.



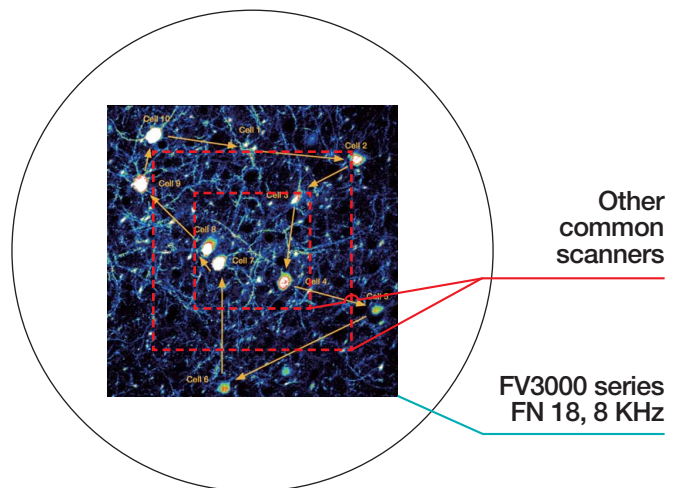
A431 cells fixed with methanol labeled with Abcam Anti-ERK1 + ERK2 antibody (Alexa Fluor 488) ab208564 and Anti-alpha Tubulin antibody (Alexa Fluor 594) ab195889 and DAPI. Sample courtesy of Abcam.

## Optimized for Live Cell Imaging

Resonant scanning greatly reduces photobleaching and phototoxicity compared to standard galvanometer scans by preventing the excitation of fluorophores into triplet states that create reactive oxygen species. These features make live cell experiments more robust and reliable. The FV3000 series has complete laser intensity control from low to high range, enabling the system to use the minimum required amount of laser power on samples. The optional laser power monitor provides consistent laser power during long-term time-lapse imaging across multiple days.

## No Compromise between Speed and Field of View

Many high-speed scanning methods restrict the field of view, limiting their usefulness for examining large areas with multiple cells. The FV3000 series' resonant scanner maintains a full 1X field of view, even at a video rate of 30 frames per second. By clipping the Y axis, additional speeds up to 438 frames per second can be achieved.

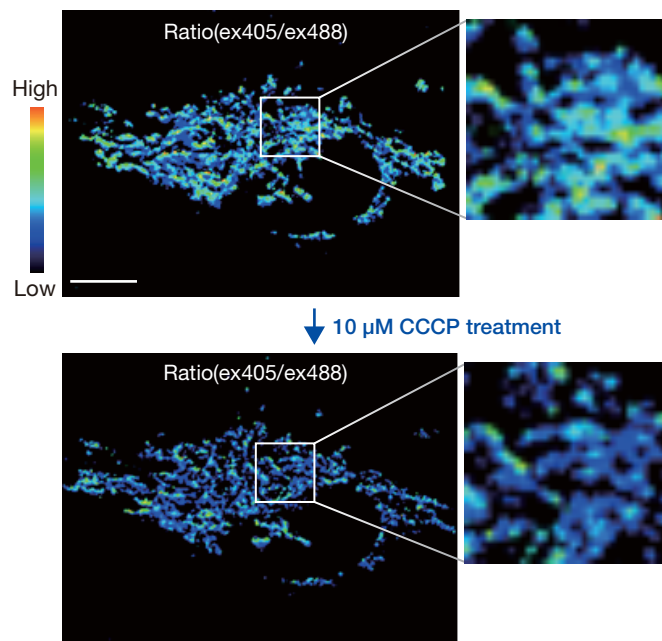


Most resonant scanners force a trade-off between speed and field of view. FLUOVIEW systems are optimized to maintain the field of view with even signal intensity so dynamic samples (e.g. calcium imaging) can be seen in the broad context of their cells and tissues. The image above shows examples of the clipped fields of view required in other resonant scanning systems.

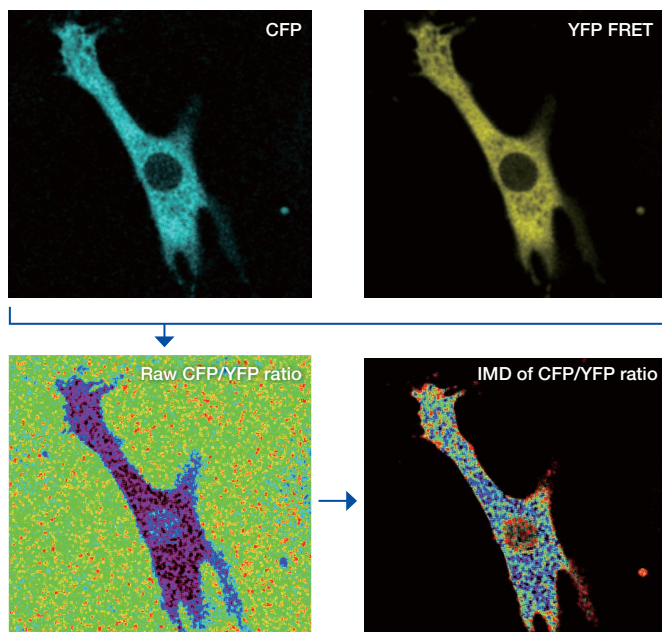


## Ratio Imaging and Intensity Modulated Display (IMD)

The FV3000's ratio imaging analysis function includes an Intensity Modulated Display (IMD) function in the software that displays quantitative fluorescence ratio changes during both standard and high-speed acquisitions. This function is particularly useful for calcium and FRET imaging where a pure ratio display provides poor contrast in background areas.



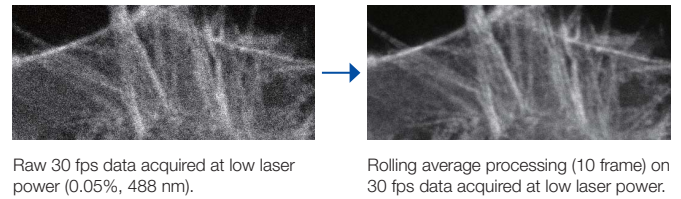
tsGFP1-mito reveals heterogeneity in mitochondrial thermogenesis in HeLa cells. Ratio images (ex 405 nm/ex 488 nm) in tsGFP1-mito-expressing cells shown before and after CCCP treatment at 37 °C. Scale bars indicate 10 μm (whole image) and 3 μm (inset). Image data courtesy of Shigeki Kiyonaka Ph.D, Yasuo Mori Ph.D Molecular Biology Field, Department of Synthetic Chemistry and Biological Chemistry, Kyoto University.



Raw and IMD ratio images of spontaneous  $\text{Ca}^{2+}$  oscillation in a beating rat cardiomyocyte expressing yellow cameleon. Image data courtesy of Yusuke Niino and Atsushi Miyawaki, Cell Function Dynamics, Brain Science Institute of RIKEN.

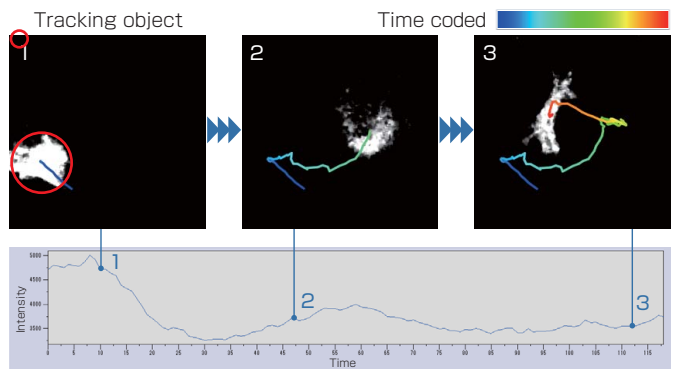
## Rolling Average Processing

High-speed scanning at low laser power to avoid phototoxicity often decreases the signal-to-noise ratio. With rolling average post-processing, users have the flexibility to adjust high-speed time-lapse images while maintaining the time scale and keeping the original data.



## Object Tracking

In time-lapse imaging, moving objects can be automatically detected, tracked, and analyzed. cellSens software's tracking function provides a powerful and intuitive tool to quantify dynamic processes such as cell movement and division.



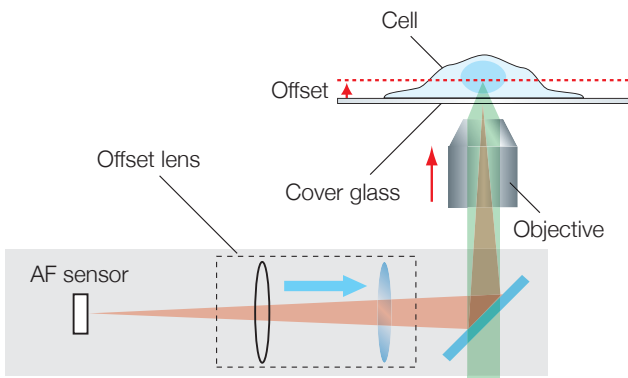
Time-dependent change in the intensity of cells

# Accurate Time-Lapse Imaging

## Maintain Focus with the Z-Drift Compensation (ZDC) System

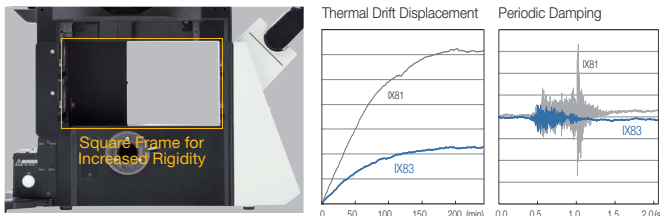
The IX3-ZDC2 Z-drift compensator uses minimally-phototoxic infrared light (laser class 1) to identify the location of the sample plane. One-shot autofocus (AF) mode enables several focus positions to be set as desired for deeper samples, enabling efficient Z-stack acquisitions in multiposition experiments. The continuous AF mode keeps the desired plane of observation precisely in focus, avoiding focus drift due to temperature changes or the addition of reagents, making it ideal for measurements that require more stringent focusing. Furthermore, the increased optical offset enables continuous AF with plastic vessels or with dry objectives. The Z-drift compensator is also compatible with silicone objectives (in AF mode).

### IX3-ZDC2 Z-Drift Compensator Optical Path Diagram



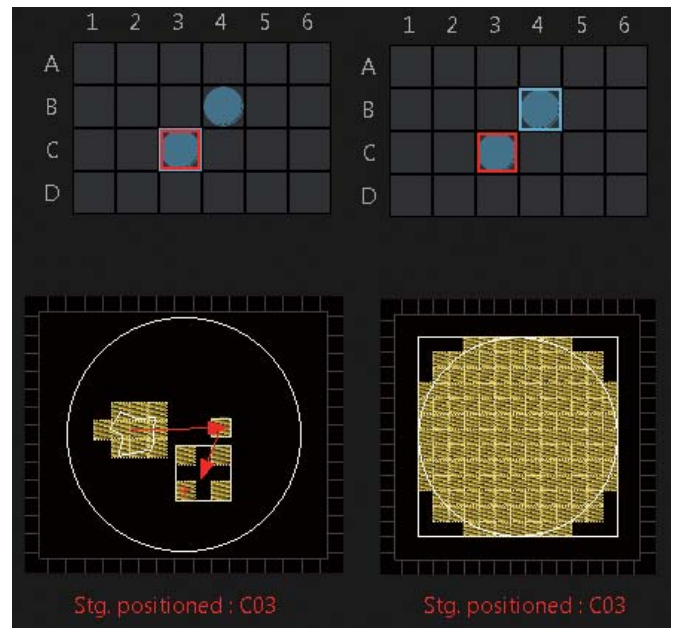
## Stable Time-Lapse Imaging with the IX83 Microscope

A Z-drive guide installed near the revolving nosepiece combines high thermal rigidity with the stability of a wraparound structure to significantly reduce the impact of heat and vibration and improve the quality of time-lapse imaging.



## Stage Control for Multi-Area Time-Lapse, Microplate, and Stitching

Multi-area time-lapse and stitching provide robust and accurate time-lapse data, and enable users to generate detailed overview images to see their data in context. The well navigator function provides sophisticated, intuitive controls for a wide range of cell culture vessels and custom plates.



## Hard Disk Recording

The microscope comes equipped with a hard-disk drive (HDD) recording function. The images are stored automatically in the HDD. Large volumes of data, such as those obtained from long-term time-lapse imaging, can be easily collected.

## High Contrast under Bright Conditions

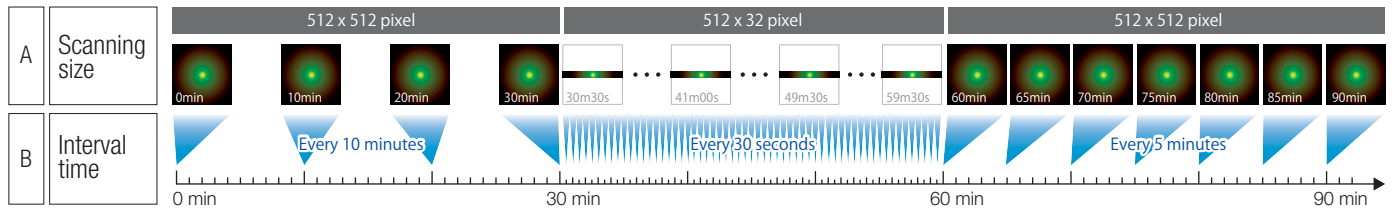
The umbra unit is designed specifically for fluorescence observation under bright room conditions. It efficiently blocks out room light, enhances the contrast of fluorescence, and enables clear fluorescence observation without the need for a dark room.





## Reduce Complexity with the Sequence Manger

With the Sequence Manager software module, complex protocols are handled with ease and accurate timing. Multi-day time-lapse experiments are controlled with microsecond scan accuracy and millisecond sequence execution accuracy. Various protocols, such as time-lapse with different time intervals, switching between high and low magnification, and photo-stimulation between imaging by FRAP or FRET (acceptor photobleaching), can be performed.

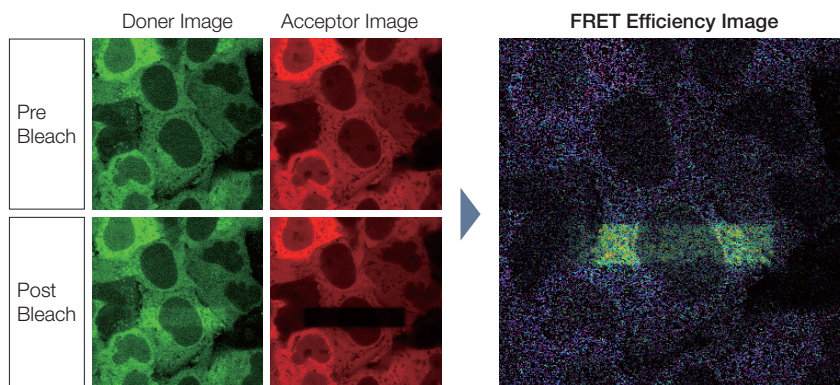


The Sequence Manager enables variable time-lapse experiments with microsecond scanning accuracy

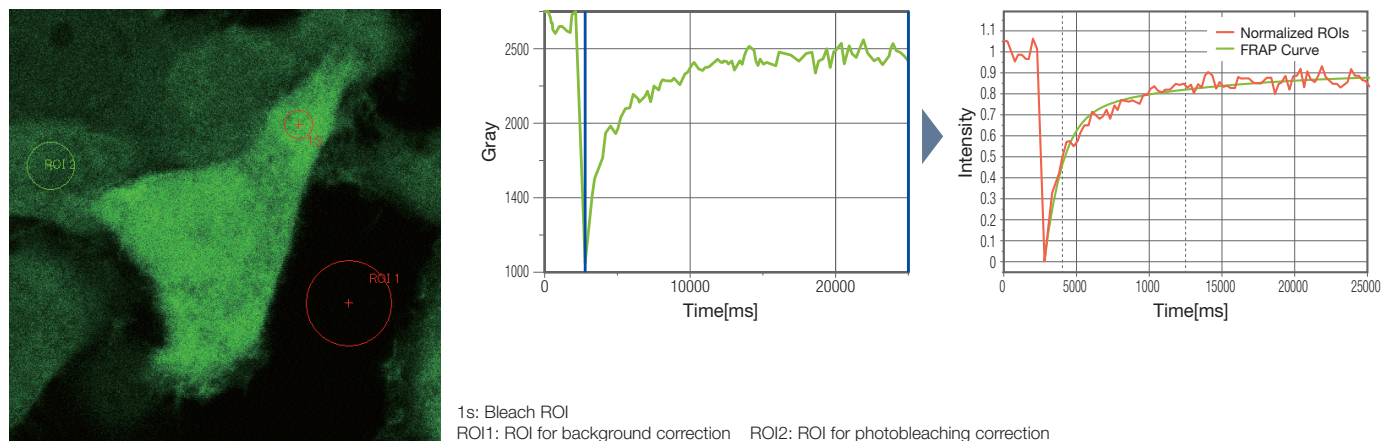
## Life Science Analysis

The cellSens Life Science Analysis module enables analysis of images from FRAP or FRET experiments. In FRAP,  $\tau/2$  and the Mobile/Immobile fraction can be estimated by fitting the curve of luminosity change caused by fluorescence recovery after bleaching. FRET enables the measurement of FRET efficiency by acceptor photobleaching, ratio imaging, and sensitized emission.

### Example of FRET analysis (acceptor photobleaching)

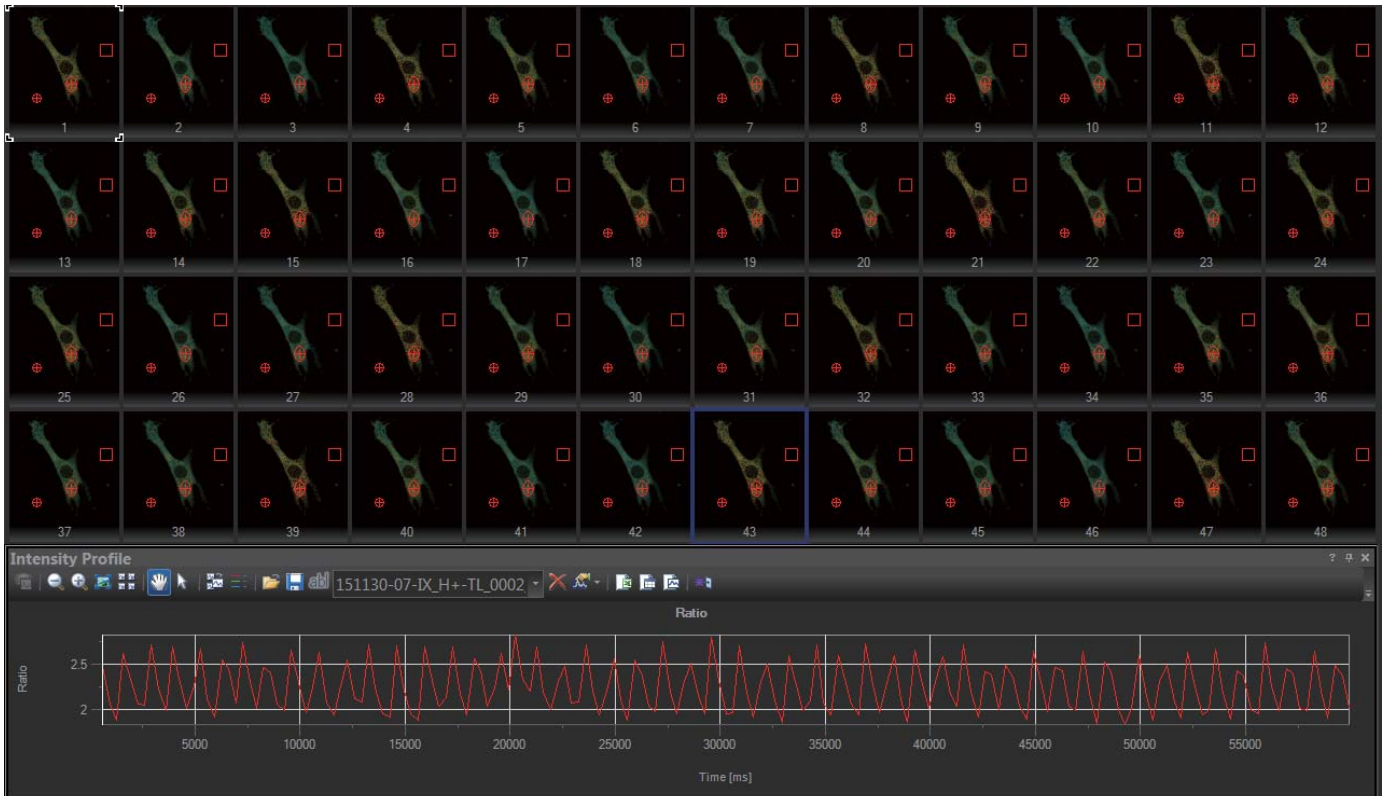


### Example of FRAP analysis

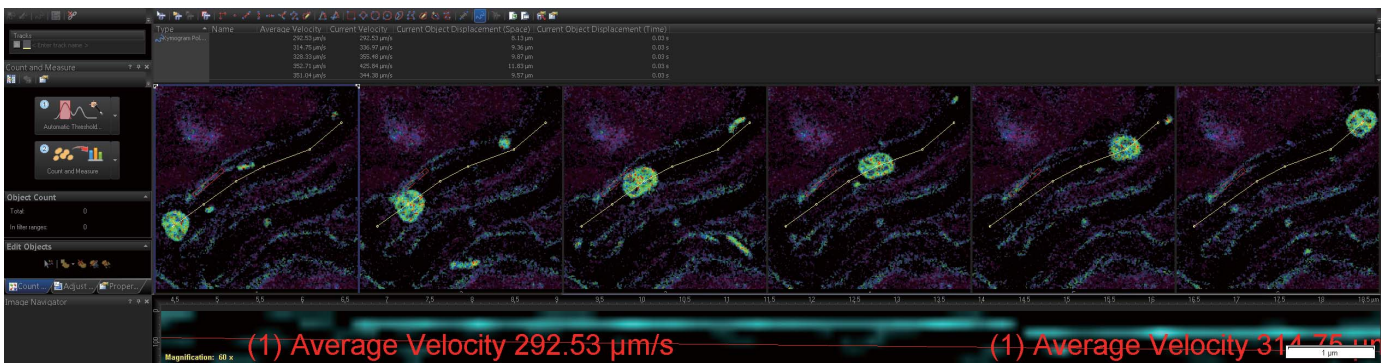


## Application: High-Speed Imaging

High-speed imaging is required to observe fast dynamic phenomena, such as a beating heart, blood flow, or calcium ion dynamics inside cells. The FV3000RS hybrid scan unit uses a galvanometer scanner for precision scanning as well as a resonant scanner that is ideal for high-speed imaging of live physiological events. The resonant scanner is capable of speeds starting from 30 fps at FN18, up to 438 fps using clip scanning. Users can switch between the galvanometer scanner and resonance scanner with the click of a button, depending on the goals of their research.



Intensity Modulated Display of the CFP/YFP ratio result during spontaneous contractions of an *in vitro* cardiomyocyte. Image data courtesy of Yusuke Nino and Atsushi Miyawaki, Cell Function Dynamics, Brain Science Institute of RIKEN.

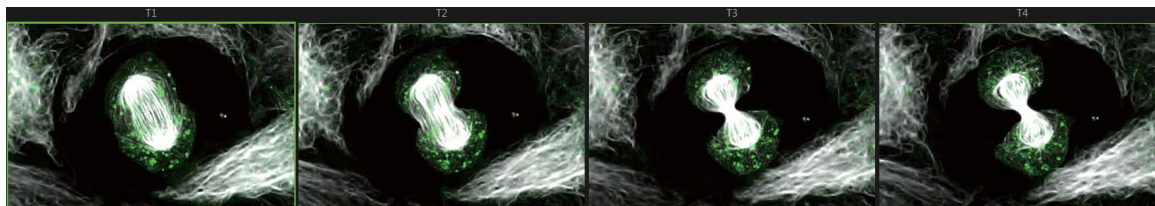


Platelets bound to a thrombosis in the blood vessel of a mouse. Images taken at 30 fps in full frame using a resonant scanner with 2 CH GaAsP PMTs. Image data courtesy of Dr. Takuya Hiratsuka, Dr. Michiyuki Matsuda, Graduate School of Biostudies, Kyoto University.

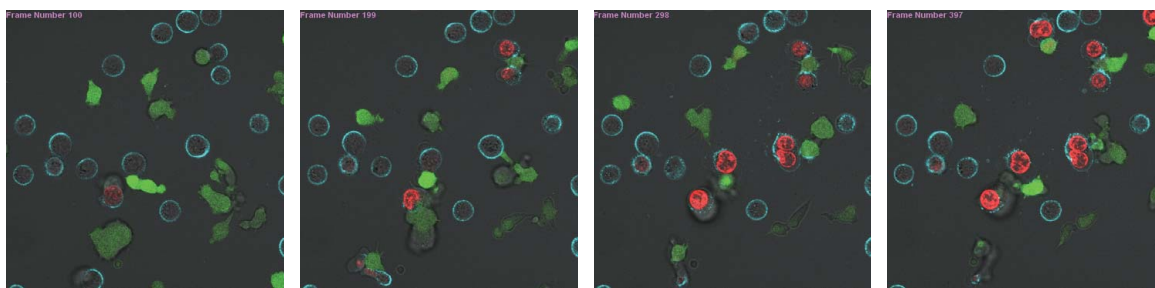


## Application: Time-Lapse Imaging

Time-lapse imaging experiments require focus to be maintained throughout with less phototoxicity to the sample. Olympus' Z-drift compensator helps enable that each well stays in focus during an experiment despite changes in temperature or the addition of reagents. Additionally, the FV3000's high sensitivity detector requires reduced laser intensity, and the resonant scanner reduces the laser illumination time, thus lowering phototoxicity for more physiologically accurate imaging data.



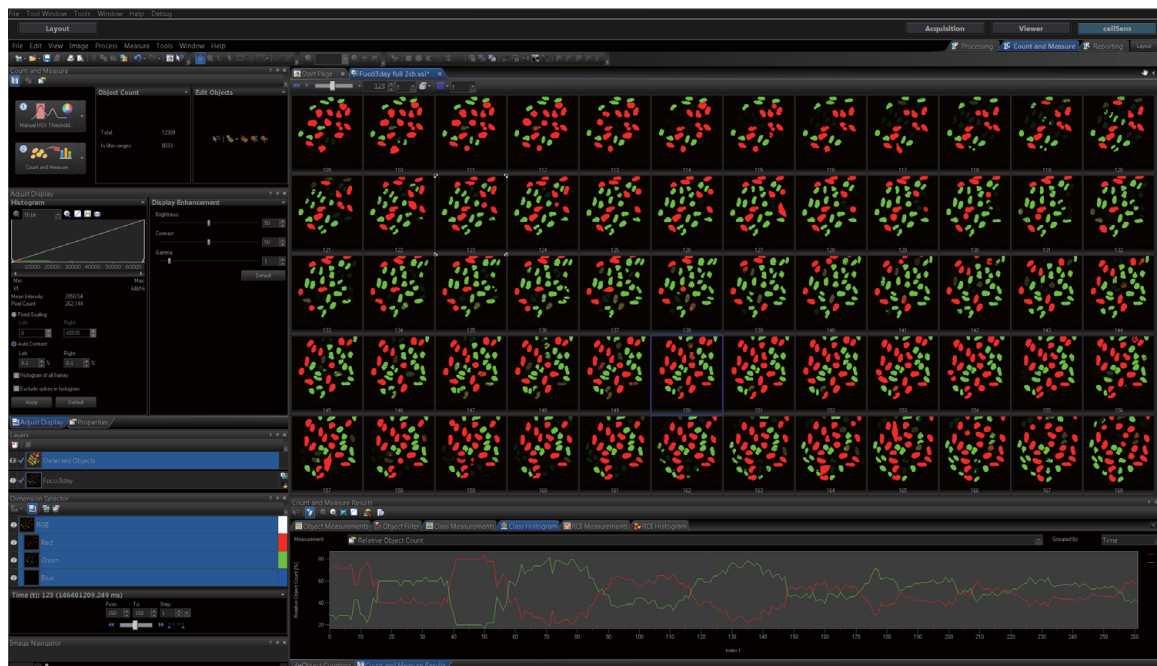
3D time-lapse imaging of mouse embryonic fibroblast labeled with silicone rhodamine docetaxol (Tubulin), imaged with a 100X silicone objective and 30 fps resonant scanning followed by cellSens deconvolution. Image data courtesy of Dr. Markus Delling, Harvard University.



NK-cell mediated cell killing after therapeutic antibody application (blue). GFP labeled NK-cells (green). DAPI uptake marking dead cells (Red). Image data courtesy of Dr. Yuji Mishima, Cancer Chemotherapy Center, Japanese Foundation for Cancer Research.

## Quantification of Images

The FV3000 confocal microscope also includes functions for analyzing and quantifying images. With just a single click, the image measurement macro enables the morphological analysis of cells, automatic cell counting, measurement of the luminosity changes of cells, and automatic cell tracking.



Fucci cell cycle counting and expansion by cellSens. Image data courtesy of Atsushi Miyawaki, Cell Function Dynamics, Brain Science Institute of RIKEN.

# Superior Objectives

## Silicone Immersion Objectives for Live Cell Imaging Deliver High-Resolution Observation at Depth

Olympus offers four high NA silicone immersion objectives that deliver excellent performance for live cell imaging. The refractive index of silicone oil ( $n_e \approx 1.40$ ) is close to that of living tissue ( $n_e \approx 1.38$ ), enabling high-resolution observations deep inside living tissue with minimal spherical aberration caused by refractive index mismatch. Silicone oil does not dry out or harden, so there is never a need to refill oil, making it ideal for extended time-lapse observations.

### UPLSAPO30XS: For a broader view and greater depth

Magnification: 30X, NA: 1.05 (silicone oil immersion), W.D.: 0.8 mm, cover glass thickness: 0.13–0.19 mm, operating temperature: 23–37 °C

### UPLSAPO40XS: For a good balance between field of view and resolution

Magnification: 40X, NA: 1.25 (silicone oil immersion), W.D.: 0.3 mm, cover glass thickness: 0.13–0.19 mm, operating temperature: 23–37 °C

### UPLSAPO60XS2: For 3D observations with superior resolution

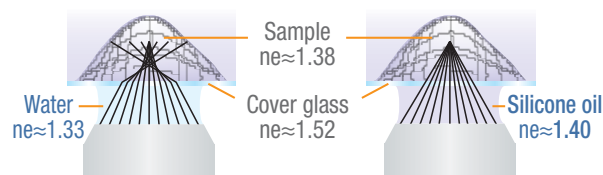
Magnification: 60X, NA: 1.30 (silicone oil immersion), W.D.: 0.3 mm, cover glass thickness: 0.15–0.19 mm, operating temperature: 23–37 °C

### UPLSAPO100XS: For greater brightness at depth in closely defined regions

Magnification: 100X, NA: 1.35 (silicone oil immersion), W.D.: 0.2 mm, cover glass thickness: 0.13–0.19 mm, operating temperature: 23–37 °C

## Refractive Index is Important with Deep Tissue Observation

In deep tissue observation, image quality depends on keeping the refractive index of the sample and immersion medium as close to each other as possible.



### Water immersion objective

When working with a water immersion objective, the difference between the refractive index of the samples and water results in spherical aberration in deep tissue, causing resolution to deteriorate and fluorescence to become dim.

### Silicone immersion objective

When working with a silicone immersion objective, the difference between the refractive index of the samples and silicone oil is minimal, so it achieves brighter fluorescence images with higher resolution for deep tissue observation.

## PLAPON60XOSC2: Enhance the Reliability of Colocalization Analysis with a Low Chromatic Aberration Objective

This oil immersion objective minimizes lateral and axial chromatic aberration in the 405–650 nm spectrum. Colocalization images are acquired reliably and images are measured with superior positional accuracy. The objective also compensates for chromatic aberration through near infrared up to 850 nm, making it the ideal choice for quantitative imaging.



### Low Chromatic Aberration Objective

Magnification: 60X  
NA: 1.4 (oil immersion)  
W.D.: 0.12 mm  
Chromatic aberration compensation range: 405–650 nm  
Optical data provided for each objective.

## Performance Comparison of the PLAPON60XOSC2 and UPLSAPO60XO

	PLAPON 60XOSC2	UPLSAPO 60XO
<b>Axial chromatic aberration (Z direction)</b> Compared for PSF fluorescent beads (405 nm, 633 nm)	Approx. 0 μm	Approx. 0.5 μm
<b>Lateral chromatic aberration (X-Y direction)</b> Compared for PSF fluorescent beads (405 nm, 488 nm, 633 nm)	Approx. 0.1 μm	Approx. 0.2 μm
<b>3D image</b> Tubulin in Ptk2 cells labeled with two colors (405 nm, 635 nm) and compared		

## Reduce Spherical Aberration

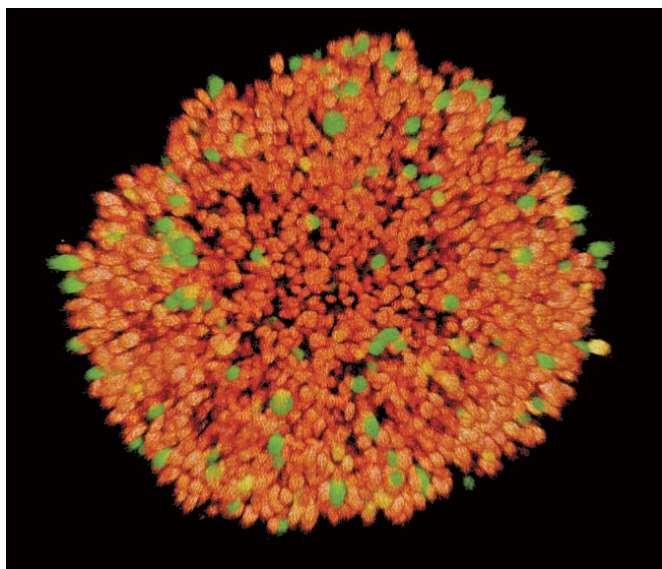
The correction collar adjusts the lens position of objectives to correct the spherical aberration caused by refractive index mismatch, resulting in the improvement of image quality, such as resolution, brightness and contrast. The correction collar is especially necessary for objectives with high NA when they are used for super resolution imaging, because they are greatly affected by spherical aberration. The remote correction collar unit is useful for easy adjustment and improvement of the image quality, and operable on all UIS2 objectives which have a correction collar.



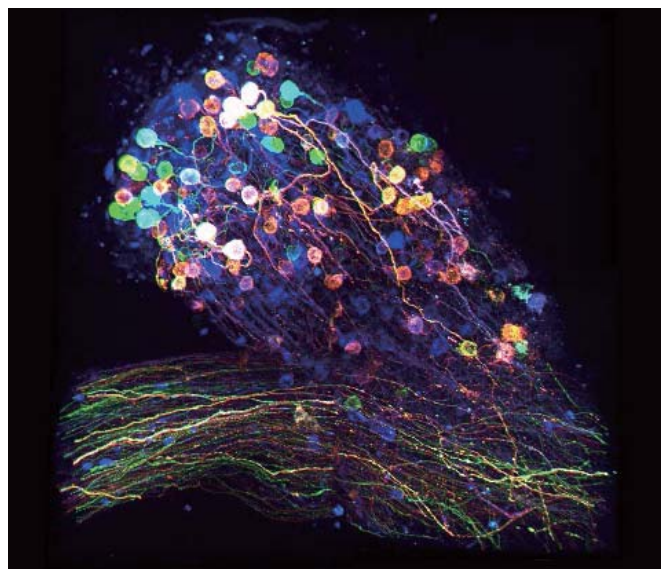


## Application: Deep Tissue Observation with Silicone Objectives

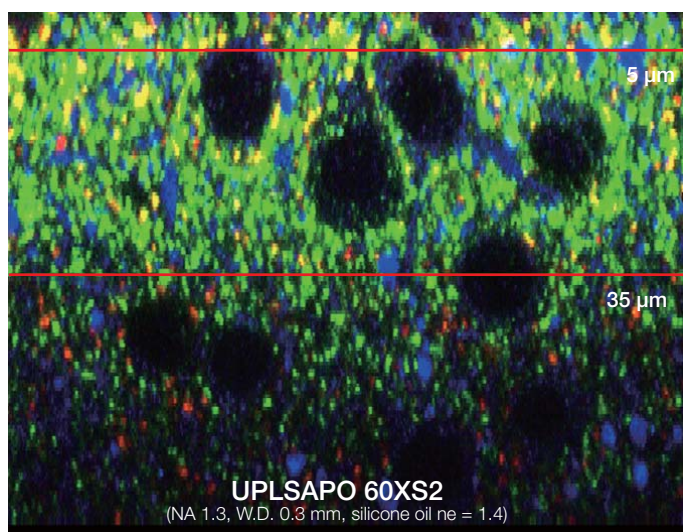
Olympus silicone immersion objectives deliver excellent performance for live cell imaging. The refractive index of silicone oil is close to that of living tissue, enabling high-resolution observation deep inside living tissue with minimal spherical aberration. This refractive index match delivers an ideal focal volume, resulting in perfect volume reconstruction and enabling high resolution confocal imaging of large living organisms. The FV3000's 3D construction software enables simultaneous image acquisition and real-time 3D construction to easily observe 3D structures.



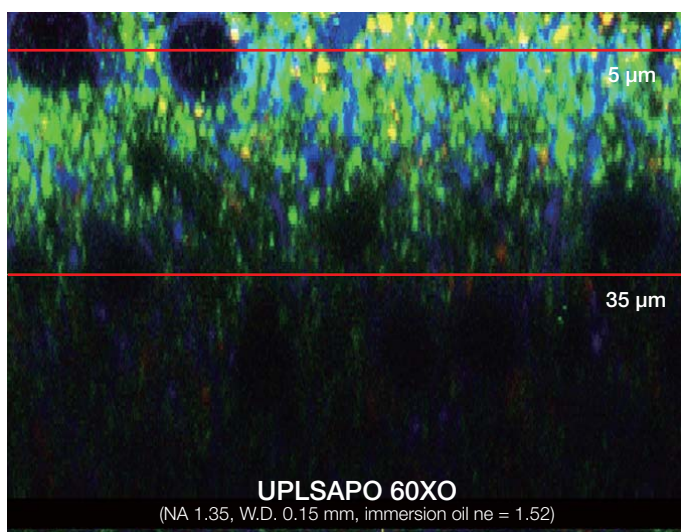
A spheroid image of an NMuMG cell line expressing Fucci2.  
Image data courtesy of Atsushi Miyawaki, Cell Function Dynamics, Brain Science Institute of RIKEN.



3D image of chick ciliary ganglion cleared by tissue clearing reagent.  
Image data courtesy of Dr. Ryo Egawa, Tohoku University Graduate School of Life Science.



ScaleA2-treated neocortex  
Image data courtesy of Motokazu Uchigashima, M.D., Ph.D., Masahiko Watanabe, M.D., Ph.D., Department of Anatomy, Hokkaido University Graduate School of Medicine.



# Choose the Frame That Suits Your Application

## Inverted microscope

- Suitable for observing cells cultured in a vessel.
- The Z-drift compensator enables time-lapse observations that remain in focus.
- Maintain the environmental conditions of cultured cells by adding a stage-top or full enclosure incubator.



## Upright microscope (configured for imaging)

- Optimized for fixed tissue and glass slide specimens.
- Motorized nosepiece precisely maintains focus position.
- Motorized 7-position nosepiece and condenser enable automated transitions from low to high magnification.



## Upright microscope (configured for electrophysiology)

- Ample space around the objectives enables patch-clamp devices to be installed.
- Add additional space by lowering the stage position for experiments that require large sample handling.
- Swing and slider nosepieces are available so objectives can be easily changed without interfering with the patch-clamp set-up.





# Designed for Electrophysiology

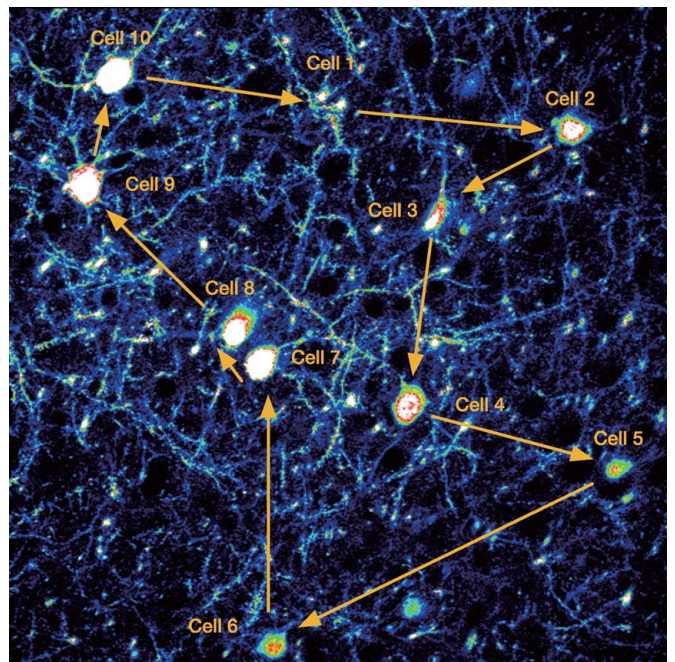
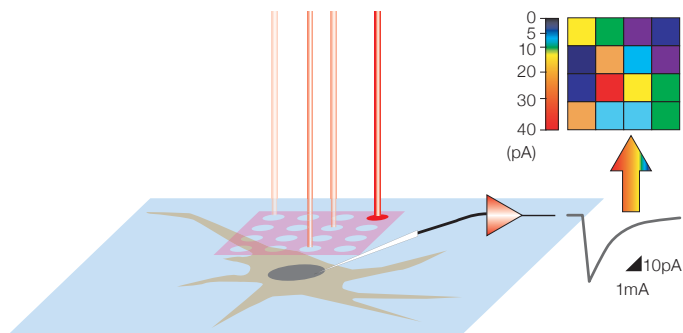
## Frame Design

Modified from the standard upright frame configuration, the large-working space design provides ample room around the objective to easily set up electrophysiology equipment.



## Meets the Needs of Electrophysiological Experiments

Synchronize confocal imaging events with electrophysiology equipment via the trigger signal I/O interface box. The I/O interface box also converts voltage signals into images that can be treated in the same manner as fluorescence images. This enables users to capture voltage-based images in synchrony with photostimulation by the confocal scanner.



## Interface to the Hardware

The analog unit is available as an I/O interface for triggering of external devices.



# Modular Units Designed for Your Applications

## Scanners



### Hybrid Scan Unit (Resonant/Galvanometer)

The hybrid scanner combines the capabilities of a galvanometer scanner with a resonant scanner for high-speed imaging in the full field of view at 30 fps and up to 438 fps at  $512 \times 32$ . The Sequence Manager makes it simple to automatically switch between resonant and galvanometer imaging in the same experiment.

### Galvo Scan Unit

The galvanometer-only scanner provides precision one-way scanning from 1 fps at  $512 \times 512$ , and up to 16 fps with bi-directional scanning and 2x zoom. High-speed multipoint stimulation or detection experiments can travel between multiple cells at over 100 Hz with data output as high as 500 kHz.

## Spectral Detectors



### High Sensitivity Spectral Detector (GaAsP PMT) with TruSpectral Technology

The 2-channel high sensitivity spectral detector (HSD) employs the same volume phase holographic (VPH) technology as the spectral detector (SD), with Peltier cooled GaAsP PMTs and a high quantum efficiency of 45% and detection up to 750 nm. This unit can be combined with the 2-channel SD for a flexible dynamic range or a second 2-channel HSD unit for powerful 4-channel sensitivity.

### Spectral Detector (Multialkali PMT) with TruSpectral Technology

The 2-channel SD employs efficient VPH transmission and an adjustable slit with 1–100 nm bandwidth from 400–800 nm detection. The multialkali PMTs provide a broad dynamic range for detection up to 800 nm.

## Laser Combiners



### Main Laser Combiner

The main laser combiner is the heart of the laser system. The combiner accommodates four standard lasers with an option to add a fifth laser or leave an open port to add an additional three diode lasers via the sub combiner.

### Sub Laser Combiner

Add this optional combiner at any time with up to 3 diode lasers for a maximum of 7 laser lines in combination with the main laser combiner.

## Illumination Units

The conventional illumination modules are designed for long-duration time-lapse experiments. Since light is introduced through fiber delivery systems, no heat is transferred to the microscope.



### Light Source/U-HGLGPS

The pre-centered fluorescence illumination source requires no adjustment and has an average lifespan of 2,000 hours.

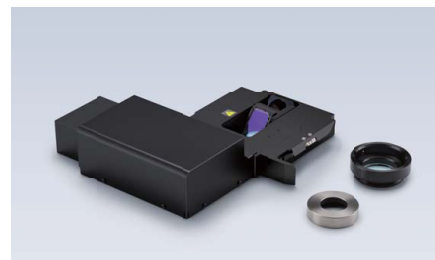


### Transmitted Detector

This unit combines an external transmitted light photomultiplier detector and LED conventional illumination for both laser scanning and conventional transmitted light Nomarski DIC observation. Users can undertake simultaneous multichannel confocal fluorescence imaging and transmitted DIC acquisition.

## Other Equipment

Available upgrade options: field-upgradable laser-based autofocus, fast and precise motorized stage control, analog input/output and TTL synchronization, and a convenient anti-vibration platform.



### Z-Drift Compensator/IX3-ZDC2

The Z-drift compensator uses minimally-phototoxic IR light to identify the location of the sample plane. The IX3-ZDC2 is also compatible with silicone objectives and plastic bottom vessels.



### Ultrasonic Stage for the IX3/IX3-SSU

With low thermal drift for improved accuracy, the ultrasonic stage can be controlled by both software and the Touch Panel Control for fast, reliable multi-area imaging.



### I/O Interface Box

This unit supports electro-physiological experiments through analog inputs and TTL I/O support. The interface box converts voltage to images that can be treated in the same manner as fluorescence images.



### Simple Anti-Vibration Plate

Designed to match the footprint of the FV3000, this simple anti-vibration plate provides a compact solution for those who do not need a full anti-vibration table.



## Specifications

### FLUOVIEW FV3000 Laser Confocal Microscope Specifications

		FV3000	FV3000RS
Laser Light	Violet/Visible Light Laser	405 nm: 50 mW, 488 nm: 20 mW, 561 nm: 20 mW, 640 nm: 40 mW One optional laser port for sub laser combiner or optional laser unit	
Optional Laser	Sub Laser Combiner	Laser as follows (max. 3 laser units ) 445 nm: 75 mW, 514 nm: 40 mW, 594 nm: 20 mW, connected to main laser combiner	
	Single Laser Unit	445 nm: 75 mW, 514 nm: 40 mW, or 594 nm: 20 mW, directly connected to main laser combiner	
Laser Light Control		Main laser combiner with implemented AOTF system, ultra-fast intensity modulation with individual laser lines, additional continuously variable shutter control (0.1%–100%, 0.1% increments) 10% or 100% maximum laser power changer by ND filter	
Scanner	Scanning Method	2 silver-coated galvanometer scanning mirrors	2 silver-coated galvanometer scanning mirrors 1 silver-coated resonant and 1 silver-coated galvanometer scanning mirrors
	Galvanometer Scanner (Normal Imaging)	Scanning Resolution: 64 × 64 to 4096 × 4096 pixels Scanning Speed (One Way): 512 × 512 with 1.1 s – 264 s. pixel time : 2 μs – 1000 μs Scanning Speed (Round Trip): 512 × 512 with 63 ms - 250 ms / 256 × 256 with 16 ms - 125 ms Optical Zoom: 1X – 50X in 0.01X increments Scan Rotation: Free rotation (360 degrees) in steps of 0.1 degree Scanning Mode: PT, XT, XZ, XY, XZT, XYT, XYZ, XYλ, XYZT, XYλT, XYλZ, XYλZT ROI scanning, rectangle clip, ellipse, polygon, free area, line, free line, and point; tornado mode only for stimulation	
	Resonant Scanner (High-Speed Imaging)	–	Scanning Resolution: 512 × 32 to 512 × 512 pixels Scanning Speed: 30 fps at 512 × 512, 438 fps at 512 × 32 Optical Zoom: 1X – 8X in 0.01X increments Scanning Mode: XT, XZ, XY, XZT, XYT, XYZ, XYλ, XYZT, XYλT, XYZ, XYλZT ROI Scanning, Rectangle Clip, Line
	Pinhole	Single motorized pinhole, pinhole diameter ø50 – 800 μm (1 μm steps)	
	Field Number (FN)	18	
	Dichromatic Mirror Turret	8 positions (high-performance DMs and 10/90 mirror)	
	Optional Unit for Scanner	Laser power monitor, optional laser port	
High Sensitivity-Spectral Detector	Detector Module	Cooled GaAsP photomultiplier, 2 channels	
	Spectral Method	Motorized Volume Phase Holographic transmission diffraction grating, motorized adjustable slit, selectable wavelength bandwidth: 1–100 nm, wavelength resolution: 2 nm	
	Dichromatic Mirror Turret	8 positions (high-performance DMs and mirror)	
Spectral Detector	Detector Module	Multi-Alkali photomultiplier, 2 channels	
	Spectral Method	Motorized Volume Phase Holographic transmission diffraction grating, motorized adjustable slit, selectable wavelength bandwidth: 1–100 nm, wavelength resolution: 2 nm	
	Dichromatic Mirror Turret	8 positions (high-performance DMs and mirror)	
System Control	Control Unit	OS: Windows 7 Professional 64-bit (English version), Windows 10 Professional 64-bit built-in dedicated I/F board and hardware sequencer for precise imaging timing	
	Display	30 or 32-inch monitor (WQUXGA 2560 × 1600)	
Fluorescence Illumination Unit		External fluorescence light source, fiber adapter to optical port of scan unit, motorized switching between LSM light path and fluorescence illumination	
Transmitted Light Detector Unit		Module with integrated external transmitted light photomultiplier detector and LED lamp, motorized switching	

### Microscope

	Inverted frame	Upright frame (for imaging)	Upright frame (for electrophysiology)
Microscope Frame	Motorized inverted microscope IX83 (IX83P2ZF)	Motorized fixed stage upright microscope BX63L	
Revolving Nosepiece	Motorized sextuple revolving nosepiece	Motorized septuple revolving nosepiece	Coded swing nosepiece Coded slider nosepiece
Condenser	Motorized long working distance condenser	Motorized luniversal condenser	Motorized long working distance condenser
Focus Stroke	Built-in motorized nosepiece focus Stroke: minimum increment: 0.01 μm		

### Software

Basic Features	GUI designed for darkroom environment. User-arrangeable layout. Acquisition parameter reload features. Hard disk recording capability, adjust laser power and HV with Z-stack acquisition. Z-stack with alpha blending, maximum-intensity projection, iso-surface rendering.
2D Image Display	Each image display: single-channel side-by-side, merge, cropping, live tiling, series (Z/T/λ), LUT: individual color setting, pseudo-color, comment: graphic and text input
3D Visualization and Observation	Interactive volume rendering: volume rendering display, projection display, animation display. 3D animation (maximum intensity projection method, α blending) 3D and 2D sequential operation function
Image Format	OIR image format 8/16-bit gray scale/index color, 24/ 32/ 48-bit color, JPEG/ BMP/ TIFF image functions, Olympus multi-tif format
Spectral Unmixing	Fluorescence spectral unmixing modes (up to 16 channels)
Image Analysis	Region and line measurements, Intensity plot over time/Z, Colocalization analysis
Statistical Processing	2D data histogram display
Optional Software	Motorized-stage control / Mapping and multipoint stimulation / Sequence manager / Virtual channel acquisition / Microplate navigation / Remote development kit / Super resolution imaging (FV-OSR) / Digital camera control function / Deconvolution / FRET&FRAP analysis / Automatic object measurement and classification / Object tracking

**Image data are courtesy of the following institutions:**

5  $\mu\text{m}$  sagittal cryosection of E14.5 mouse embryo (cover and page2), stained with TSA reagents.

Image data courtesy of Dr. Guan Yang and Prof. Xiao Yang, Genetic Laboratory of Development and Diseases, Beijing Institute of Biotechnology, AMMS, China.

3D rendered image of Xenopus endoderm labeled with malachite green and methylene blue. 3 channel image captures label and autofluorescence. (1, page 1)

3D rendered image of Xenopus endoderm labeled with malachite green and methylene blue. (2, page 1)

2  $\times$  2 tiled image of whole rat embryo, 20 mm total field of view. H&E fluorescence with 640 nm laser diode. (3, page 1)

Image data courtesy of Dr. Mike Davidson. Image presented with lasting gratitude for his lifetime commitment to science and microscopy.



- OLYMPUS CORPORATION is ISO14001 certified.
- OLYMPUS CORPORATION is ISO9001 certified.
- Illumination devices for microscope have suggested lifetimes. Periodic inspections are required. Please visit our website for details.

- This product is designed for use in industrial environments for the EMC performance. Using it in a residential environment may affect other equipment in the environment.
- All company and product names are registered trademarks and/or trademarks of their respective owners.
- Images on the PC monitors are simulated.
- Specifications and appearances are subject to change without any notice or obligation on the part of the manufacturer.

[www.olympus-lifescience.com](http://www.olympus-lifescience.com)

**OLYMPUS**<sup>®</sup>

**OLYMPUS CORPORATION**  
Shinjuku Monolith, 2-3-1 Nishi-Shinjuku, Shinjuku-ku, Tokyo 163-0914, Japan