OLYMPUS®

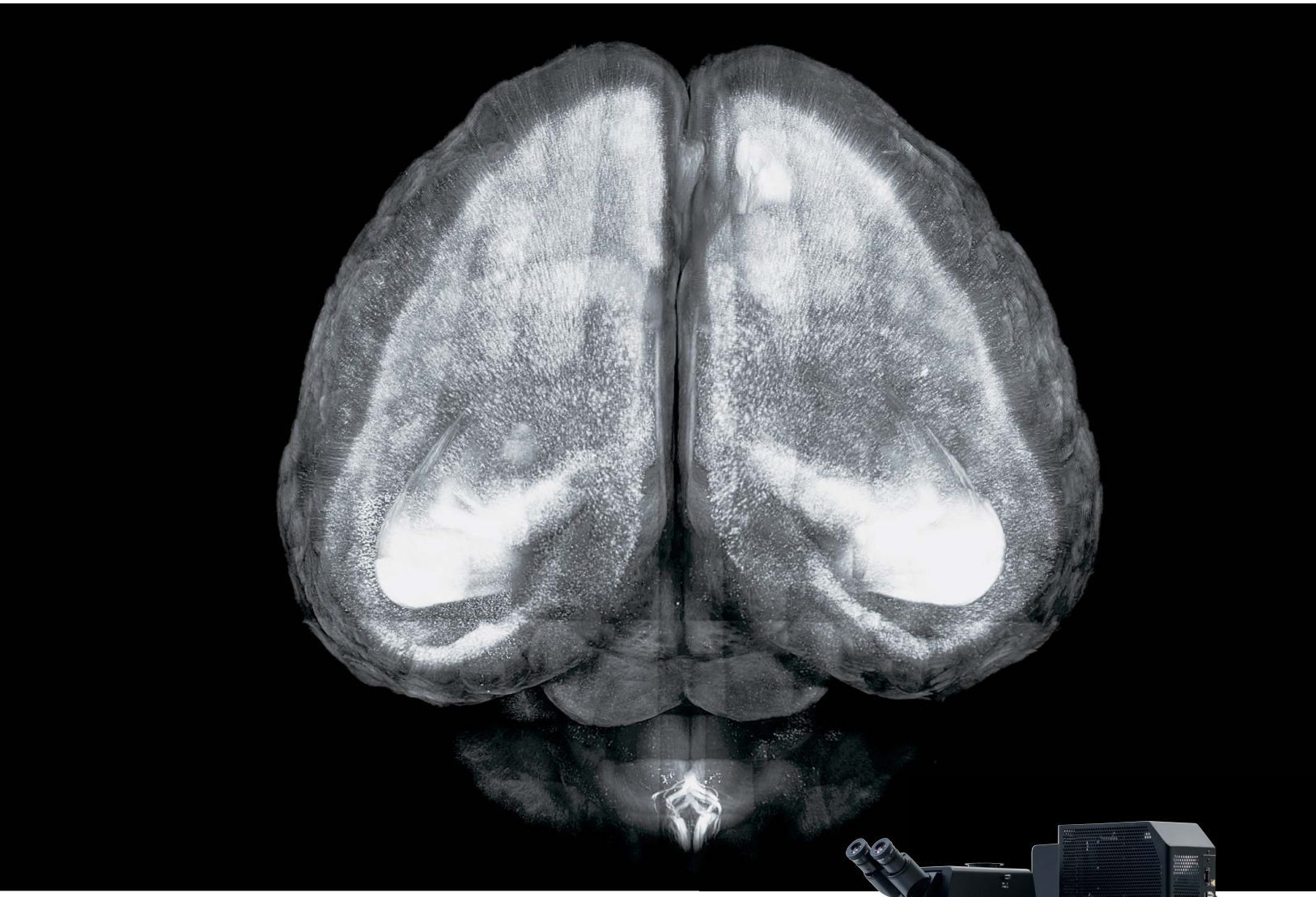
Your Vision, Our Future

Multi Photon Laser Scanning Microscope

FVMPE-RS

FLUOVIEW

Advanced Multi Photon Microscopy with Extended IR Range at High Speed



INNOVATIVE, PRECISE, HIGH SPEED, AND DEEPLY FOCUSED

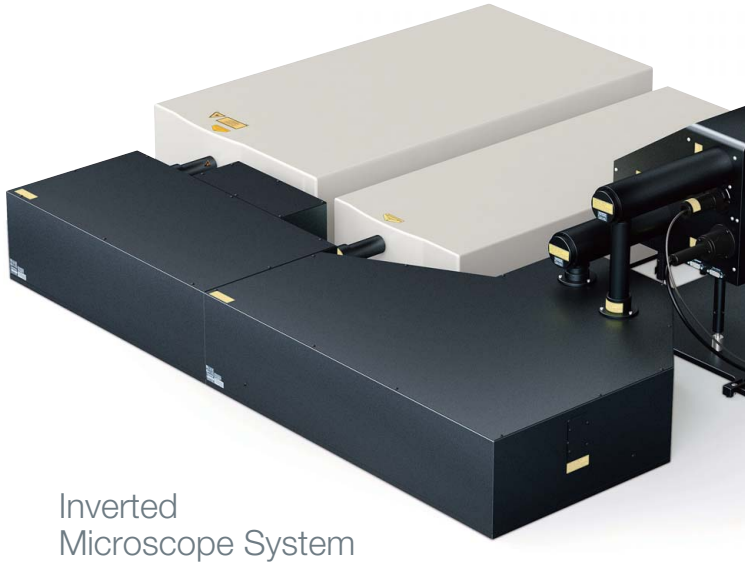
High Speed

Multi-color

Extended IR Range

Laser Light Stimulation

Deep Observation

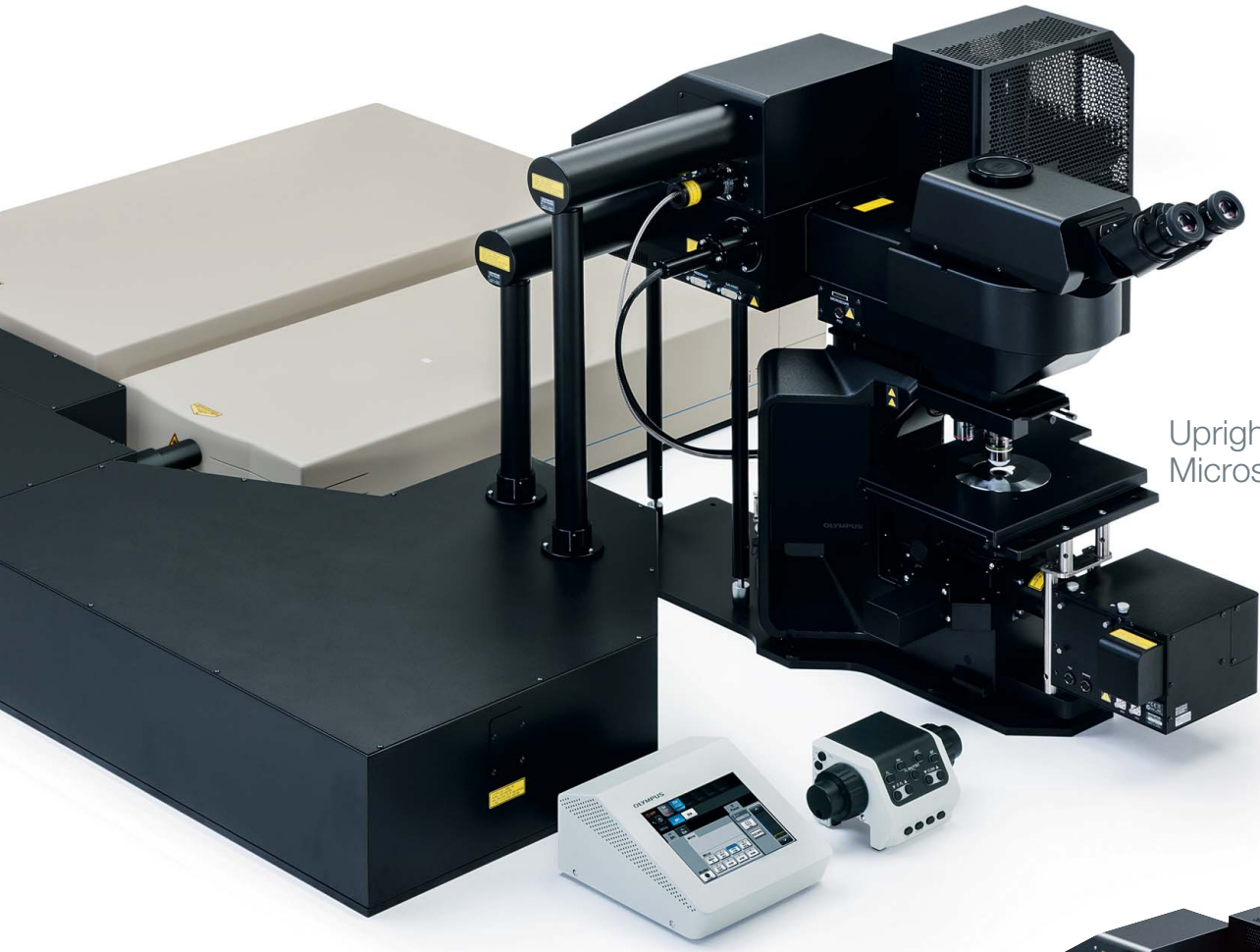


Inverted
Microscope System

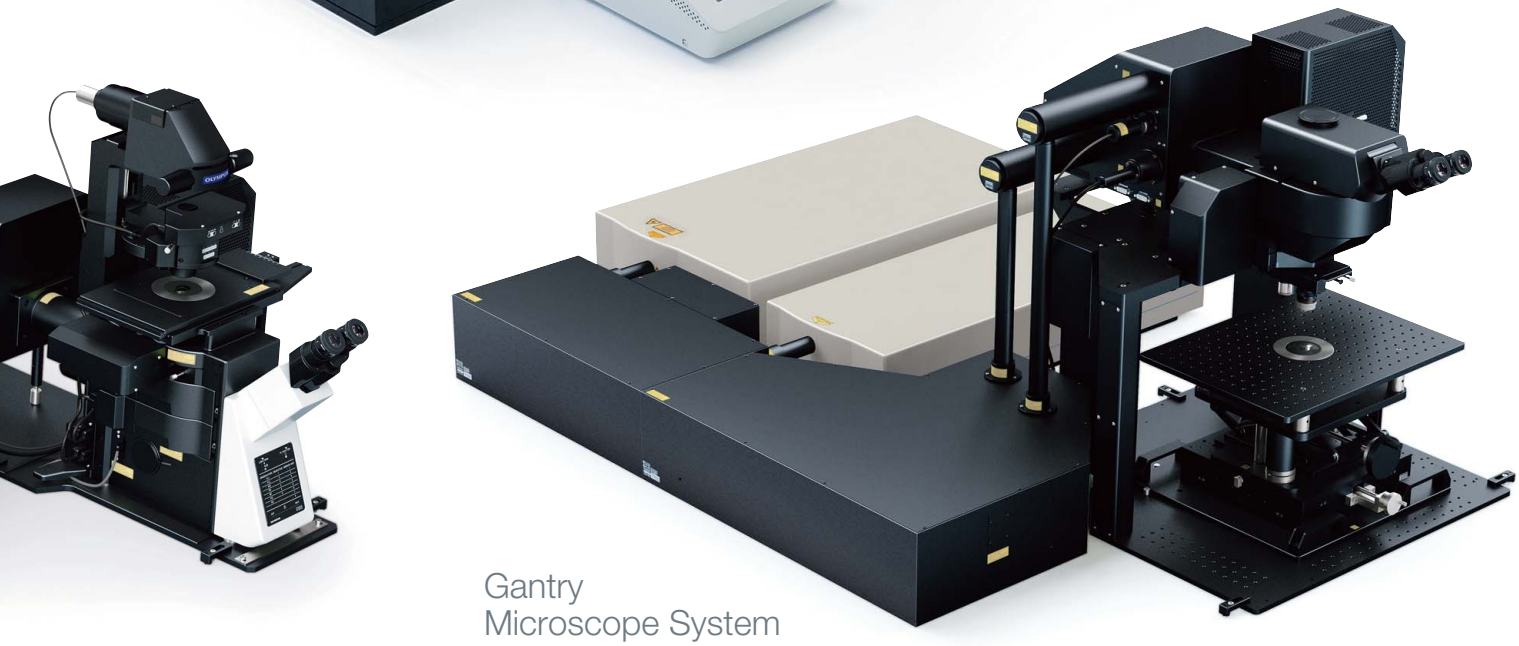
The Olympus FVMPE-RS satisfies a myriad of performance needs for deep observation.

It delivers high speed, millisecond imaging essential for the capture of rapid *in vivo* responses, and offers ideal spot excitation with intense energy — even at deep sites. It also offers high S/N imaging for efficient detection of scattered fluorescence photons, simultaneous dual wavelength excitation at deep sites, visible or multi photon laser light stimulation, and synchronization with patch clamp data.

Put simply, the Olympus FLUOVIEW FVMPE-RS combines high speed, deep observation capability with multi-color imaging and powerful laser light stimulation for the researcher who refuses to compromise.



Upright
Microscope System

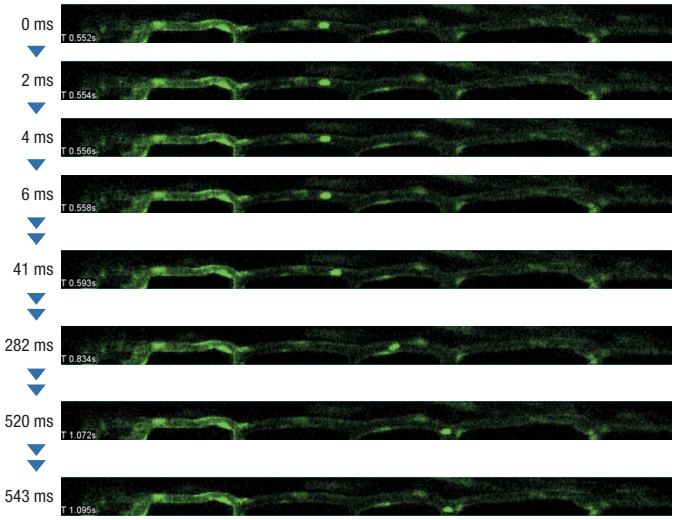


Gantry
Microscope System

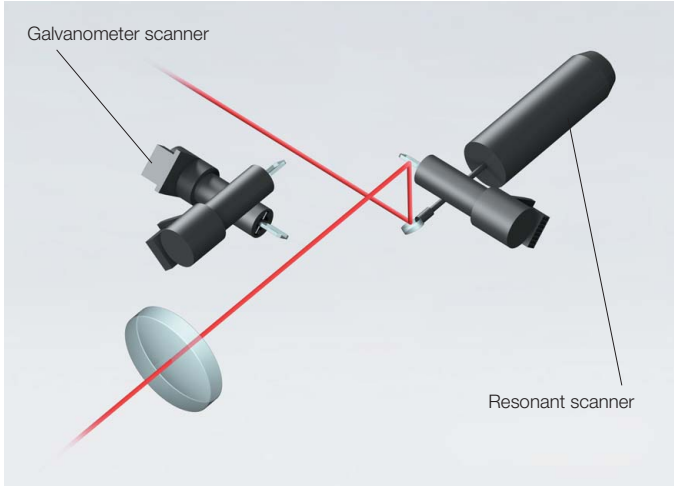
HIGH SPEED SCANNING CAPTURES *IN VIVO* RESPONCES WITH 438 fps

A High Speed Scanner Providing Unique 438 fps at 512 × 32 Scan Performance

The scanner unit combines a newly developed high speed resonant scanner with a conventional galvanometer scanner to provide high speed and high definition imaging in a single system. High speed imaging delivers 30 fps at 512 × 512 at full field of view (FN 18), while clip scans optimize return time to achieve 438 fps at 512 × 32 pixels — making it possible to capture rapid calcium channel activity and membrane potential-sensitive dyes in action.



Images show data acquired with 438 fps frame rate. Fast moving fluorescently labeled cells are captured without distortion in blood vessel of zebrafish.

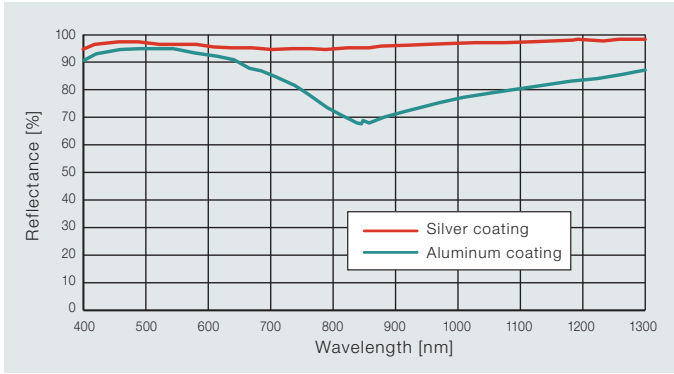


High speed imaging

A Proprietary Silver Coating Improves Excitation Efficiency by 50%*

Silver-coated scanner mirrors achieve extremely high reflectance characteristics across a broad wavelength range from visible to near infrared. Total reflectance for the XY scanner is enhanced by more than 25% in the near-infrared range compared to conventional aluminum coating mirrors, and this increased reflectance provides more than a 50% improvement when converted to multi photon excitation efficiency. The result is a highly effective apparatus that delivers the superior power needed for deep *in vivo* experiments.

*Compared to standard aluminum coating.



A Cooled, High-Sensitivity GaAsP Detector Acquires High S/N Images

High S/N imaging can be acquired even under faint fluorescence through the use of gallium arsenide phosphide (GaAsP) in the photomultiplier tube (PMT) — delivering greater quantum efficiency than multialkali PMT along with Peltier cooling that improves S/N even further.

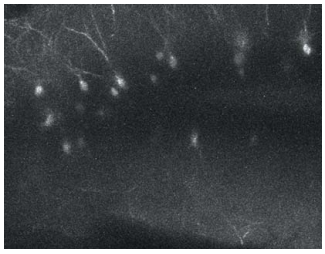


Image multialkali with current detector

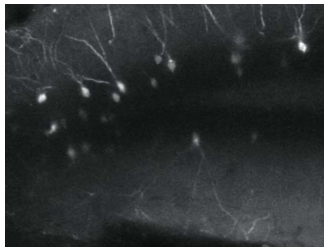
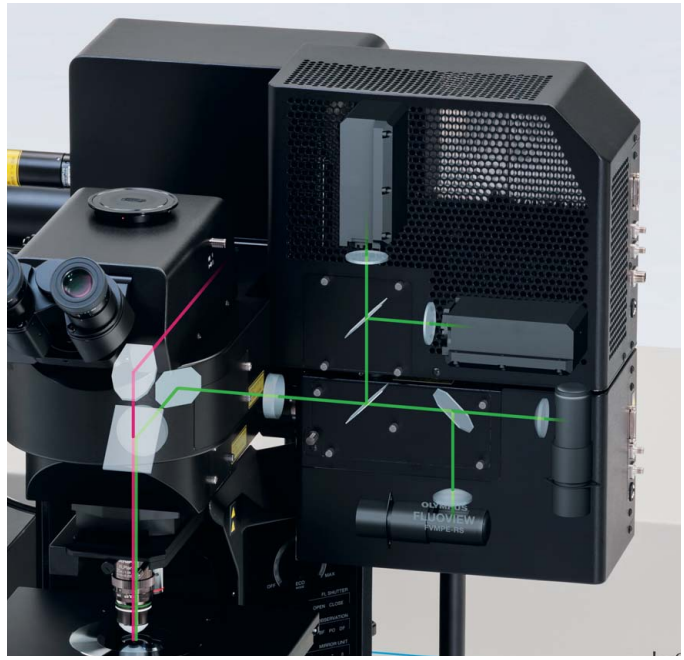


Image captured with GaAsP detector

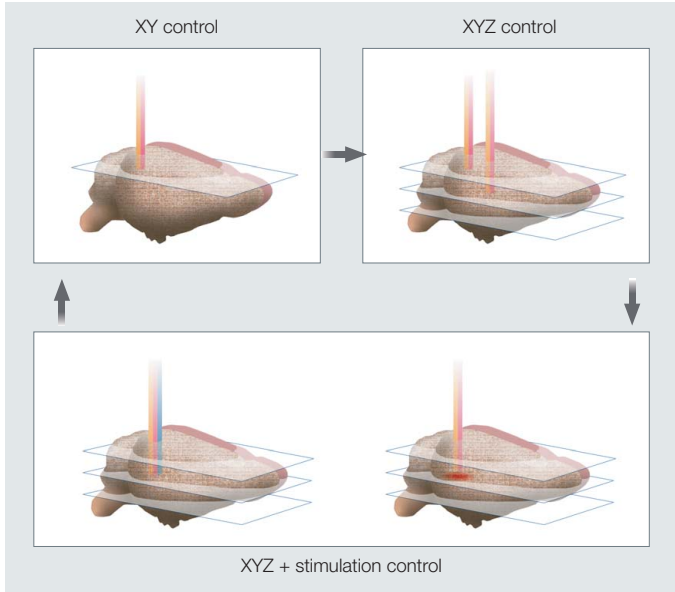
Arc-dVenus transgenic mouse (8-week-old), coronal brain block, hippocampal dentate gyrus
Projection image of 300 — 400 µm depth (5 µm steps)

Image data courtesy of:
Dr. Norio Takata, Dr. Hajime Hirase
Laboratory for Neuron-Glia Circuitry, RIKEN BSI
Dr. Shun Yamaguchi
Gifu University Graduate School of Medicine



Microsecond Timing Precision and Hardware Sequencer Control

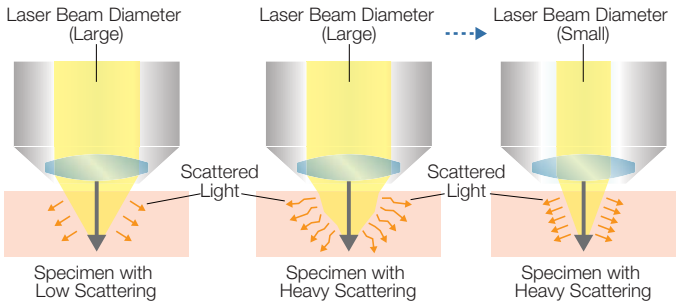
Microsecond repeatability precision provides the power needed for precise control of triggering and stimulation. Microsecond repeatability precision is critical for many applications requiring high speed. This is particularly true for electrophysiology and optogenetic stimulation, where microsecond timing can mean the difference between observing synchronous and asynchronous stimulus responses. For extra long (two-week) acquisitions with complicated experimental procedures that require switching between different imaging tasks, the optional sequence manager can still maintain millisecond precision, ensuring data integrity in the most demanding *in vivo* and *in vitro* experiments.



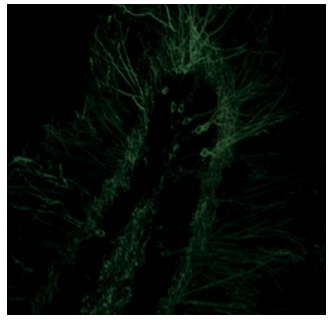
OPTIMIZED FOR DEEP OBSERVATION

Deep Focus Mode Elevates Light-Condensation Performance for Specimens with Heavy Scattering

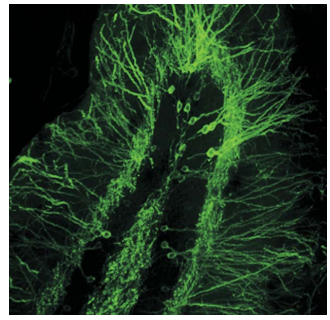
A newly developed Deep Focus Mode responsively adjusts the laser beam diameter in accordance with laser scattering conditions across specimens. For *in vivo* specimens with heavy laser scattering, more excitation photons reach deep sites with the Deep Focus Mode and produces brighter high resolution images.



Both maximum intensity projection from 23 slices, Deep Focus Mode improves image brightness



Normal image



Deep Focus Mode image

Image data courtesy of:
Urs Ziegler and Jose Maria Mateos,
Center for Microscopy and Image Analysis, University Zurich.
Mouse line L15 kindly provided by Pico Caroni, FMI, Basel

Depth-Brightness Compensation Keeps Brightness Consistent from the Surface to Deep Levels

When observing thick specimens, images can often get darker as the focal point goes deeper. But with depth-brightness compensation, detector sensitivity and laser power are constantly adjusted to maintain brightness at a consistent level.

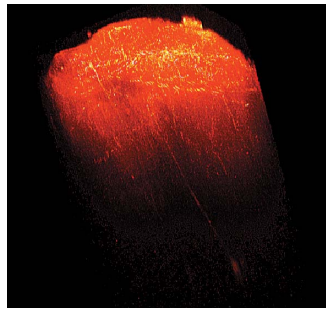


Image without compensation

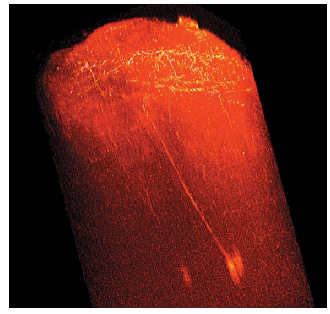
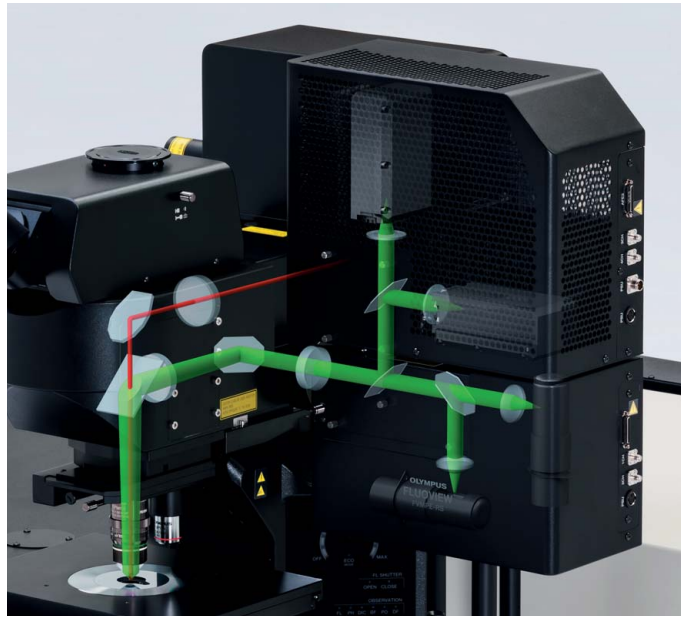


Image with compensation

Detection Light Path Redesigned for More Efficient Fluorescence Capture

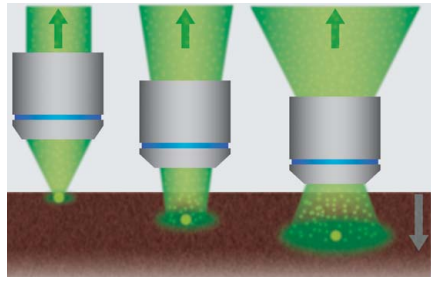
The non-descanned detection light path has been positioned close to the specimen. The signal collecting optics have been enlarged to increase the detection efficiency of scattered fluorescence.



Deep Observation of *In Vivo* and Fixed Transparent Specimens Through Dedicated Multi Photon Objectives with a Maximum Depth of 8 mm

For use in combination with Optical Clearing Agents, the XLSLPLN25XGMP glycerin immersion objective enables the observation of bright, high definition 3D images up to a maximum depth of 8 mm* without harming tissue. Such performance enables observation of a mouse brain extending from the surface to the hippocampus and beyond, for example. A diverse lineup of dedicated multi photon objectives includes the XLPLN10XSVMP that enables more efficient deep observation over a wide field at low magnification and offers compatibility with a wide range of immersion solutions from water to oil.

*When used in combination with the FLUOVIEW FVMPE-RS multi photon excitation laser scanning microscope while using specimen-clearing techniques.



•Wide Field of View

Despite efficient excitation, fluorescence light is scattered deep within the specimen. These wide field of view objectives can collect scattered fluorescence photons to generate brighter images.

Dedicated Multi Photon Objectives	NA	W.D. (mm)	Immersion Index
XLPLN10XSVMP	0.6	8.0	1.33–1.52
XLPLN25XWMP2	1.05	2.0	1.33
XLPLN25XSVMP2	1.00	4.0	1.33–1.40
XLSLPLN25XSVMP2	0.95	8.0	1.33–1.40
XLSLPLN25XGMP	1.00	8.0	1.41–1.52

XLPLN10XSVMP
W.D. 8 mm

XLPLN25XWMP2
W.D. 2 mm

XLPLN25XSVMP2
W.D. 4 mm

XLSLPLN25XSVMP2
W.D. 8 mm

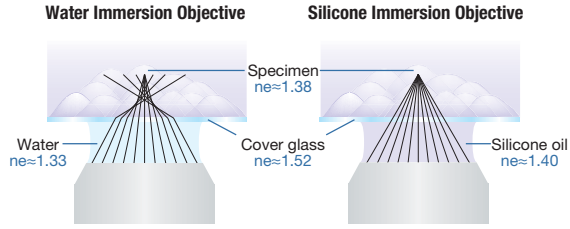
XLSLPLN25XGMP
W.D. 8 mm

Silicone Immersion Objectives for Live Imaging

This immersion objective is designed exclusively for use with silicone oil, which has a refractive index even closer to live cells than that of water. The objective features a large numerical aperture and wide-ranging transmission capability from UV to IR for use in both multi photon and single photon microscopy. Time lapse observations become more reliable and less elaborate, because silicone oil does not dry at 37ºC and its refractive index remains constant. This objective also offers a long working distance to enable observation at deeper tissue levels and across wide field of views. In a nutshell, this silicone objective offers a comprehensive solution for both macro- and deep-tissue observation in the fields of developemental and regenerative science.

Silicone Immersion Objective
UPLSAPO30XSIR
Magnification: 30x
NA: 1.05 (silicone immersion oil)
W.D.: 0.8 mm
Cover glass thickness: 0.13–0.19 mm
Operation temperature: 23ºC–37ºC

Refractive Index is Important with Deep Tissue Observation

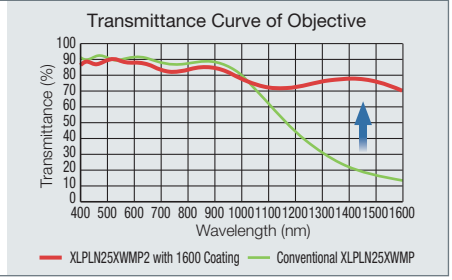


When working with a water immersion objective, the difference between the refractive index of the sample and water results in spherical aberration in deep tissue, causing resolution to deteriorate and fluorescence to become dim.

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

Optics with Outperforming New IR Coating

An innovative IR coating (1600 coating) for the dedicated multi photon objectives and scanner optics further refines deep observation quality. This coating with improved long wavelength transmittance enables excitation without a decrease in laser power, even at deep sites. Since transmittance at 405 nm also remains high, the feature is suited to uncaging applications that employ a 405 nm laser.



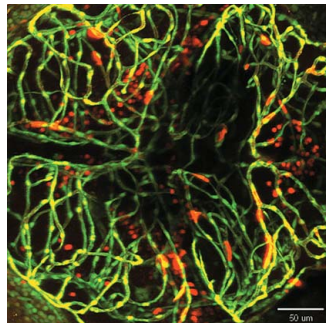
HIGH PRECISION LASER BEAM CONTROL UP TO 1300 nm FOR FLEXIBLE DUAL-LINE MULTI PHOTON IMAGING

Multi Wavelength Excitation and Multi Photon Imaging

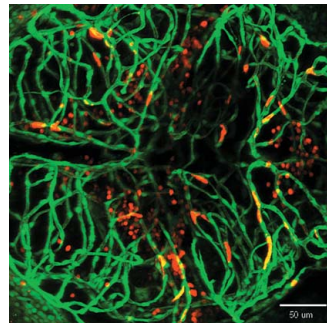
Multichannel, multi photon excitation imaging is accomplished with a dual wavelength IR pulsed laser or two independent IR pulsed lasers — enabling simultaneous excitation of chromophores with different wavelengths. Thanks to the flexible and precise IR introduction optics both lines are accurately merged. Simultaneous excitation provides perfected registration and balanced images for different chromophores. Optimal excitation wavelengths for individual chromophores may also reduce auto fluorescence by avoiding the use of excitation at around 800 nm.

InSight X3 Supports Simultaneous Two Laser Line Excitation and Extended NIR Multi Photon Imaging

The InSight X3 pulsed IR laser systems ideally support multi photon imaging with excitation from 680–1300 nm. The Dual-line version of the InSight X3 system offers two laser beam outputs: main output with a tunable line from 680–1300 nm and the second output at 1045 nm. Higher laser power beyond 1000 nm provides a host of new multi photon imaging capabilities, covering a variety of dyes, fluorescence proteins, and Third Harmonic Generation imaging without UV damage.



Crosstalk occurred after simultaneous excitation of GFP and DS-RED with a single wavelength IR pulsed laser (950 nm).

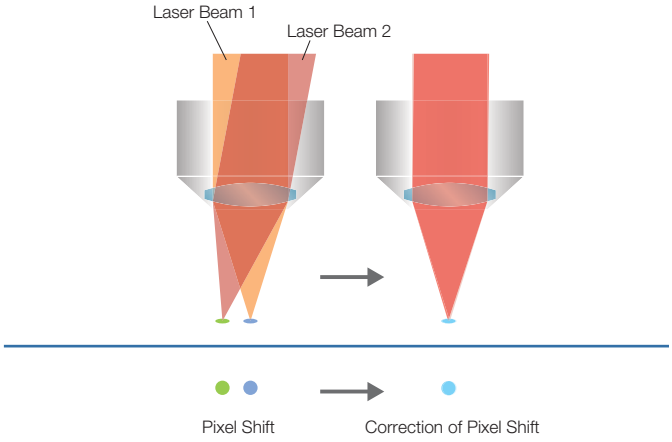


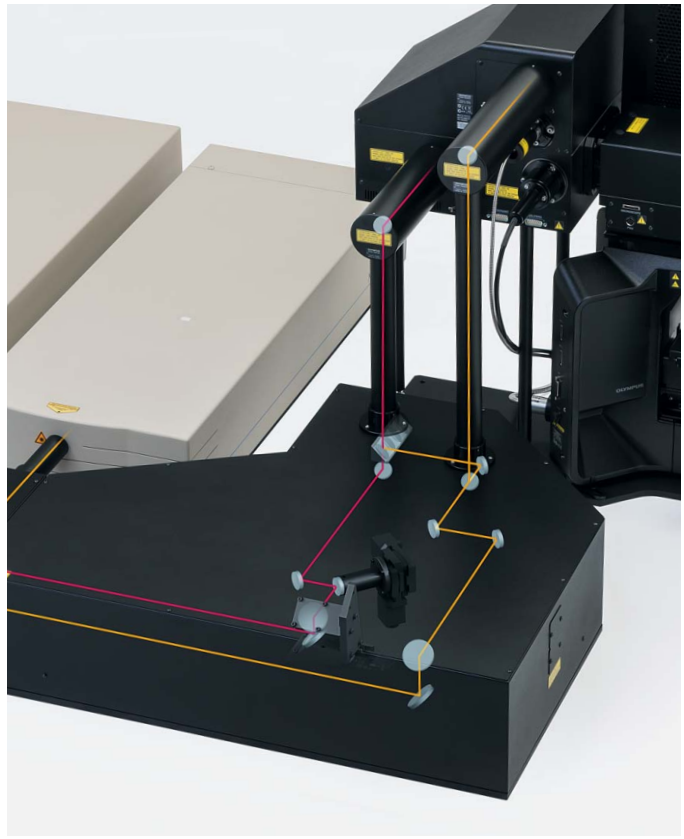
Two fluorescence proteins are clearly separated after individual excitation of GFP and DS-RED with a dual wavelength IR pulsed laser (950 nm/1040 nm).

Image data courtesy of:
Director Naoki Mochizuki
Department of Cell Biology, National Cerebral and Cardiovascular Center

Auto Correct for Laser Beam Misalignment and Pixel Shift with Quadralign 4 Axis Auto Alignment

Multi-color multi photon laser acquisition provides optimized excitation of different fluorophores, reducing channel crosstalk and photobleaching due to the need to choose a suboptimal middle wavelength for excitation. To ensure proper colocalization of fluorescent signals, the Quadralign 4 axis auto alignment is incorporated into 2 horizontal and 2 angular axes per laser line, and single click compensation is enabled for laser beam position as well as incident laser angle — a common cause of pixel shift. Saving time and effort, this auto alignment mechanism tunes the optical axes of the lasers to the laser wavelength used during multi-color excitation. Software-based fine-tuning is also available.

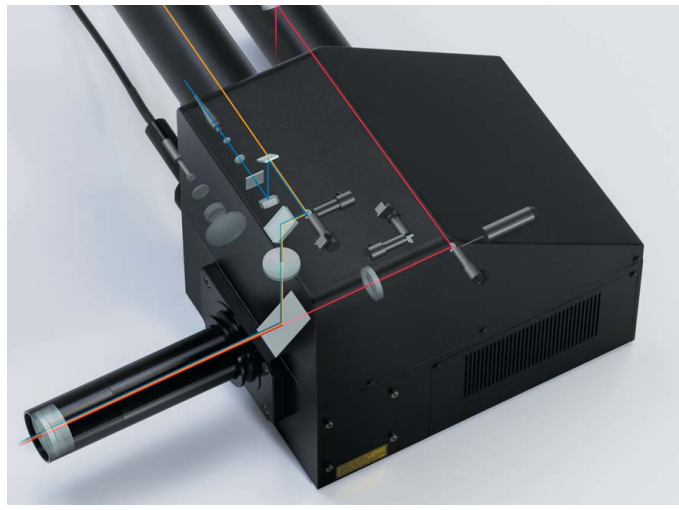




TOOLS FOR ADVANCED APPLICATIONS

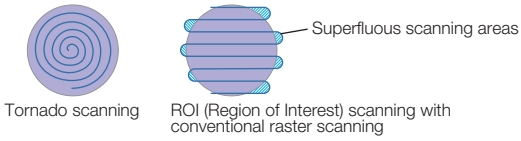
Light Stimulation SIM Scanner from the Visible to IR Range

A laser light stimulation scanner can be installed separately — enabling optogenetics laser light stimulation of channel rhodopsin (ChR2) and halorhodopsin (NpHR) with simultaneous realtime imaging of neural cell activity with a visible or IR range laser.



•Wide Choice of Scan Modes

The FVMPE-RS comes with AOM as standard and provides fine position and time control of imaging and light stimulation. Using Olympus' proprietary tornado scanning allows rapid bleaching and laser light stimulation of desired fields in experiments.

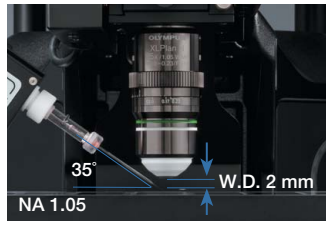


Tornado scanning

ROI (Region of Interest) scanning with conventional raster scanning

Analog Unit Synchronizes Electro-Physiological Data and Laser Light Stimulation

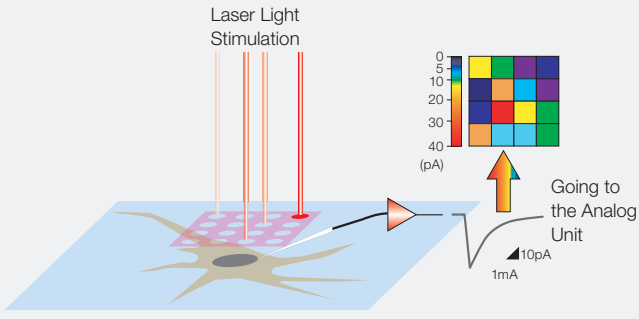
Electro-physiological experiments are enabled through analog inputs and TTL I/O support. The analog unit converts voltage to images that can be treated in the same manner as fluorescent images — enabling light stimulated electrical signals measured with patch clamps to be synchronized with image capture and displayed as a pseudo color intensity overlay.



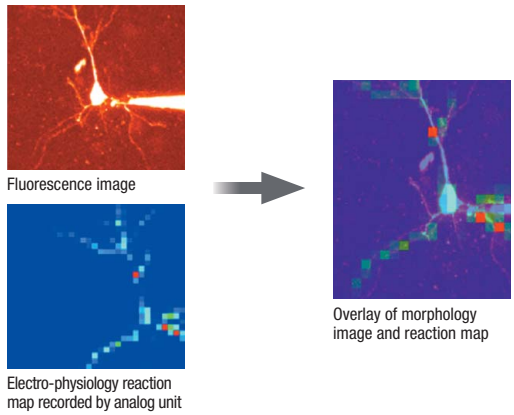
3D Mapping Stimulation Creates Reaction Maps Based on Multiple Coordinates

Highly targeted laser light stimulation is achieved through dividing the observation domain into a grid and laser irradiating each specific area in a software-controlled sequence while eliminating adjacent areas from stimulation. The Z-position setting is available to enable stimulation at a depth different from that of the imaging layer. Changes in intensity during stimulation can also be mapped to the image and reaction maps can be created for multiple coordinates.

A Typical Combination of Laser Light Stimulation and an Analog Unit



*Optional highly rigid piezo nosepiece is necessary for 3D mapping stimulation.



*Data acquired with equivalent function in FV1000

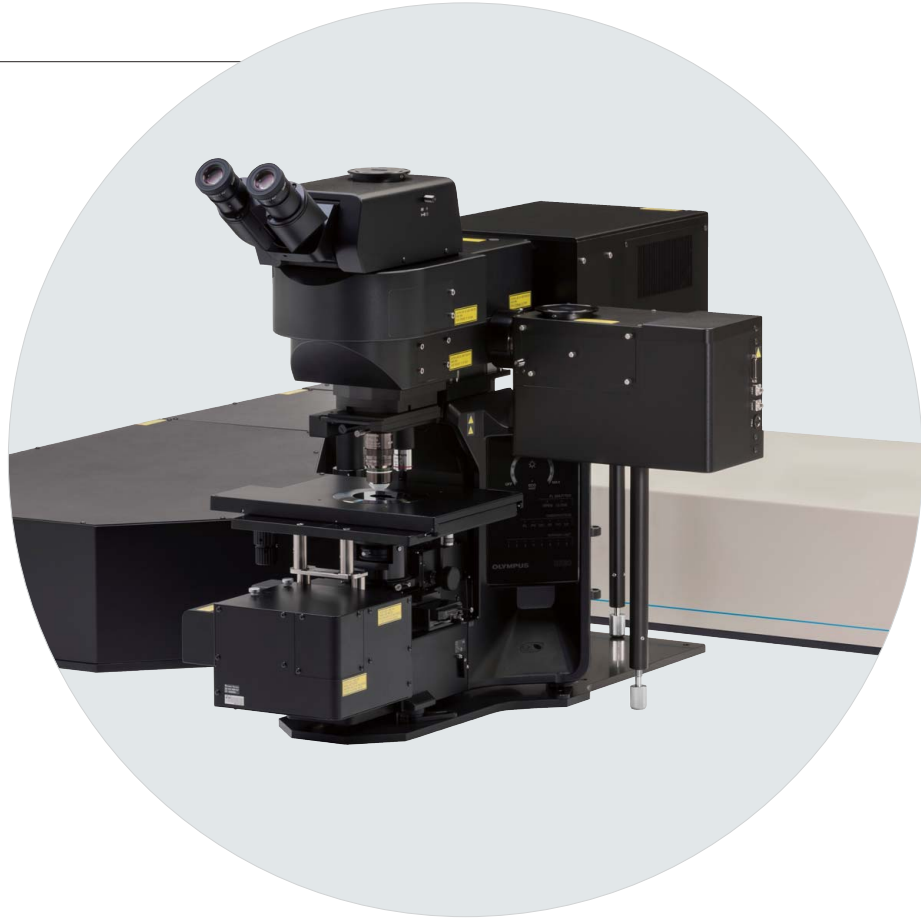
Image data courtesy of:
Haruo Kasai
Center for Disease Biology and Integrative Medicine, Faculty of Medicine, The University of Tokyo

A VARIED LINEUP OFFERING HIGH FLEXIBILITY AND INNOVATIVE IR LASER BEAM CONTROL

Dedicated to Multi Photon Microscopy Tasks

Upright Microscope System

- Non-descanned detectors are very sensitive towards any interfering light. The black microscope frame reduces undesired light reflections.
- The big focus stroke accommodates a wide range of specimens, from slices to mice and other small animals.
- The optional transmitted fluorescence light detector expands your system's capabilities in multi photon imaging. The dedicated high NA condenser detects transmitted fluorescence as well as transmitted second- and third-harmonic generation (SHG and THG) signals.
- The new double deck design motorized fluorescence illuminator suppresses vibration to a minimum during mirror unit exchange. Observation conditions (i.e. multi photon observation, fluorescence observation) can also be switched with minimal effort, even during simultaneous patch clamp experiments.



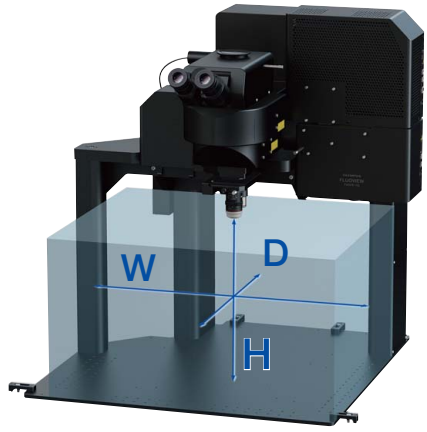
Optional Detecter

Microscope System	Cooled GaAsP PMT	Transmitted Fluorescence	Transmitted Brightfield
Upright	✓	✓	✓
Gantry	✓	–	–
Inverted	✓	–	✓

For *in vivo* Observation Requiring Maximum Space

Gantry Microscope System

- A removable manual XYZ stage enables height adjustments. Changing between thin sample and whole animal imaging can be easily accomplished.
- A large workspace is preserved beneath the objective, while the Gantry frame enables self-constructed experimental equipment to be ideally positioned for imaging.



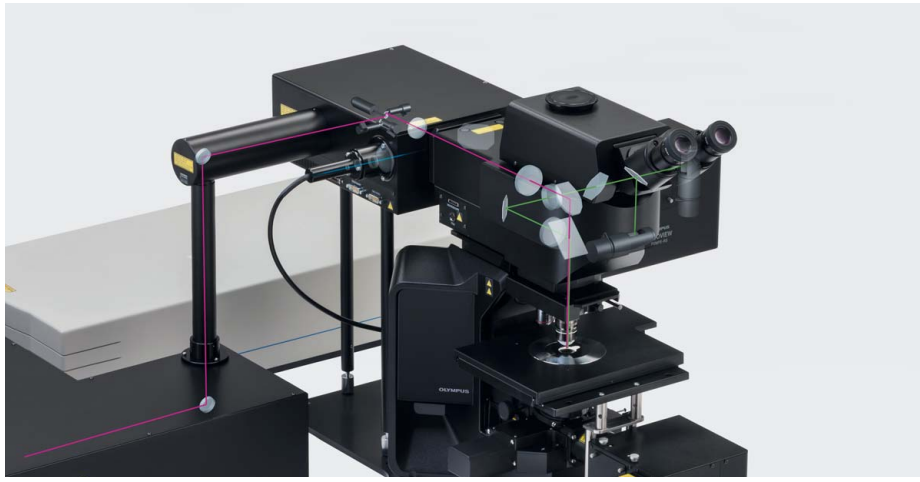
640 (W) x **355 (H)** x **520 (D)** mm

For Time Lapse Observation of Thick Cultivated Cells such as Tissue Cultures, Three-Dimensional Cultures, and Cell Cultures (Spheroid), as well as Intravital Time Lapse Observation of Organs and Tissues through a Body Window

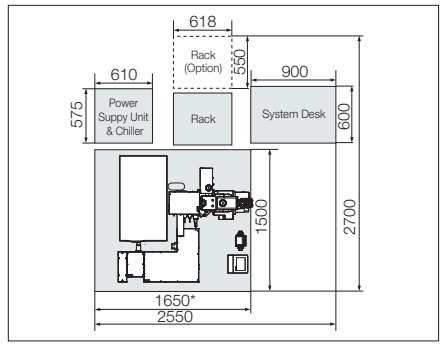
Inverted Microscope System

- An inverted frame ideally supports observations of 3D cultures and multi-cellular clusters (spheroids), which were difficult to manipulate with upright frames.
- The optical performance of the IX83 fully automated high-end research microscope was optimized to efficiently collect scattered fluorescence light. Non-descanned detection performance is improved as compared to conventional inverted multi photon microscope systems.

One Laser System

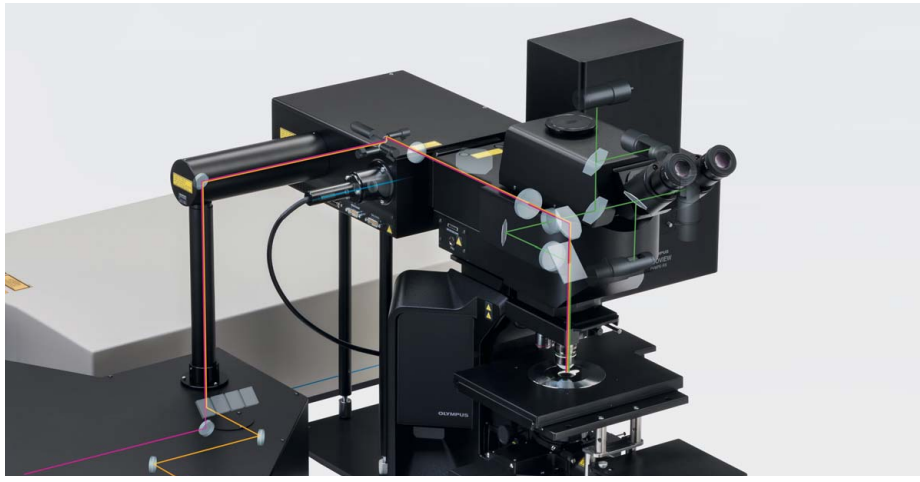


This streamlined system uses a single multi photon IR laser for imaging. SIM scanner for visible laser light stimulation is optional.

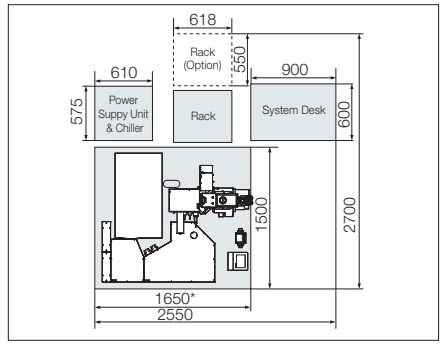


*1800 mm with Inverted Microscope System

Dual Lines System

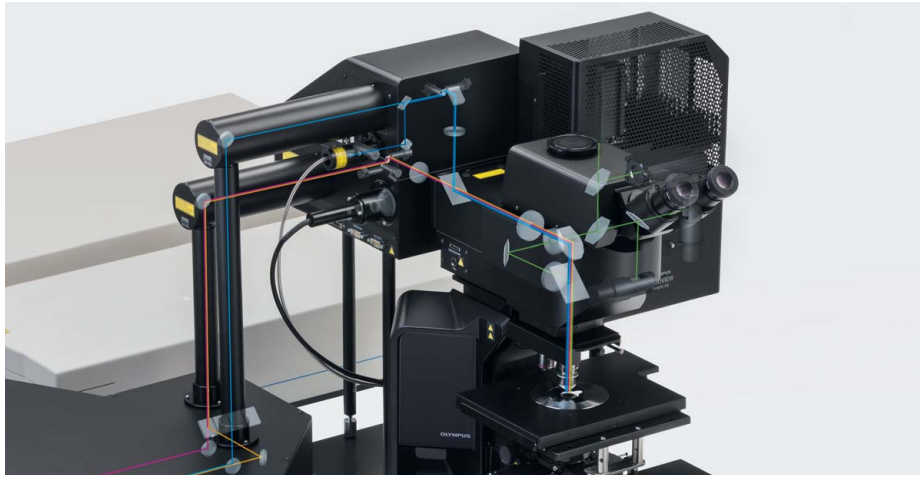


Employing the InSight X3 Dual laser, this system supplies dual wavelengths for multi photon, multi-color imaging. SIM scanner for simultaneous laser light stimulation is also optional.

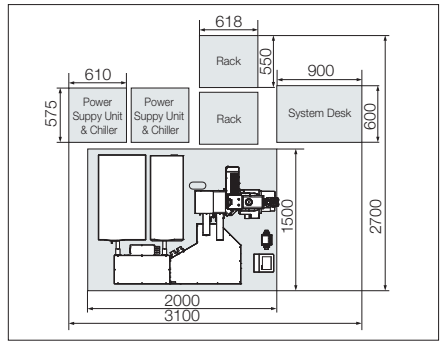


*1800 mm with Inverted Microscope System

Twin Lasers System (with SIM Scanner)



This system employs two multi photon IR lasers for imaging. In addition to multi photon, multi-color imaging, simultaneous laser light stimulation is also supported in combination with an optional SIM scanner.



Lasers Adapted for a Range of Multi Photon Configurations

InSight X3 Dual-OL enables dual wavelength simultaneous imaging for deep observation — with a high peak power with short 120 fs pulse widths, a continuously variable broadband range from 680 nm to 1300 nm, and a fixed wavelength of 1045 nm. A broad selection of dedicated models is available to make the most of multi photon performance, including the MAITAI-OL (Spectra-Physics).

Spectra-Physics

Model	Wavelength covered
MAITAI HPDS-OL	690 nm — 1040 nm
MAITAI eHPDS-OL	690 nm — 1045 nm
INSIGHT X3-OL	680 nm — 1300 nm
INSIGHT X3 DUAL-OL	680 nm — 1300 nm
INSIGHT X3 DUALC-OL	1045 nm (fixed)

Visible Beam Combiner for Laser Light Stimulation

The laser combiner allows solid-state laser combinations for laser light stimulation at wavelengths of 405 nm, 458 nm, and 588 nm.

Light Guide Illumination Source U-HGLGPS

This light guide illumination source is equipped with a liquid light guide that minimizes the impact of vibration and lamp heat on the microscope and specimens alike. Employing a high pressure mercury bulb, the light source offers a durable average lifetime of 2000 hours.

Transmitted Non-Descanned Light Detector

A high NA condenser and transmitted non-descanned light detector for multi photon imaging detect fluorescence emitted from the focal plane and light scattered within the specimen.

Multialkali PMT 2CH Detector

Basic configuration of multialkali PMT 2CH.

Multialkali PMT 2CH + 2CH Detector

Multialkali PMT 2CH and optional addition of multialkali PMT 2CH.

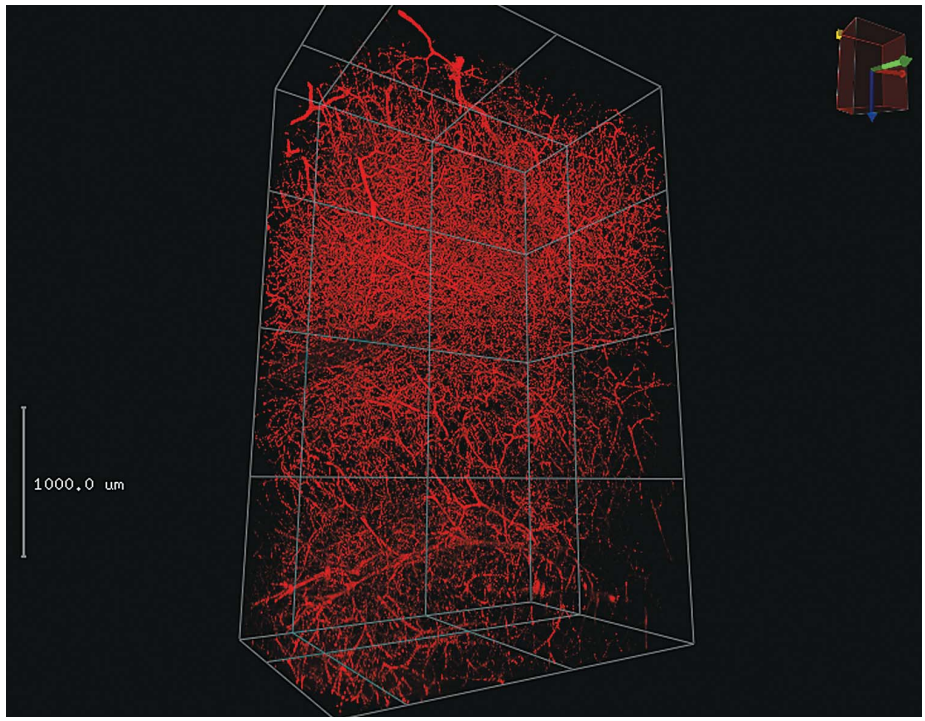
Multialkali PMT 2CH + Cooled GaAsP PMT Detector

Multialkali PMT 2CH and optional cooled GaAsP PMT 2CH in combination.

STREAMLINED SOFTWARE FOR MULTI PHOTON IMAGING

Software Architecture Supports Massive Data Needs

Smooth, 3D rendered display is possible for massive Z-stack data comprising high definition images captured from the sample surface to deep sites. Key frame registration is also available, making it easy to create animated views of 3D images that zoom and transition to different camera angles.

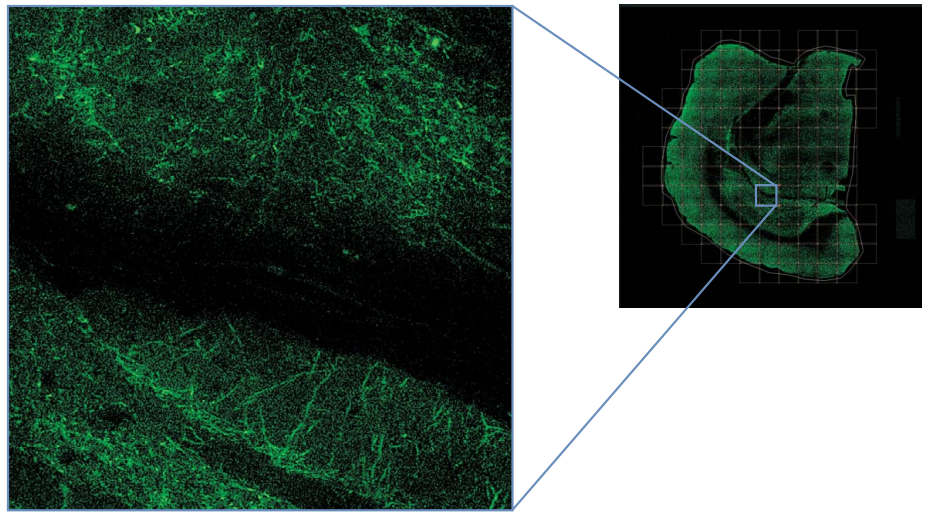


4 mm 3D Stack on Blood Vessel Label with Texas Red in Mouse Brain

Image data courtesy of:
Hiroshi Hama, Rie Ito, Atsushi Miyawaki
Laboratory for Cell Function Dynamics, RIKEN Brain Science Institute

Tiling Significantly Extends the Imaging Range

The tiling function scans multiple adjacent views and stitches them together to build a large image beyond the physical field of view. Use of a motorized stage supports tiling for an even wider field of view, while the mapping feature makes it easy to locate a specific cellular position within the resulting large image.



Using the Map Function with Motorized Stage, Finding Target Field of View is Easy

Image data courtesy of:
Urs Ziegler and Jose Maria Mateos
Center for Microscopy and Image Analysis, University Zurich
Mouse line L15 kindly provided by Pico Caroni, FMI, Basel

Specifications

FLUOVIEW FVMPE-RS SPECIFICATIONS

		One Laser System	Dual Lines System	Twin Lasers System
Unit	Qualified IR pulsed lasers with negative chirp for multi photon excitation	· Spectra-Physics products : MAITAI HPDS-OL: 690 nm — 1040 nm MAITAI eHPDS-OL: 690 nm — 1040 nm INSIGHT X3-OL: 680 nm — 1300 nm INSIGHT X3 DUAL/DUALC-OL: 680nm — 1300nm + 1045nm		
	Main IR pulsed laser	MAITAI HPDS-OL MAITAI eHPDS-OL INSIGHT X3-OL	INSIGHT X3 DUAL-OL INSIGHT X3 DUALC-OL	MAITAI HPDS-OL MAITAI eHPDS-OL INSIGHT X3-OL
	Additional IR line/ Laser: Use as second imaging line/ laser or for simultaneous stimulation (optional SIM scanner)		1045 nm fixed line from INSIGHT X3 DUAL/DUALC-OL	MAITAI HPDS-OL MAITAI eHPDS-OL
	Automatic Introduction Optic	Introduction optic with AOM attenuation (0% — 100%, 0.1% increment) Including fully automated beam expander, XY shifter and two axes angle alignment. (4 Axes Quadralign Auto Alignment optic) Direct coupling to laser port of scanning unit.	Introduction optic with 2 sets of AOM attenuation (0% — 100%, 0.1% increment) Including 2 sets of fully automated beam expander, XY shifter and two axes angle alignment. (4 Axes Quadralign Auto Alignment optic) Direct coupling to laser port of scanning unit.	
	IR laser combining optic		Motorized light path switcher, with DM900, DM1000R, DM1100 to combine two IR wavelength for imaging.	
	Optional Visible light laser for stimulation	405 nm/50 mW, 458 nm/20 mW, 588 nm/20 mW laser source with AOTF attenuation. 0% — 100%, 0.1% increment, < 2 µs rising time		
Scanning Unit	Scanning Method	Light deflection via 2 silver-coated galvanometer scanning mirrors, or silver-coated resonant scanning mirror.		
	Scanning Speed	Galvanometer Scanner (Normal Imaging) : 512 × 512 with 1.1 s — 264 s. Pixel time : 2 µs — 1000 µs. Resonant Scanner (High Speed Imaging) : 30 fps at 512 × 512, 438 fps at 512 × 32		
	Scanning Mode	XY, XYZ, XYT, XYZT, free line, XZ, XT, XZT, PointT		
	Galvanometer Scanner (Normal Imaging)	Galvanometer ROI scanning: Rectangle Clip, Ellipse, Polygon, Free Area, Line, FreeLine & Point. Zoom: 1.0× — 50.0× with 0.01× increment, support 0° — 360° rotation and pan Scanning Field Number: 18 Image Size: 64 × 64 — 4096 × 4096		
	Resonant Scanner (High Speed Imaging)	Resonant ROI scanning: Rectangle Clip, Line. Zoom: 1.0× — 8.0× with 0.01× increment Scanning Field Number: 18 Image Size: 512 × 512		
	Optical Coating	IR support optic with 1600 Coating.		
	Non Descanned MPE imaging detectors	Reflected detection: 2 or 4 channel configuration: 2 PMTs configuration, 4 PMTs configuration or 2 PMTs + 2 cooled GaAsP-PMTs Transmitted detection: 2 PMTs unit with high NA condenser.		
	Transmitted light detector	Module with integrated external transmitted light photomultiplier detector and 100 W Halogen lamp, motorized switching, fiber adaptation to microscope frame		
	Z-Drive	Integrated motorized focus module of the microscope, minimum increment 0.01 µm Optional: highly rigid piezo nosepiece.		
	Control Unit	OS: Microsoft Windows 7 Professional 64bit Hardware sequencer for highly precise timing repeatability control		
	Optional Simultaneous Stimulation Scanner	Highly synchronized simultaneous stimulation scanner, including a set of Galvanometer Scanner, VIS and IR laser port. ROI scanning: Rectangle Clip, Ellipse, Polygon, Tornado, Free Area, Line, FreeLine & Point.		
	Optional analog and digital in out box	4 channels analog signal input, 6 channels digital TTL trigger input, 5 channel digital TTL trigger output. Scanner timing output		
Operation Environment		Room temperature: 20 — 25°C, humidity: 75% or less at 25°C, requires continuous (24-hour) power supply		
Size of Anti-vibration table		1500 mm × 1650* mm *1800 mm with Inverted Microscope System	1500 mm × 1650* mm *1800 mm with Inverted Microscope System	1500 mm × 2000 mm
Software	Basic Feature	Dark room matching GUI design. 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		
	IR laser controlling	Fully integrated IR laser wavelength control and Deep Focus Mode		
	Optional Motorized Stage software	XY motorized stage control, Map image acquisition for easy target locating. Tiling acquisition and software image stitching. Define multiple area for time lapse imaging.		
	Optional Mapping and Multiple point stimulation software	Multiple point stimulation and data acquisition software. Mapping multiple point stimulation to generate reaction map. Filtering feature to select points. Multiple point stimulation. Single or repeat stimulation. Each point independent stimulation wavelength selection.		
	Optional Sequence Manager	Advanced programmable software to define multiple imaging/stimulation tasks and execute by hardware sequencer. Minimum gap 100 ms delay between tasks.		

Image data on cover page: YFP-H mouse Brain of 20 weeks old treated by New Sca/e
Courtesy of:
Hiroshi Hama, Atsushi Miyawaki
Laboratory for Cell Function Dynamics RIKEN Brain Science Institute

- **OLYMPUS CORPORATION is ISO14001 certified.**
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Printed in Japan M1780E-072017