OLYMPUS®

Your Vision, Our Future
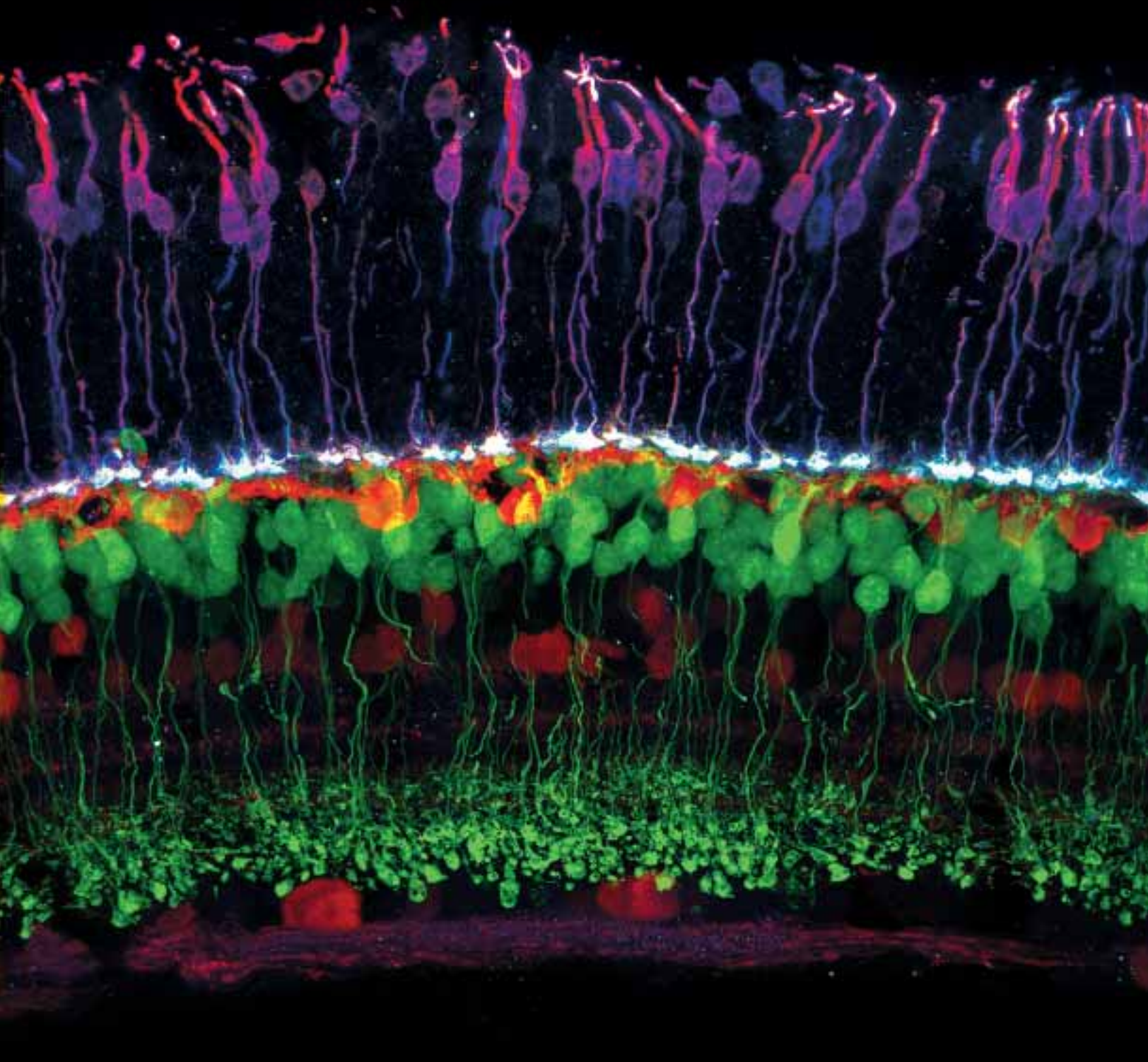
CONFOCAL LASER SCANNING
BIOLOGICAL MICROSCOPE

FV1000

FLUOVIEW

UIS2
World-leading optics

Next Generation
Live Cell Imaging System

THE EVOLVED FV1000

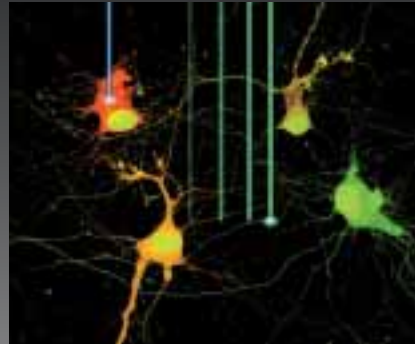## EVOLVED STABILITY

Diode Laser

Stability

- Greater stability, longer service life and lower operating cost are achieved using diode lasers.
- Reduced heat generation and noise alleviate operator discomfort.
- With increased power, both imaging and laser light stimulation can be done with one laser.

EVOLVED LASER LIGHT STIMULATION

SIM Scanner

Laser Light Stimulation

- Synchronized laser stimulation and imaging with the SIM (SIMultaneous) scanner ensures cellular reactions
  during or immediately following stimulation are not overlooked.
- The stimulation/imaging positions and laser wavelengths can be set separately with two independent beams.
- Laser light stimulation position can be changed during imaging.
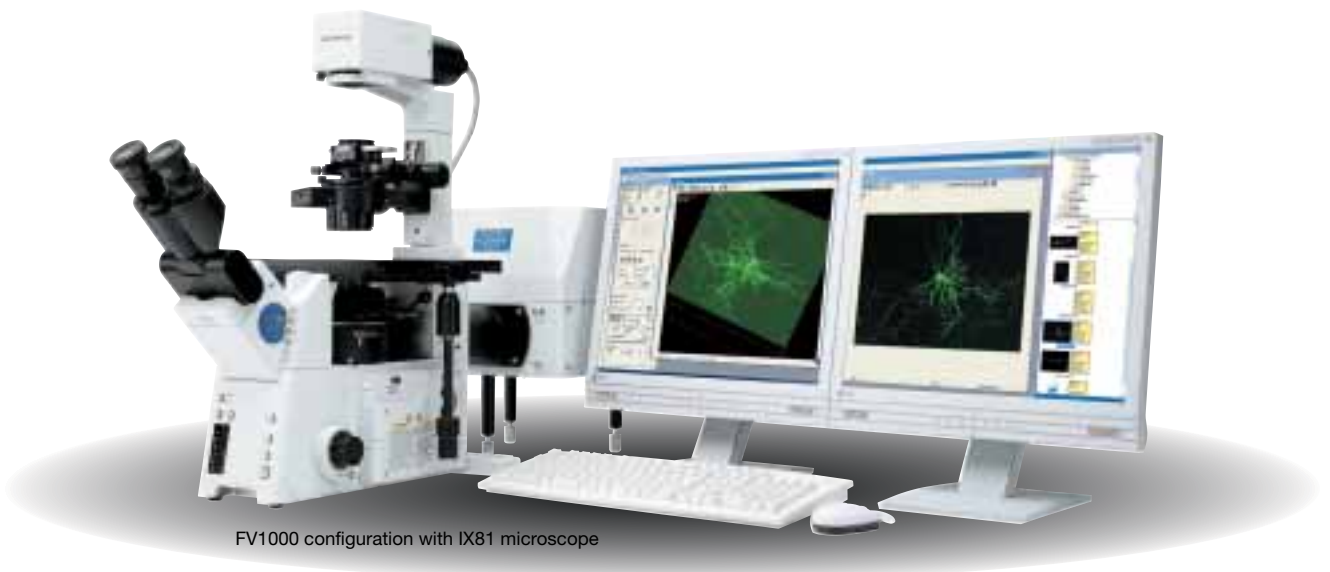
FV1000 configuration with IX81 microscope

The FV1000 delivers all of the key performance functions required from a confocal laser scanning microscope, minimizing specimen damage during high-speed imaging of living organisms, and accurately captures a full range of related information.

EVOLVED BASIC PERFORMANCE

Dependable Performance

With laser power monitor   Without laser power monitor
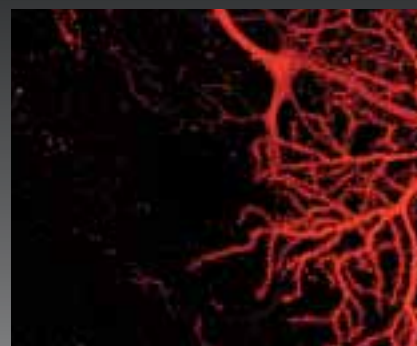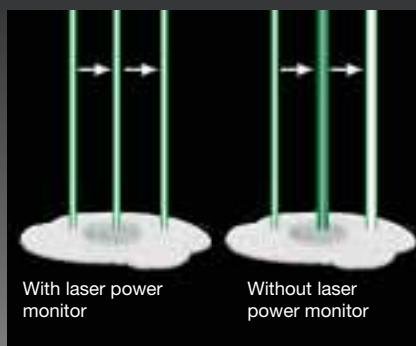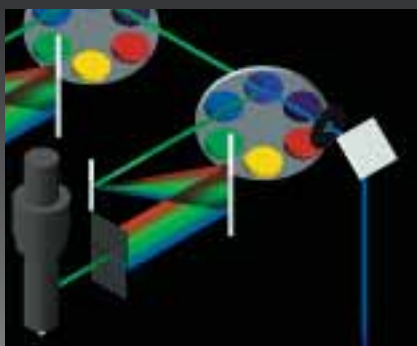
High-precision VBF

- Variable Barrier Filter (VBF) for maximally utilizing the grating function.
- Thanks to the variable VBF with 2 nm resolution, acquisition wavelength can be set freely to suit various fluorochromes.
- Simple separation of fluorescent cross-talk using the lambda scanning and unmixing functions.

Quantification

- Laser power monitoring for stable stimulation.
- Stable excitation light via advanced laser intensity feedback system for greater stability.
- Measurement of fluorescent intensity during observation using live-plot.

High Sensitivity

- Newly designed Analog Accumulation Circuit (AAC).
- Uniquely coated filters and dichromatic mirrors enhance sensitivity.
- Highly sensitive photomultipliers selected specifically for the FV1000.
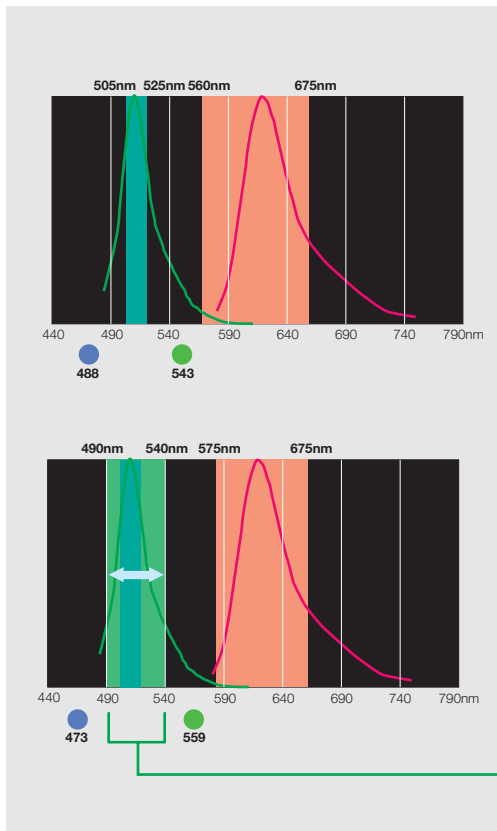- Superlative optics with minimum optical return loss.

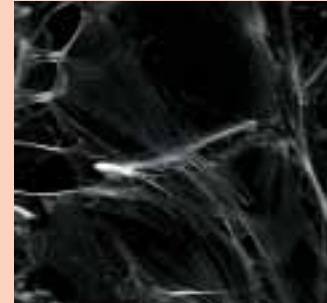FV1000 configuration with BX61 microscope

MULTI-COMBINER

Multi-combiner: the cutting-edge light source.

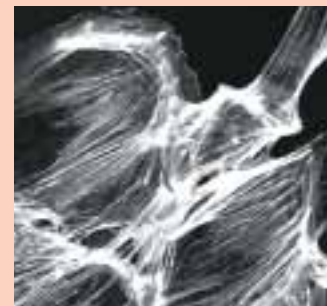### Incorporation of a wide wavelength range of diode lasers.

The Olympus multi-combiner system can be equipped with diode lasers corresponding to a wavelength range from 405nm to 635nm, and enables efficient observation of fluorescent proteins which change into multiple colors. The combination of a 473nm and 559nm laser is powerful for combinations of EGFP and pigments having an excitation wavelength peak in the range from about 550nm to 600nm. Diode lasers have numerous advantages, including improved stability, reduced heat and noise, and reduced operating cost due to conservation of electric power.

505nm 525nm 560nm 675nm

440 490 540 590 640 690 740 790nm

● **488** ● **543**

490nm 540nm 575nm 675nm

440 490 540 590 640 690 740 790nm

● **473** ● **559**

488nm/543nm/633nm laser combination

473nm/559nm/635nm laser combination

***C.elegance* expressing EGFP**
Due to excitation at 473nm, the width of acquired fluorescent light is broader than before, and a brighter EGFP image can be acquired.
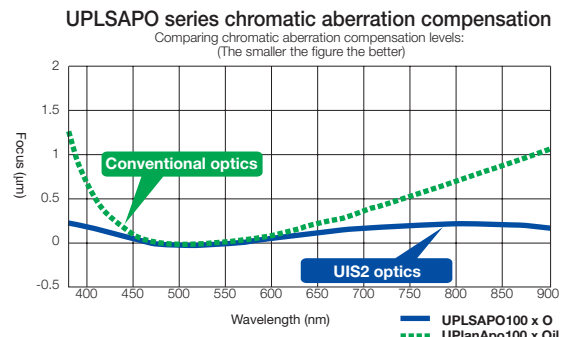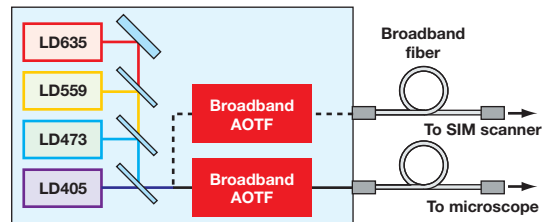
HeLa cell with actin dyed by AlexaFluor594
559nm has better excitation efficiency than 543nm for AlexaFluor594, and this enables acquisition of brighter images.

Double fiber system allows an imaging laser to be used for laser light stimulation.
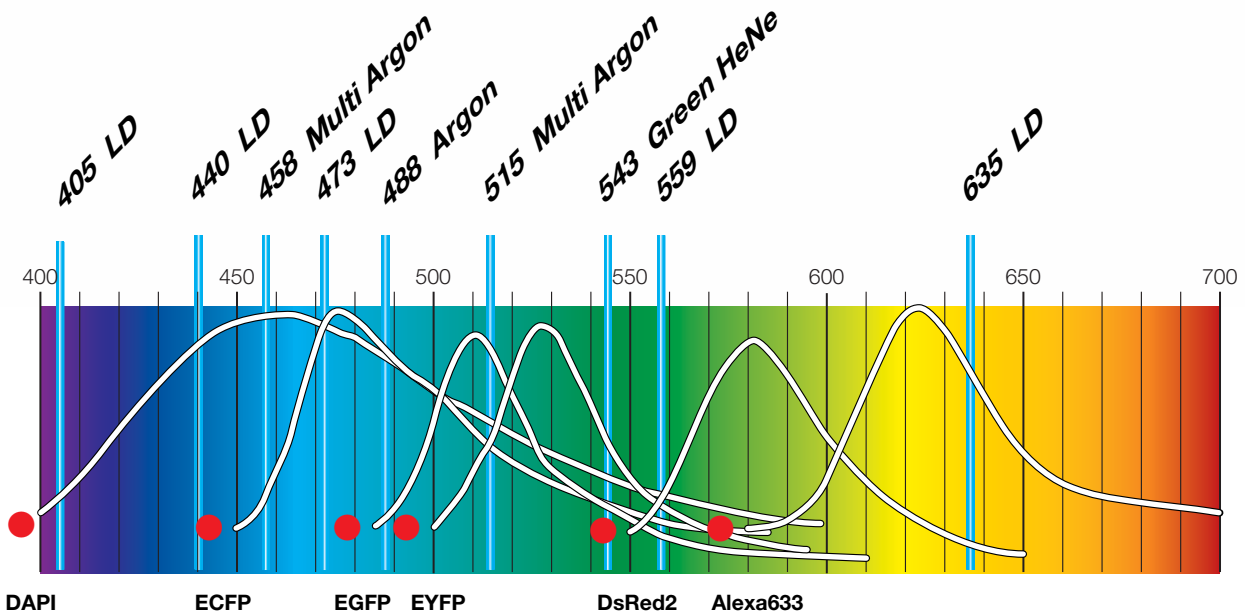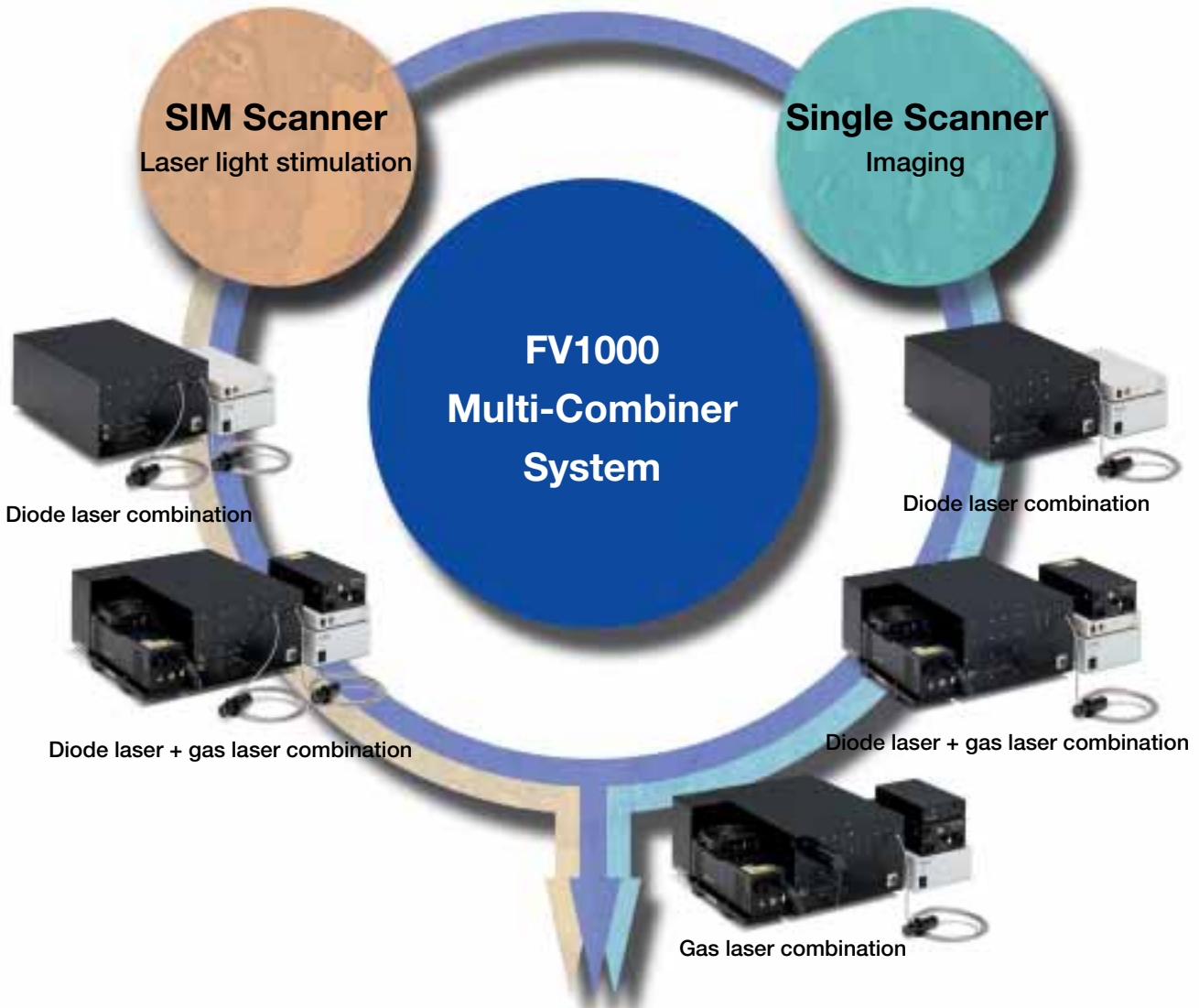
The laser used for observation can also be used for laser light stimulation. There is potential for new possibilities in combination with the SIM scanner system whose breadth of compatible applications has widened.

Broadband fiber and UIS2 optics integration for reduced axial chromatic aberration.

Converting the laser-based optic system to broadband makes it possible to introduce a visible laser and 405nm laser into the microscope with a single fiber. Furthermore, the system employs UIS2 objectives, so shift in the axial direction is minimized and the system can exhibit outstanding performance. Color shift and position shift between images is small, and high-reliability can be obtained in colocalization.

| LD635 |
| LD559 |
| LD473 |
| LD405 |

Broadband AOTF

Broadband fiber

To SIM scanner

To microscope

UPLSAPO series chromatic aberration compensation

Comparing chromatic aberration compensation levels:
(The smaller the figure the better)

Focus (μm)

2
1.5
1
0.5
0
-0.5

400 450 500 550 600 650 700 750 800 850 900

Wavelength (nm)

Conventional optics

UIS2 optics

―――― UPLSAPO100 x O
‒ ‒ ‒ ‒ UPlanApo100 x Oil

SIM Scanner
Laser light stimulation

Single Scanner
Imaging

FV1000 Multi-Combiner System

Diode laser combination

Diode laser combination

Diode laser + gas laser combination

Diode laser + gas laser combination

Gas laser combination

405 LD
440 LD
458 Multi Argon
473 LD
488 Argon
515 Multi Argon
543 Green HeNe
559 LD
635 LD

400   450   500   550   600   650   700

DAPI   ECFP   EGFP   EYFP   DsRed2   Alexa633

SIM SCANNER

SIM (SIMultaneous) scanner system: simultaneous laser light stimulation and imaging.

Lasers used for imaging can be used for laser light stimulation

Branching of laser in laser combiner
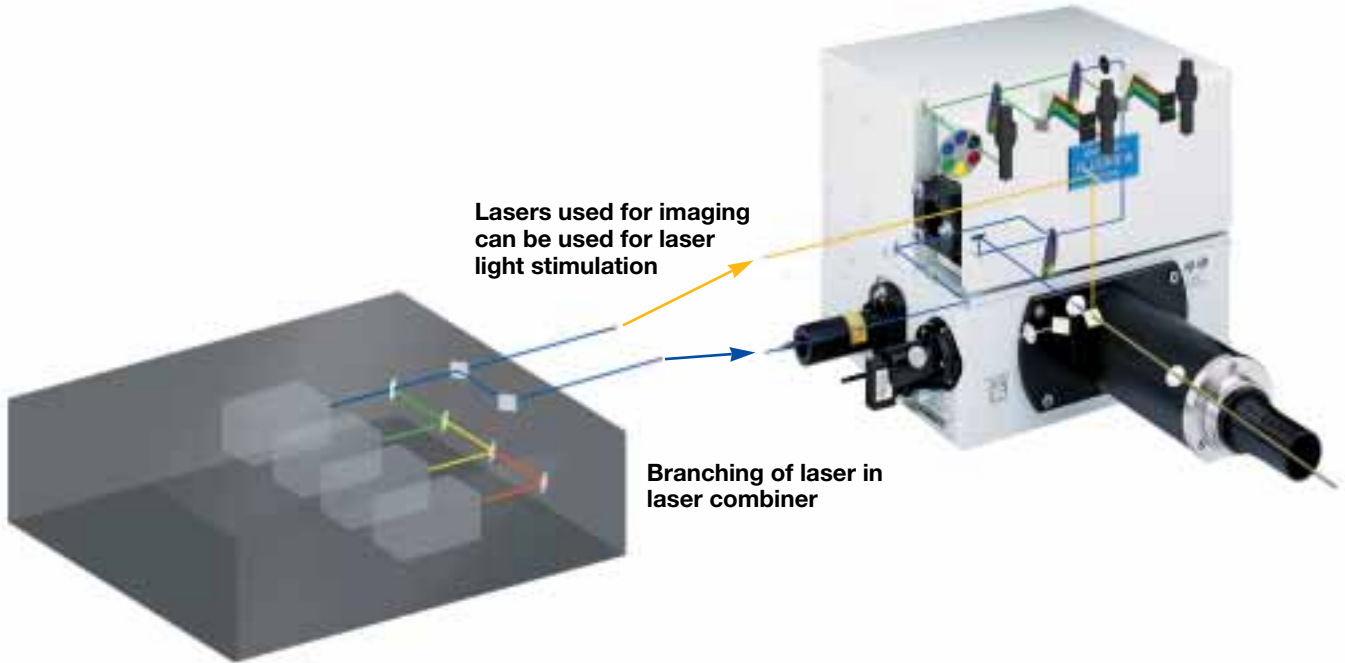
Assured capture of reactions immediately following laser light stimulation.

The compact design incorporates two laser scanners, one for confocal imaging and the other for simultaneous stimulation. They can be illuminated separately and independently, making it possible to stimulate the specimen during observation. As a result, the rapid cell reactions that occur right after laser stimulation can be accurately and reliably captured, making the FV1000 ideal for such applications as FRAP, FLIP, photoactivation and photoconversion.

Stimulation area can be changed during imaging.

Two laser beams, one for imaging and one for laser light stimulation, can be controlled independently and separately. The stimulation area can be moved to a different position on the cell during imaging, so the system is powerful for a variety of experiments.

Wide choice of different bleaching modes.

Various scan modes can be used for both the observation area and stimulation area. This enables free-form bleaching of designated points, lines, free-lines, rectangles and circles.
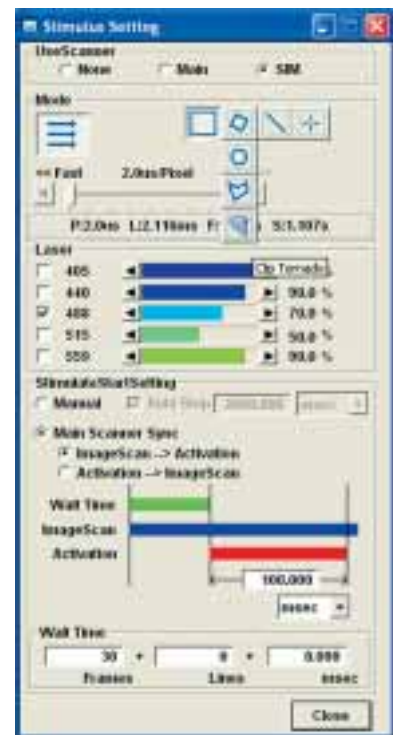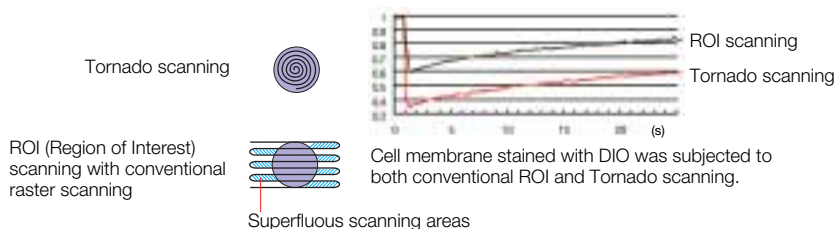
Multi-laser combiner enables 2-channel laser output.

Laser light is branched in the laser combiner, and each laser wavelength provided in the system can be selected and used for imaging and stimulation.

Tornado scanning for highly efficient bleaching.

Conventional raster scanning cannot always complete photobleaching quickly. Tornado scanning makes the procedure much more efficient by significantly reducing unnecessary scanning.

*Tornado scanning is available only with the SIM scanner.

Tornado scanning

ROI (Region of Interest) scanning with conventional raster scanning

Superfluous scanning areas

ROI scanning

Tornado scanning

Cell membrane stained with DIO was subjected to both conventional ROI and Tornado scanning.

Stimulus setting
This enables selection of the laser wavelength, intensity, scanning mode and other parameters for the stimulation laser.
When using the SIM scanner, control can be linked with imaging in µs units.

Positive fluorescence imaging of live cells by laser light stimulation while imaging is in progress.

In contrast to bleaching, light stimulation enables many changes inside and outside cells, such as intensifying fluorescence or changing colors, and can be performed with minimal light. Live cell imaging is dramatically enhanced by capturing a diffusion or transfer of fluorescence-labeled molecules, or by marking a specific live cell with an appropriate color stain. The FV1000's SIM system can provide excitation scanning separately from image scanning; this allows the user to illuminate the specimen as required, without the restrictions of image settings.

The illumination area is not limited to the imaging field of view, but covers areas outside as well. The imaging area and laser light stimulation areas can be freely set up on the reference image.

Kaede

This fluorescent protein is made through cloning from Trachyphyllia geoffroyi. It emits a strong green light after synthesis, but when stimulated by ultraviolet laser illumination, changes its color from green to red, like a maple tree in autumn. Its name is derived from this characteristic ("kaede" means "maple tree" in Japanese).

When violet (or ultraviolet) laser illumination is directed onto a Kaede-expressing cell, diffusion of the reddish Kaede can be monitored throughout the whole area of the cell, providing an easy and accurate method for capturing the whole cell labeling. The FV1000 allows this to be done while observation is in progress.

When Kaede is expressed in the cytoplasm of a live cell, it shows a high-speed diffusion coefficient value (about $30\mu m^2/s$) in spite of its tetramic structure. Taking advantage of this property, the movements of Kaede molecules in the cell can be observed more easily than molecules in general GFP/FRAP experiments. Since Kaede photoconversion requires only slight laser light, less intense than that used in ordinary photobleaching, labeling can be completed in a very short time.

| Before stimulation | After stimulation |
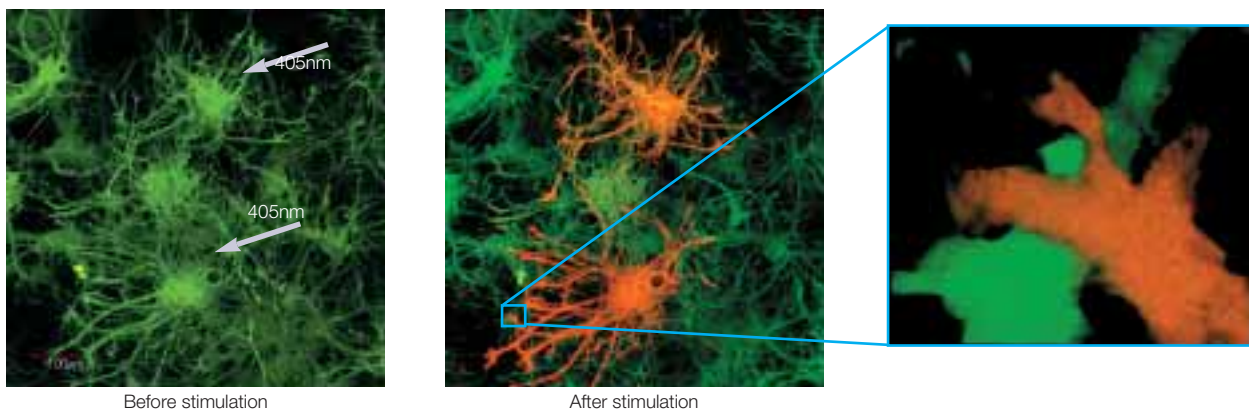
Kaede-expressing astroglia cells are stacked on the Kaede-expressing neurons. By illuminating two colonies with a 405nm laser, the Kaede color can be photoconverted from green to red. The glial cells in contact with the neurons are observed while they are forming colonies and extending their processes, and the nuclei of these colonies can also be observed. The FV1000 SIM scanner makes it easy to change cell colors from green to red while conducting an observation, and to control neutral colors between red and green.

Data courtesy of:
Dr. Hiroshi Hama, Ms. Ryoko Ando and Dr. Atsushi Miyawaki
RIKEN Brain Science Institute Laboratory for Cell Function Dynamics
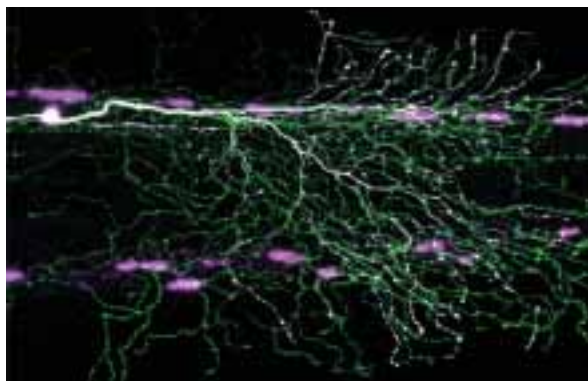
Zebra fish
Embryo where Huc:Kaede has been injected into a fertilized egg (5th day after insemination)
Image observed after 3 days via 405nm stimulation of sensory spinal nerve (Rohon-Beard) cells.
Objective: LUMPlan FL20 x W (N.A.0.8)

Dr. Tomomi Sato, Dr. Hitoshi Okamoto
RIKEN Brain Science Institute Laboratory for Developmental Gene Regulation
Reference:
Sato T, Takahoko M, Okamoto H. (2006) HuC:Kaede, a useful tool to label neural morphologies in networks in vivo. Genesis. 44:136-42.

Zebra fish
Embryo where Huc:Kaede has been injected into a fertilized egg (5th day after insemination)
Image observed approximately 1 hour after 405nm stimulation of the trigeminal ganglion
Objective: LMPlanFL60 x W (N.A.0.9)

Dr. Tomomi Sato, Dr. Hitoshi Okamoto
RIKEN Brain Science Institute Laboratory for Developmental Gene Regulation
Reference:
Sato T, Takahoko M, Okamoto H. (2006) HuC:Kaede, a useful tool to label neural morphologies in networks in vivo. Genesis. 44:136-42.

SIM SCANNER

FRAP (Fluorescence Recovery After Photobleaching)

Conventional FRAP applications are performed using a laser controlled via the AOTF (Acousto-Optical Tuneable Filters). Introduction of the SIM scanner further improves FRAP performance. For instance, while conducting an observation with the main scanner laser, the user can use a second laser to carry out photobleaching on a particular targeted area. As a result, the rapid movements of fluorescence molecules that come from outside the targeted area immediately after photobleaching are not overlooked.

FRAP: Fluorescence recovery after photobleaching is a method for analyzing molecular movements. It measures diffusion rates, tethering to other structural components and the separation speed of molecules.
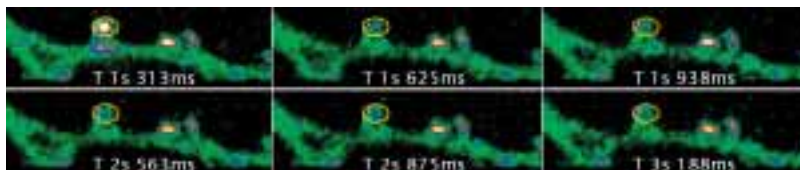
Fluorescence intensity is used to analyze molecule concentration and mobility in a postsynaptic density site in hippocampal neurons. FRAP analysis reveals differences in the molecules' speed of integration with the postsynaptic density. The intensity of fluorescence recovery, and the time it requires, allow the degree of molecular mobility to be measured.

Specimen: Hippocampal neurons, Shank-GFP stain, 488nm excitation (Multi-argon laser)
Image acquisition time: 100ms / bleach time: 80ms., 488nm excitation (Sapphire 488 laser)

Data courtesy of: Dr. Shigeru Okabe
Department of Anatomy and Cell Biology, Tokyo Medical and Dental University

## PA-GFP (Photo Activatable-Green Fluorescent Protein)

Fluorescent protein PA-GFP can be used to mark targeted cells, organelles and proteins. The SIM scanner allows illumination of any designated area at any time. In the following photos, 405nm laser excitation is conducted intermittently on part of a PA-GFP expressing cell, and time-lapse changes of fluorescent signals on different points of the cell are monitored. This provides information about protein diffusion within the cell.
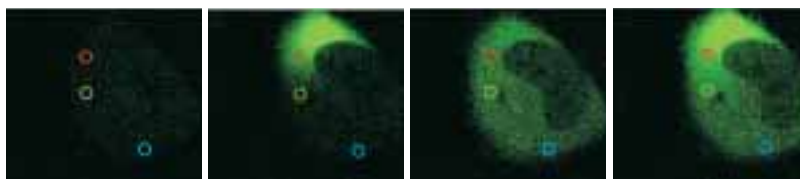
Specimen: PA-GFP labeled HeLa cell, 488nm excitation, image acquisition every 1 second.

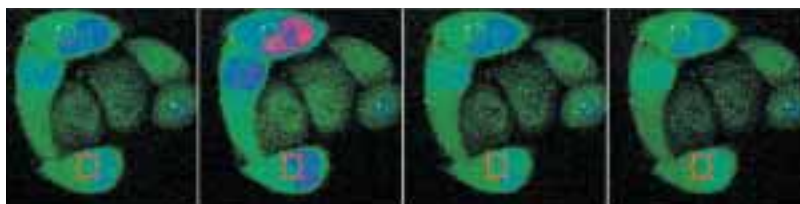Light stimulation: 405nm laser with intermittent stimulation

Data courtesy of: Dr. Takeharu Nagai, Dr.Takayuki Miyauchi, Dr. Atsushi Miyawaki
RIKEN Brain Science Institute Laboratory for Cell Function Dynamics
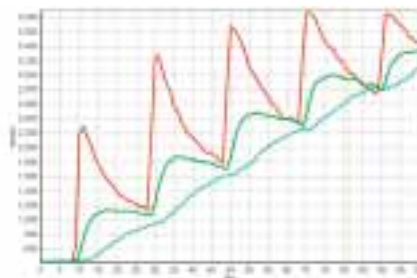
Uncaging

With a 405nm laser or ultraviolet laser attached, the SIM scanner system can be used for uncaging. Caged compounds can be uncaged point-by-point or as an ROI operation, while the FV1000's main scanner captures images of the uncaging without any time lag.

Caged-ATP

Introduction of fluorescent calcium fluorochrome Ca Green into the HeLa cell

Fluorescent calcium indicator Calcium Green in HeLa cells.
Using caged ATP, it is possible to observe an increase in calcium ion concentration inside the cell in response to the release of caged ATP via a pulse from a 405nm laser.

Data courtesy of: Dr. Takeharu Nagai, Dr.Takashi Fukano, Dr. Atsushi Miyawaki
RIKEN Brain Science Institute Laboratory for Cell Function Dynamics
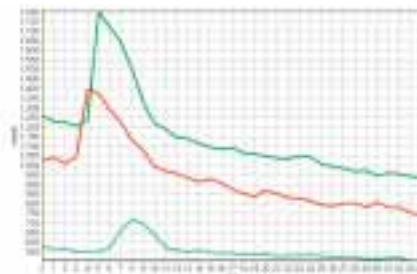
FLIP (Fluorescence Loss In Photobleaching)

The FV1000 SIM scanner system optimizes FLIP operation. Instead of the conventional method of alternating between photobleaching and imaging, this system conducts both procedures at the same time, enabling reliable capture of even rapid molecular movements.
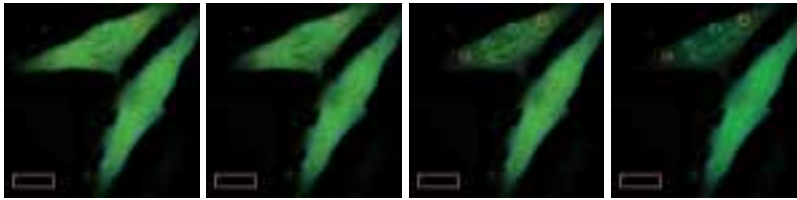
The diffusion mobility of fluorescence molecules in a cytoplasm is difficult to detect by FRAP, but can be detected by FLIP. By continuously bleaching a single point in the cytoplasm, different fluorescence fade speeds are captured according to the distances from the bleached point. These differences allow the ease of molecular transfer to be measured.

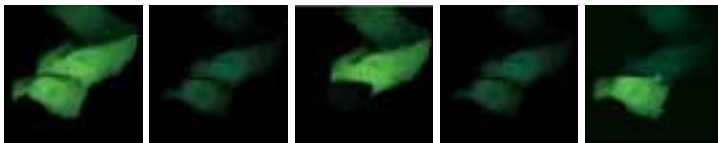Specimen: HeLa cell, GFP (Free), 488nm excitation (Multi-argon laser)

Image acquisition time: 100ms./ bleach time: 100 sec. continuously, 405nm bleaching
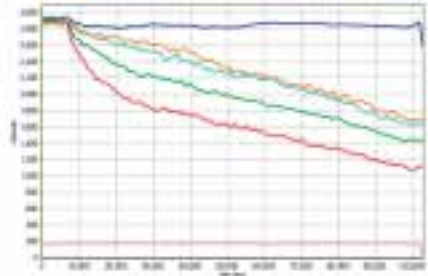
Dronpa

Dronpa is a new fluorescent protein with photochromic properties. Using photoactivation like PA-GFP, this method is suitable for observation of molecular movement, but has unique additional functions.
1. Since fluorescence emission is extremely faint before activation of PA-GFP, it is difficult to confirm expression and cell fixation before imaging. By contrast, Dronpa provides very bright fluorescence signals before activation, so that expression and cell fixation are easy to confirm.
2. Photochromism appears broadly similar to photobleaching, with fluorescence intensity reduced by irradiation with a strong 488nm laser. However, fluorescence intensity reduced by photochromism is readily restored with illumination from a faint 405nm laser. As a result, user-selected control of fluorescence intensity using 405nm (for excitation) and 488nm (for imaging) enables repeated photoactivation experiments on the same cell.

3. The fluorescence intensity before fading and previous photoactivation results are used as control values, which makes experiments much quicker to perform.

Laser wavelength: 488nm (excitation)/ 488nm (photobleaching) /405nm (stimulation)

Data courtesy of: Ms. Ryoko Ando. Mr. Hideaki Mizuno, Dr. Atsushi Miyawaki,
RIKEN Brain Science Institute Laboratory for Cell Function Dynamics
References: Ryoko Ando, Hideaki Mizuno, Atsushi Miyawaki, Science 19 Nov. 2004, Volume 306, pp. 1370-1373
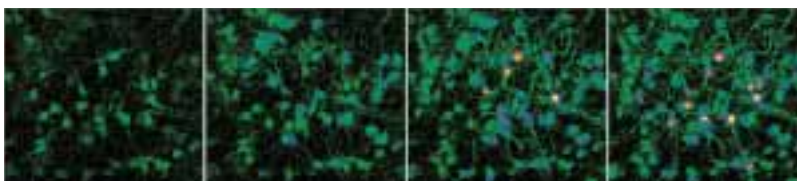
Caged-Glutamate

Fluorescent calcium indicator Fluo-3 in HeLa cells. Image acquisition at 1-second intervals

Using the caged compound Bhcmoc-Glutamate, an increase in calcium ion concentration inside the cell can be observed in response to glutamate stimulation, released via 405nm laser illumination.

Data courtesy of: Dr. Hiroshi Hama, Dr. Atsushi Miyawaki
RIKEN Brain Science Institute Laboratory for Cell Function Dynamics
Caged compound Bhcmoc-Glutamate presented by Dr. Toshiaki Furuta, Department of Science, Toho University

MAIN SCANNER
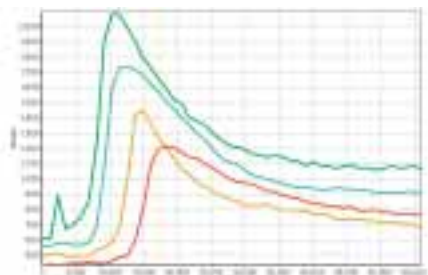
Spectral and filter scan units provide two types of high-precision scanning.

Variable Barrier Filter (VBF).

With the spectral detection system, once a fluorochrome combination is selected within the software, the ideal wavelength spectrum is automatically selected. Of course, manual adjustment of the wavelength ra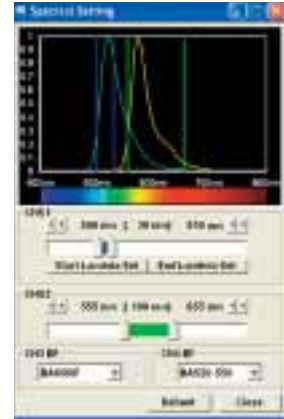nge is also possible according to the fluorescence emission wavelength peaks. The spectral scan unit can be used to optimize image acquisition since the sensitivity of each channel can be adjusted with the same responsiveness as the filter version.

### High performance mirror unit with special coating.

High-performance mirrors with a special new coating are introduced to all emission filters and dichromatic mirror units, achieving sharper transitions which were not possible with the conventional vacuum deposition method. In particular, these mirrors cover the full fluorescence emission spectrum for fluorescent proteins, enabling clear observation even with weak fluorescence emission and minimizing damage to live cells.

High sensitivity detection system.

High-sensitivity, high S/N ratio optical performance is achieved through integration of a pupil projection lens within the optics and employment of a high-sensitivity photomultiplier and analog processing circuit with minimal noise.

Photon counting detection mode.

The Olympus original Hybrid Photon Counting Mode (HPCM) can successfully capture images even when fluorescence emission is weak. This mode optimizes photomultiplier control conditions to acquire images with minimum noise and high quantitative performance.

Excitation DM488/543/633 comparison

- - - - - Conventional   ——— High performance mirror
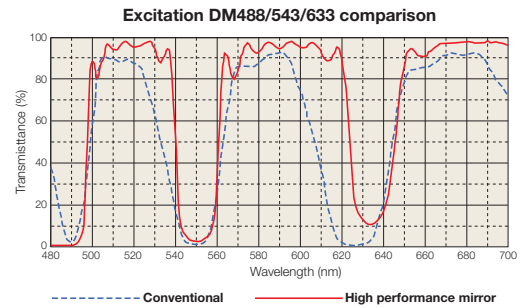
Original spectral system.

An independent photomultiplier is incorporated into the original optics for each channel. The special spectral scanning system uses a diffraction grating, which has excellent linear performance with no wavelength deviation and ensures high-precision, high-resolution, high-speed spectroscopy.

High-speed spectroscopy.

High-speed galvanometer diffraction grating delivers high wavelength change speed (100nm/ms), enabling very fast acquisition of XYλT images.

High-precision spectroscopy.

Cross talk derived from two fluorochromes with similar emission wavelength peaks can be clearly separated by the system's highly accurate 2nm wavelength resolution. Even when working with specimens whose fluorescence emission wavelength is similar to the excitation wavelength, it is possible to obtain images unaffected by the excitation light.

Grating
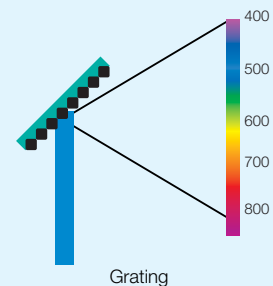
XYλ series, wavelength range 495-695nm

| 495nm — 515nm | 565nm — 585nm | 575nm — 595nm | 595nm — 615nm | 675nm—695nm |
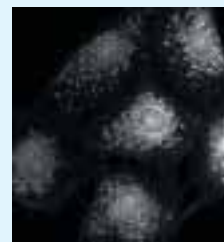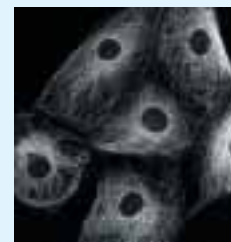
Spectral Version Scan Unit

CH3

IR laser port

UV laser port

VIS laser port

Fluorescence illumination

Barrier filter

DM

Grating

Slit

CH2

CH1

Slit

DM

Grating

Confocal pinhole

Pupil projection lens

Galvanometer scanning mirrors

Excitation DM

to Microscope

Filter Version Scan Unit

IR laser port

UV laser port

VIS laser port

Fluorescence illumination

Barrier filter

Barrier filter

DM

DM

Confocal pinhole

Barrier filter

Pupil projection lens

Galvanometer scanning mirrors

Excitation DM

to Microscope

SOFTWARE

Compatible with laser light stimulation and spectral unmixing.

Suitable for multiple users

For the convenience of multiple researchers, each user may create an independent log which contains individual settings such as the display window and toolbar. To change researchers, the new user simply logs in at software start-up.

On-line help

The comprehensive On-line help explains each command's function and usage, as well as the overall flow of operations.

One-touch exchange between confocal and fluorescence observation

Since the FV1000 is fully automated, one-touch exchange between confocal and fluorescence observation is possible. In addition, microscope settings can be easily changed through the Microscope Controller.

Microscope Controller

Wide choice of scanning modes

Several scanning modes are available, including ROI, point and high-speed bi-directional scanning. These can be used together freely, in such combinations as XYZ, XYT, XYZT, XYλ, XYλT, etc.

### Easy designation of scan area

The observation field of view and scanning area are displayed graphically, and settings can be altered while confirming the magnifications with the scroller. The operator can move the image scan area at will using the Pan button. Rotation scanning of the image field is also possible.

Eliminate cross talk by using sequential mode

Both frame and line sequential modes can be selected. The order of image acquisition, or image combinations, can be freely changed.

Free emission wavelength selection

When selecting fluorochromes from within the software, the fluorescence spectra are displayed and the ideal detection wavelengths are automatically selected. Naturally, manual adjustment of the wavelength emission range, in as little as 1nm step increments, is also possible in order to optimize acquisition of specific fluorescence emission peaks.
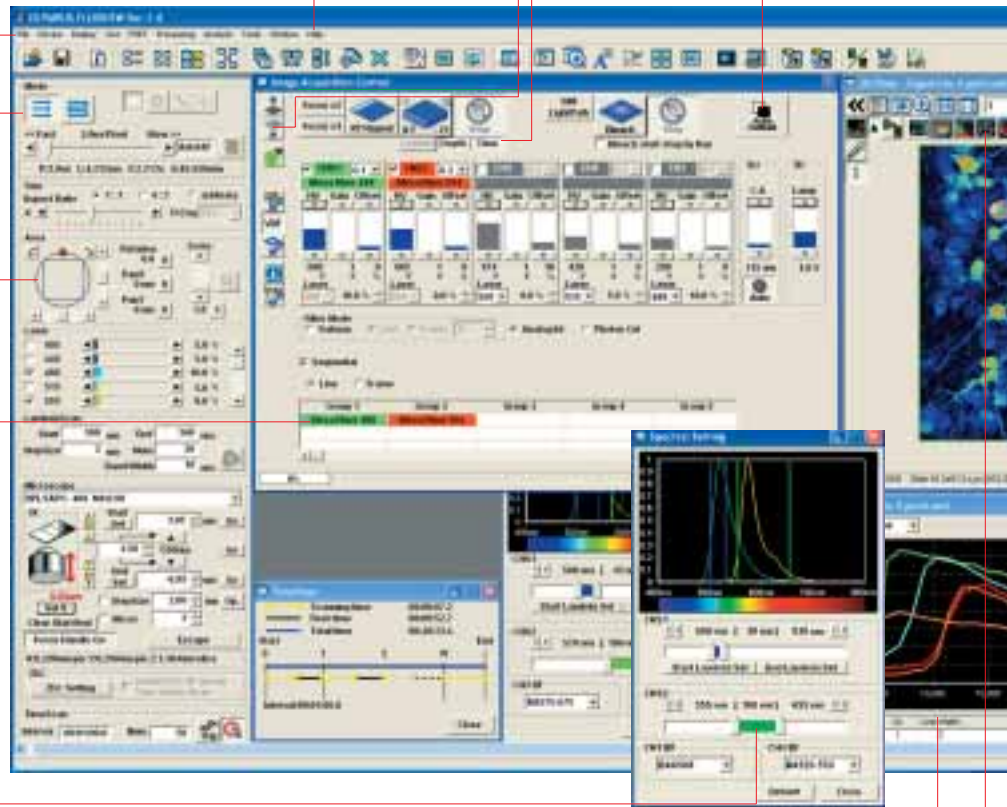
Real-time emission intensity graph display

For image acquisition in real time, live emission intensity is displayed in a graph. Since the images are captured using the full 12-bit capacity, this is also convenient for setting sensitivity levels.
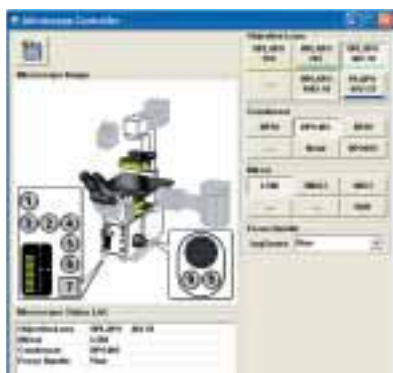
Easy image acquisition for 3D, 4D and 5D series

Multi-dimensional image acquisition, such as λ series, Z series and time-lapse, is easily performed.

Automatic contrast

Automatic contrast selects optimal photomultiplier voltage settings for each detection channel, simplifying image acquisition.

2D image display

The 2D control panel enables free manipulation of the image display. Tiling display or multi-dimensional image display can also be freely selected.
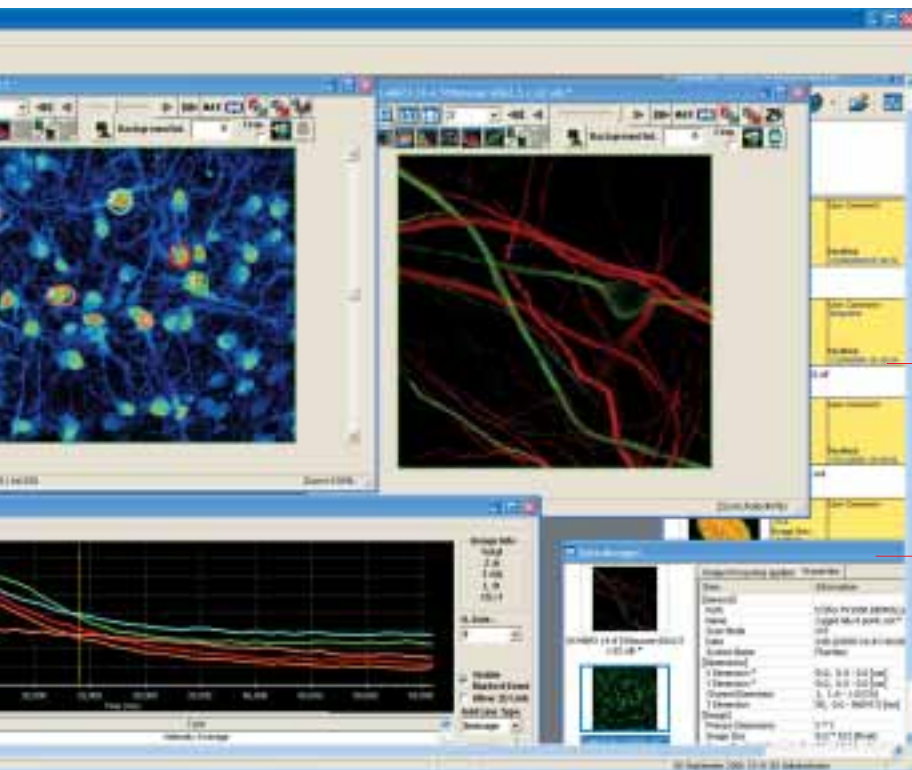
Easy image search

Explorer software provides simultaneous thumbnail image display, making it easy to search for previously stored data.

Data manager

The data manager provides thumbnail displays and different types of file information.

Re-use function

Previously-set scanning conditions can be recalled and applied to new or subsequent experiments.

File Input/Output

OIB (Olympus Image Binary format) is employed to acquire both scanning and microscope setting conditions together with image acquisition data. This software also handles all widely used image formats (TIF, BMP, JPEG, AVI, MOV etc) with high interchangeability.

2D View Analysis Tool

Background Correction:
Subtracts background.

Region Measurement:
Measures size or intensity of region designated as ROI.

Intensity Profile:
Displays intensity profile of region designated with ROI or Line.

Histogram:
Displays histogram of intensity values of region designated as ROI.

Series Analysis:
Analysis of variation in intensity along Z-axis/time-axis in region designated as ROI.

Line Series Analysis:
Analysis of variation in intensity along Z-axis/time-axis on a designated line.

Colocalization:
Analysis of degree of overlap, between two channels, of pixels at or higher than a certain intensity.

Ratio:
Create an image using the intensity ratio between two channels.

SCANNING MODES

Live tile mode, Look back function.

This allows the user to check the acquired images during time lapse experiment. Adjustment of focus and/or image brightness can be done during rest time.

High-speed image acquisition (4KHz/line).

High-speed scanning mode can capture confocal images at 16 frames per second with 256 x 256 pixel resolution. In combination with clip scanning, images can be acquired even faster than video rate.

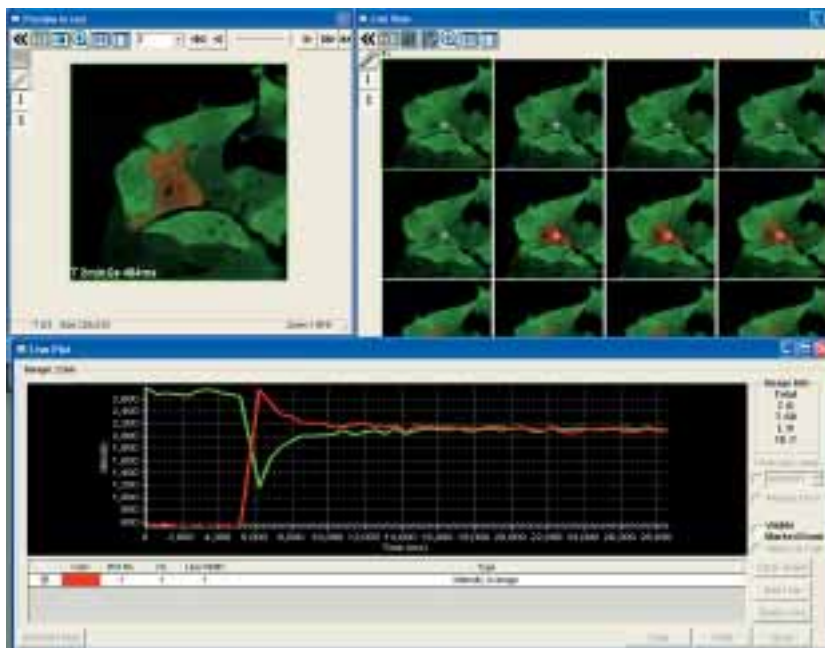**Wide variety of line scanning modes.**

Line scanning for a straight line, slanted line or free curve enables easy analysis of changes over time at the msec order. Observation of complex time lapse combined with Z or λ element is also possible.

Trigger function.

The system also has a trigger function for synchronizing scanning with external devices. Scanning can be started/stopped with a trigger signal, and it is also possible to gather frames for each external trigger.

Live plot function

Changes in fluorescent intensity in a region designated as ROI are plotted in real-time during image acquisition.

Laser monitoring function.

Feedback is applied to the laser output so that the sample is always exposed to a fixed level of excitation light. There is no need to pay attention to variation in laser output, and the amount of fluorescence can be measured accurately.

TIME COURSE

Using the Time Controller to schedule experiment flows and protocols.

• The Time Controller allows image acquisition conditions to be changed easily while observation is in progress. In addition to experiments such as FRAP and FLIP, the following protocols are supported:

1. Image acquisition while storing data on the hard disk.
2. Changing time-lapse intervals during the course of an experiment.
3. Image acquisition while changing the excitation laser in mid-procedure.
4. Data output from a specified point, using an external trigger.
5. After acquisition of reference images, laser intensity and excitation area can be changed.

• The experiment protocol can be entered from the taskbar in the protocol schedule area. Settings can be entered freely by clicking and dragging the mouse on the column of items to be scheduled.

Imaging execution time

Protocol schedule area
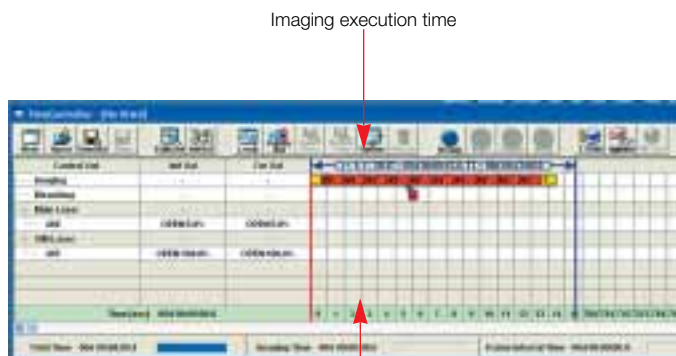
FRET

FRET (Fluorescence Resonance Energy Transfer).

FRET is the phenomenon by which the excitation energy of fluorescent molecules transfers to other fluorescent molecules, with the degree of transfer efficiency depending on the relative positions of two molecules. FRET allows observation of the interaction between two protein molecules, analysis of structural changes, and imaging of the calcium concentration in cells.

- Equipped with FRET analysis functions using the Ratio Imaging method, Acceptor Photobleach method and Sensitized Emission method.
- For the Acceptor Photobleach method, the system is also equipped with an experiment protocol setting function using a Wizard format, and this facilitates setting of experimental procedures.

Acceptor bleach can be set easily using wizard setting.
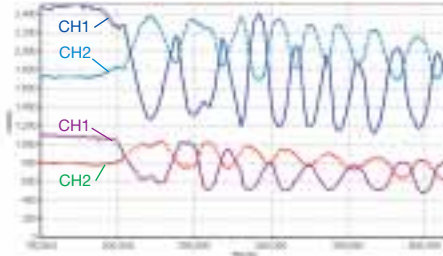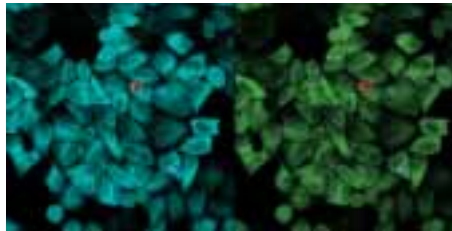
YC3.60 (Yellow Cameleon 3.60)

Due to structural improvement using circular permutation, YC3.60 offers superior performance and achieves a relatively high rate of change. Among other advantages, it allows high-precision, high-speed calcium concentration imaging, which is difficult by conventional methods. In LSM observation, calcium dynamic movements are observed with simple ratio imaging: this has contributed to making FRET imaging more popular. In the above ratio image photos, YC3.60 is expressed on HeLa cells, with calcium concentration changes captured when they are stimulated by histamine.
Laser for observation: LD440, 0.3% output
Objectives: 40x, zoom 1.3x

References: Takeharu Nagai, Shuichi Yamada, Takashi Tominaga, Michinori Ichikawa, and Atsushi Miyawaki 10554-10559, PNAS, July 20, 2004, vol. 101, no.29
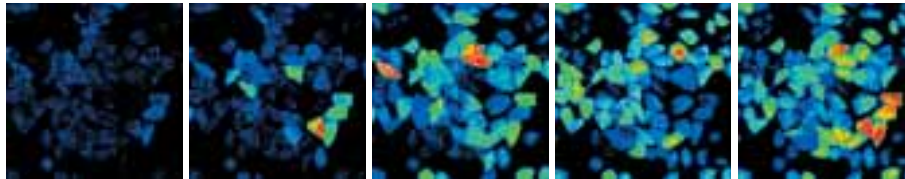
PHOTO STIMULATION

- Using the laser light stimulation setting function (Stimulus Setting), laser light stimulation experiments can be done with either the main scanner (image scanner) or SIM scanner. Even with the main scanner only, it is possible to perform laser light stimulation during a time course by instantaneously switching the laser scanning mode (laser wavelength, laser irradiation range). However, images cannot be acquired during laser light stimulation.
- Tornado scanning can also be used for laser light stimulation (with SIM scanner only). This is suitable for FRAP, Kaede protein and other photoconversion experiments.
- In intensity analysis, the timing of laser light stimulation is displayed simultaneously with the changes in fluorescent light intensity.
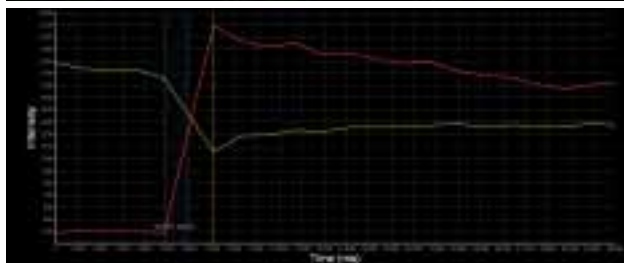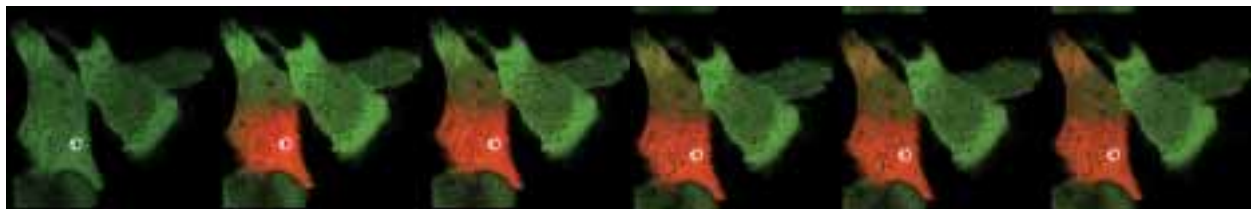
Photoconversion experiment to the Kaede protein expressed in the HeLa cells is done using tornade scanning with imaging laser.

Selectable rendering modes.

The Alpha Blend method and Maximum Intensity Projection method can be selected as the rendering method for the 3D display function. Select the optimal rendering method to suit the specimen.
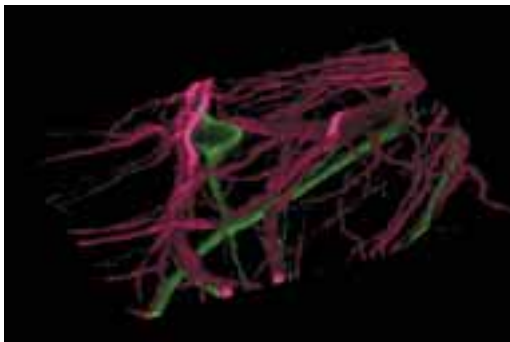
Alpha Blend method
(Cultured nerve cells derived from mouse hippocampus)
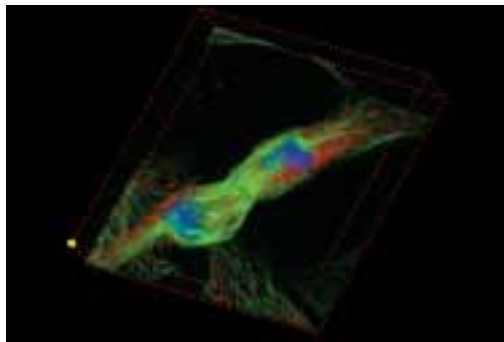*Dr. Koji Ikegami, Dr. Mitsutoshi Setou*
Molecular Geriatric Medicine, Mitsubishi Kagaku Institute of Life Sciences

Maximum Intensity Projection method (Ptk2 cells)

Interactive volume rendering.

Using the interactive volume rendering method with the 3D display function, the angle of a 3D rendered image can be freely changed to the direction you wish to see by operating the mouse. A variety of display functions are available, including the ability to display a cross section at an arbitrary location, and extended focus images.
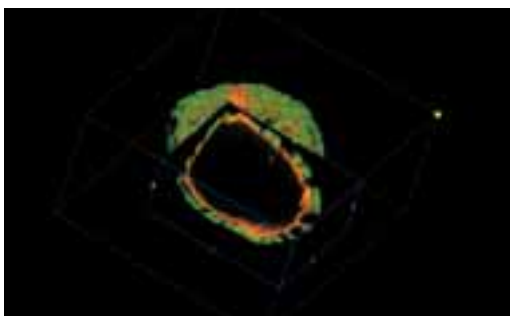
Lily pollen
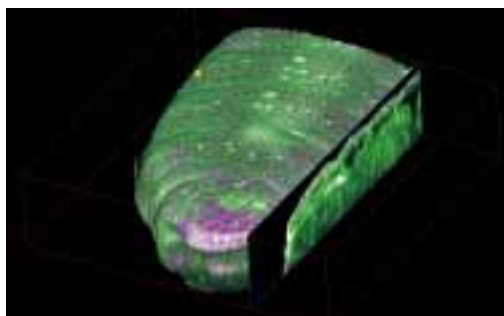
Wild-type embryo in stage 17 of drosophila
Dr. Tetsuya Kojima
Laboratory of Innovational Biology, Department of Integrated Biosciences
Graduate School of Frontier Sciences, University of Tokyo

4D animation creation function.

3D structures which change with the passage of time can be animated for images acquired with XYZT.

View of PKC moving to membrane due to PMA stimulation in HeLa cell (PKC-GFP)

Brightness compensation function for the depth direction.

When acquiring 3D images, the images darken in the depth direction. The system is equipped with a function for increasing laser intensity or PMT sensitivity in the depth direction, so that images of more distant parts can be acquired while retaining a fixed brightness.

Image without compensation

Image with compensation

UNMIXING

Easy fluorescence separation.

Fluorescence can easily be separated through two modes (Normal and Blind). In Normal mode, separation is performed based on a designated ROI fluorescence spectrum using already-known fluorochr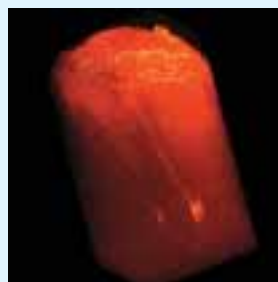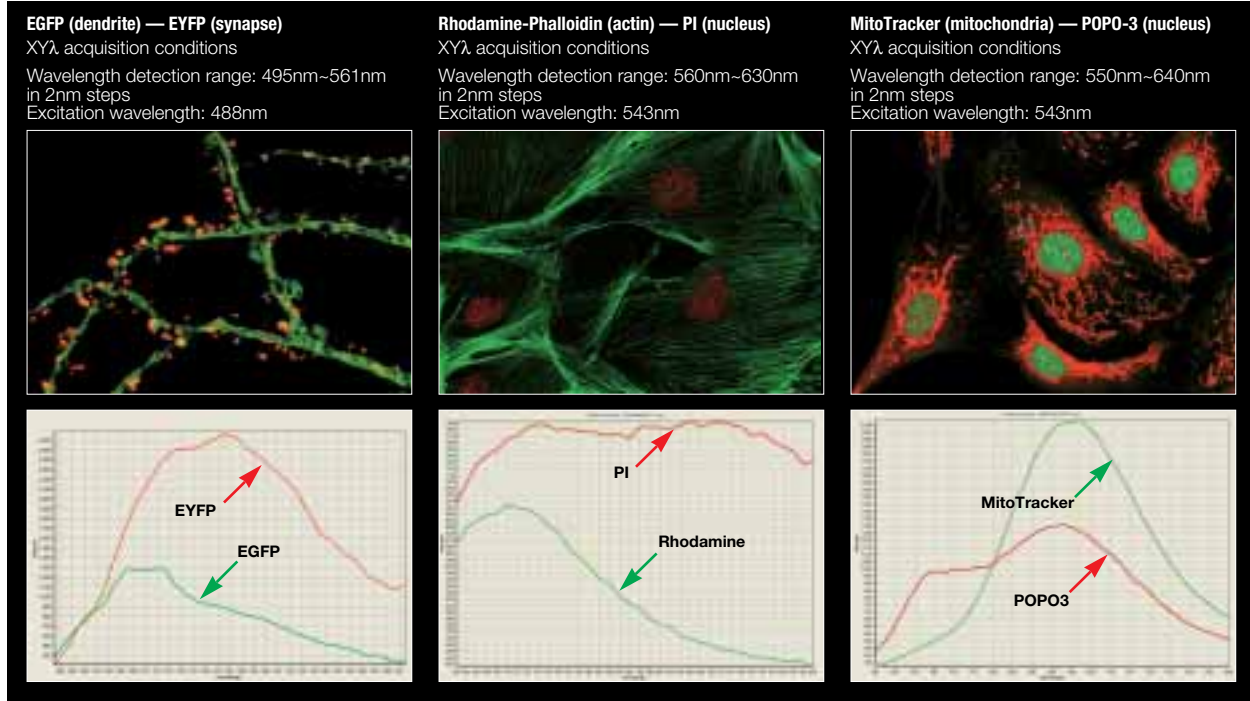ome wavelength data, or data derived from acquired images. In Blind mode, the separation uses an iterative process to derive the best fit of a given number of fluorescence spectra.

• 2nm spectral resolution allows two fluorochromes with similar emission peaks to be clearly separated

• Spectral unmixing is successful even when there are emission intensity differences in each fluorochrome

• A fine diffraction grating is used to gain precise separation for unmixing.

EGFP (dendrite) — EYFP (synapse)
XYλ acquisition conditions
Wavelength detection range: 495nm~561nm in 2nm steps
Excitation wavelength: 488nm

Rhodamine-Phalloidin (actin) — PI (nucleus)
XYλ acquisition conditions
Wavelength detection range: 560nm~630nm in 2nm steps
Excitation wavelength: 543nm

MitoTracker (mitochondria) — POPO-3 (nucleus)
XYλ acquisition conditions
Wavelength detection range: 550nm~640nm in 2nm steps
Excitation wavelength: 543nm

EYFP
EGFP

PI
Rhodamine

MitoTracker
POPO3

Data courtesy of: Dr. Shigeru Okabe
Department of Anatomy and Cell Biology, Tokyo Medical and Dental University

COLOCALIZATION

Analysis of emission intensity overlap between channels.

In the analysis of multi-stained specimens, it is easy to determine whether labeled molecules are localized in the same region. The degree of localization ("colocalization degree") may be quantified as Pearson's correlation coefficient, the overlap coefficient and the colocalization coefficient index, and enables comparison of the degree of colocalization between different specimens. This method is also applicable for analysis of an image series.

Colocalization image (white area)

Threshold Mode

Threshold lines can be interactively altered.

Regions/Min-Max Mode

Setting the ROI on the histogram makes it possible to create a colocalization image. Values can also be obtained for Pearson correlation, overlapping coefficient and colocalization index.
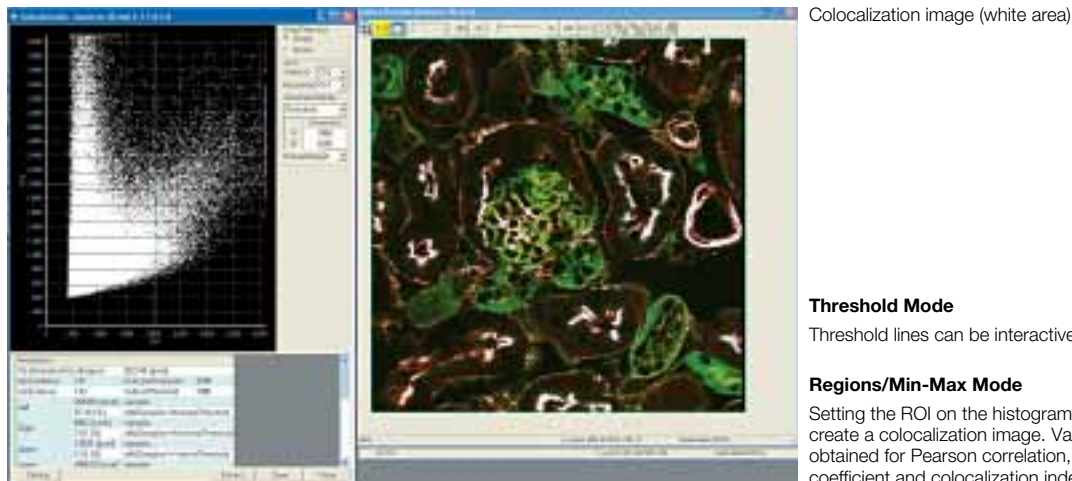
MULTI-POINT

Long-term, efficient time lapse of multiple live cells.

By equipping the system with a motorized XY stage, repeated image acquisition of multiple points located over a wide range is performed automatically. Furthermore, time lapse observation of the cells under different conditions is accomplished by using a well plate. These functions dramatically improve throughput of experiments requiring long-term observation.

- Repetitive operation can be done in sequence for multiple registered points, by simply setting the cells or specific points you wish to observe.
- Cell observation using well plates can be done more efficiently.
- Tiling image acquisition: After automatic registration of the neighboring visual field, a wide observation area can be automatically acquired while maintaining high resolution. (Separate software is needed to integrate the acquired images.)
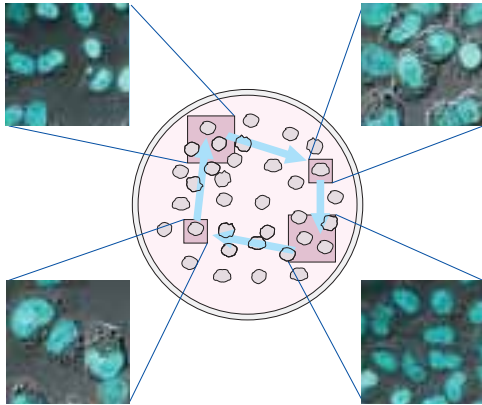
Simultaneous time lapse of cells in multiple areas
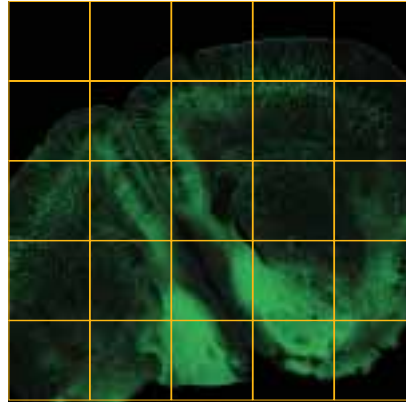
Tiling image acquisition

Image made by acquiring images at multiple points of a mouse brain section (YFP) and integrated.
Objective: UPlanSApo10x
Acquired visual fields: 25 fields
XYZ acquisition conditions for each visual field: 512 x 512, 15 slices

Sample provided by:
Ms. Mikako Sakurai,
Mr. Masayuki Sekiguchi (Section Chief)
Department of Degenerative
Neurological Diseases,
National Institute of Neuroscience,
*National Center of Neurology and*
Psychiatry

FV1000-ZDC

Corrects for thermal drift during confocal time-lapse imaging.

During long time-lapse observations, temperature changes around the microscope and drug administration during the observation cause focal drift, resulting in a loss of focus on the target. For confocal laser scanning biological microscopes with high resolution in the Z-direction, even slight focal drift can impair image acquisition to the point that images are no longer useful to researchers. Olympus is the world leader in equipping a confocal laser scanning biological microscope with zero thermal drift compensation.

Corrects automatically for thermal drift during confocal time-lapse imaging.

- In time-lapse imaging, focus is automatically corrected immediately prior to imaging.
- Compensation is performed in reference to the bottom surface of the dish, allowing target Z-slice images to be obtained regardless of sample conditions.
- Without thermal drift compensation, several Z-slice images must be taken to ensure acquisition of target image plane. Thermal drift compensation eliminates this need, minimizes sample exposure to irradiation.
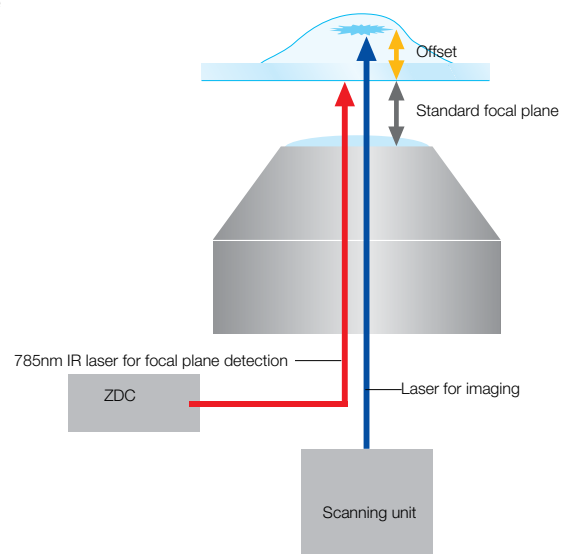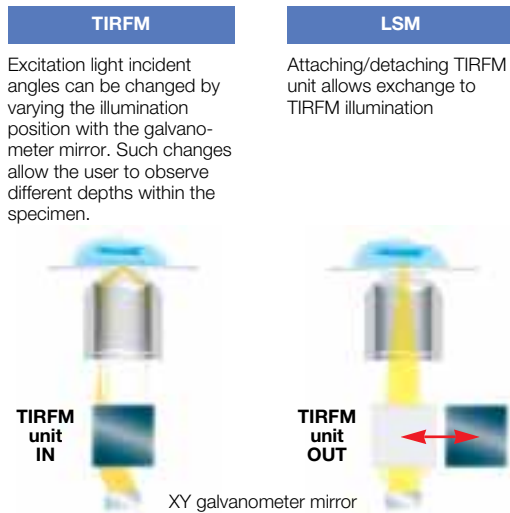
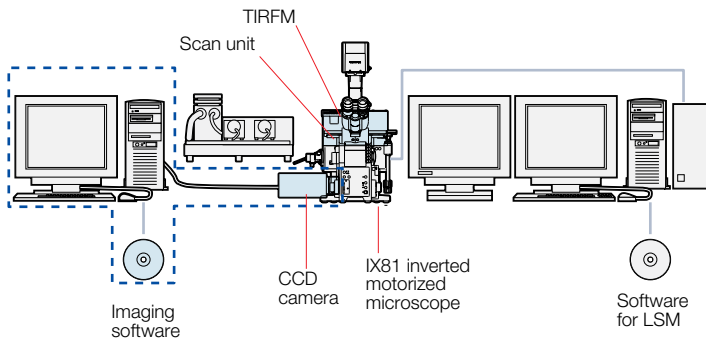# TIRFM *(Total Internal Reflection Fluorescence Microscopy)*

High S/N images near the cell surface: automated control of necessary volume of laser filtering light enables easy reproduction of TIRFM observation.

This special TIRFM unit employs the FV1000's laser for TIRFM illumination. The incident angle of the excitation laser toward the specimen is controlled through FV1000 software FV10-ASW, to set up the necessary laser filtering light volume. The optimum light path length is provided automatically through the selection of excitation wavelength and the objective. Since TIRFM observation can be done by exchanging confocal observation, protein localization on the cell surface and cross section images of the cell interior may be acquired simultaneously. A CCD is required for TIRFM image acquisition and image-capturing software is required. Note that time-lapse imaging by interchanging CCD and confocal images and then overlapping them is not possible.

* Cannot be incorporated with SIM scanner

TIRFM	**LSM**
Excitation light incident angles can be changed by varying the illumination position with the galvano-meter mirror. Such changes allow the user to observe different depths within the specimen.	Attaching/detaching TIRFM unit allows exchange to TIRFM illumination

		TIRFM	**LSM**
Features		Acquisition of high S/N images at 50-200nm depths from the specimen surface (surface observation only)	Allows observations from the surface to the intracellular structure Capable of 3D observation for each Z-axis position
		Acquisition of high S/N images with minimal influence from the background	Capable of composing 3D images from slice images
Method		Imaging through CCD camera Frame rate depends on camera performance	Point scanning Detector incorporates photomultiplier

TIRFM unit IN

TIRFM unit OUT

XY galvanometer mirror

TIRFM
Scan unit
Imaging software
CCD camera
IX81 inverted motorized microscope
Software for LSM

FV1000MPE

Multiphoton laser microscope for deep imaging.

With the FV1000MPE, a laser radiates high-density light at wavelengths up to several times the emission wavelength, causing only those fluorescent molecules located exactly at the focal point to become excited. Confocal-type optical sectioning can be achieved without the use of a pinhole, since light is not emitted from areas outside the focal plane. Also, the FV1000MPE allows deeper imaging of thickly sliced specimens as the system uses a near-infrared laser with superior tissue penetration.

Efficient fluorescence detection via external 2-channel system.

The FV1000MPE provides more efficient detection with minimal loss using a newly developed external two-channel photomultiplier detection system exclusively for two-photon. The system provides effective fluorescence detection without the need for a pinhole for de-scanning, because fluorescence excitation occurs only in the confocal plane.

- Clear morphological observation of thickly sliced specimens such as brain slice is achieved, which was not possible with conventional laser microscopes.
- When combined with a BX61WI fixed-stage upright microscope with motorized focusing, the laser microscope system can be used with a fixed stage and XY translation platform for patch clamping.

APPLICATIONS GALLERY
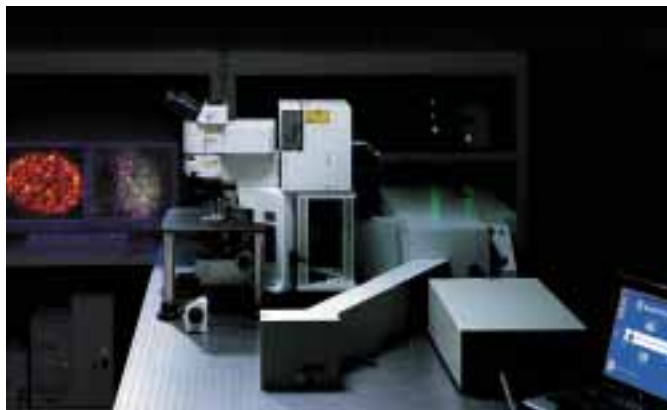
Cultured nerve cells derived from the mouse hippocampus

Mouse brain section
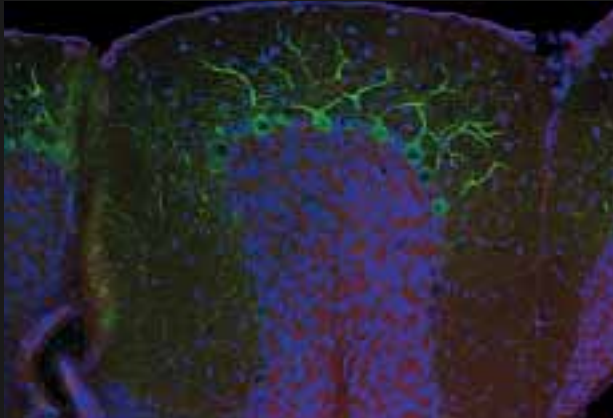
Mouse cerebellum
Green (AlexaFluor488): TTLL7
Magenta (AlexaFluor568): MAP2
Blue (TOTO-3): Nuclei
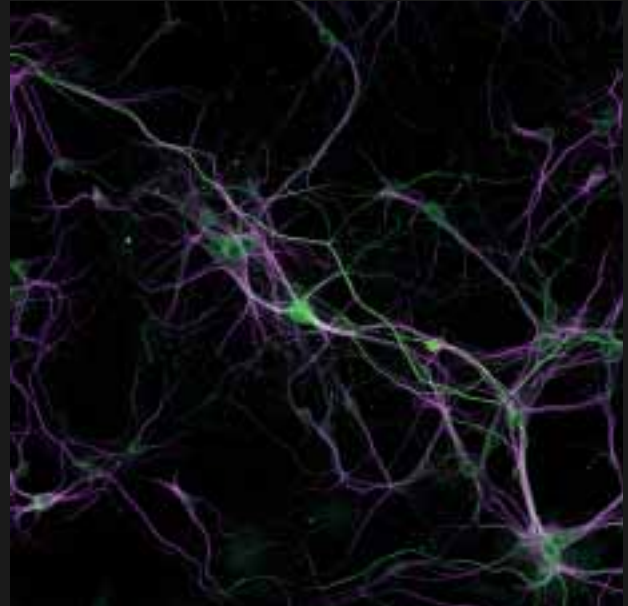
Green (AlexaFluor488): Protein χ
Magenta (AlexaFluor568): MAP2
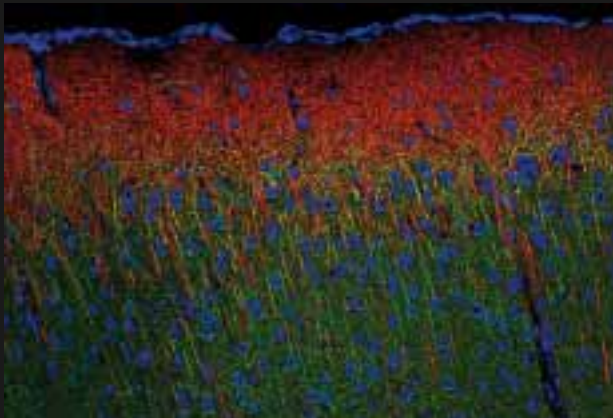
Mouse cerebral cortex
Green (AlexaFluor488):TTLL7
Magenta (AlexaFluor568): MAP2
Blue (TOTO-e): Nuclei

Dr. Koji Ikegami,
Dr. Mitsutoshi Setou
Molecular Geriatric Medicine,
Mitsubishi Kagaku Institute of Life Sciences

Mouse embryo: Posterior vena cava and somite
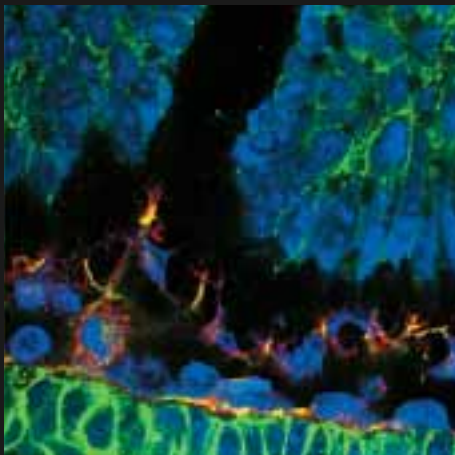
AlexFluor488: ß-Catenin
AlexFluor 546: Vascular endothelial
cell PECAM-1
TOTO-3: Nucleus

Data courtesy of:
Dr. Masanori Hirashima
Developmental Biology, School of
Medicine, Keio University

Drosophila

Drosophila, Stage 14
Green (FITC): Src42A(src of Drosophila)
Magenta (Cy3):E-cadherin

Rudimentary limbs of larva in latter part of 3rd instar
Green (FITC): POU-homeodomain protein Nubbin
Magenta (Cy3): Homeodomain protein Aristaless
Blue (Cy5): Homeodomain protein Bar

Cross-section viewing from dorsal side of wild-type embryo of
Drosophila in stage 17
Green (FITC): Src42A (src of Drosophila)
Magenta (Cy3): E-cadherin

Dr. Tetsuya Kojima
Laboratory of Innovational Biology,
Department of Integrated Biosciences
Graduate School of Frontier Sciences,
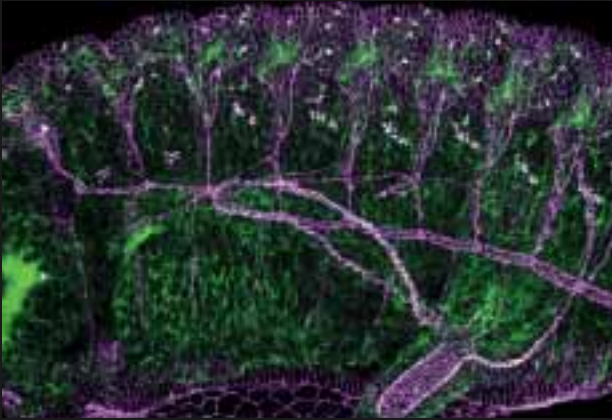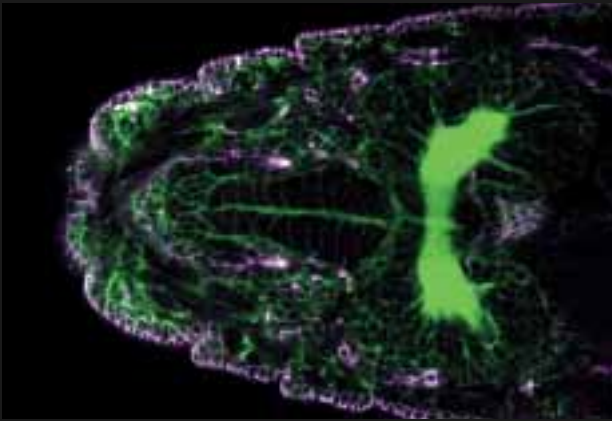*University of Tokyo*

Cell division (HeLa cell)
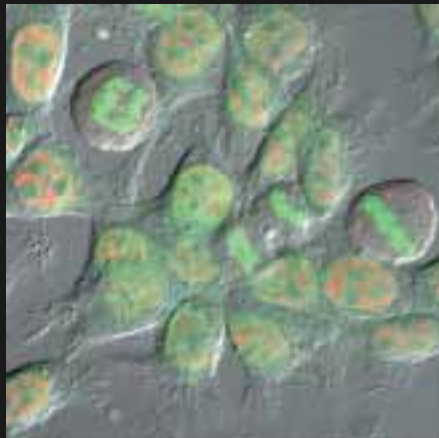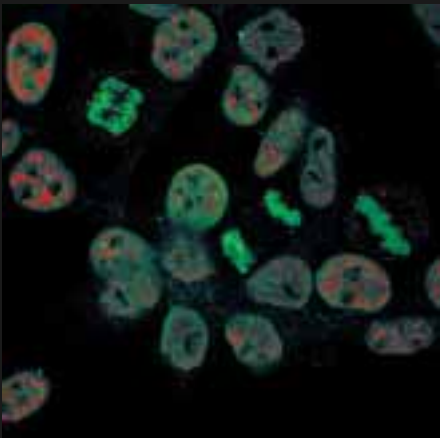
Immunostaining of HeLa cell is done using mouse monoclonal antibody with splicing factor SC35 and Alexa555 as secondary antibody. DNA is stained with DAPI. Characteristic patterns of intracellular localization are shown for interphase and mitotic period.
Green: DAPI (DNA)   Red: Alexa555 (SC35, splicing factor)

Data courtesy of: Dr. Takeshi Fukuhara,
Department of Functional Genomics,
Medical Research Institute & Laboratory of Gene Expression,
School of Biomedical Science, Tokyo Medical and Dental University Graduate School

APPLICATIONS GALLERY

Cerebellum Purkinje cell
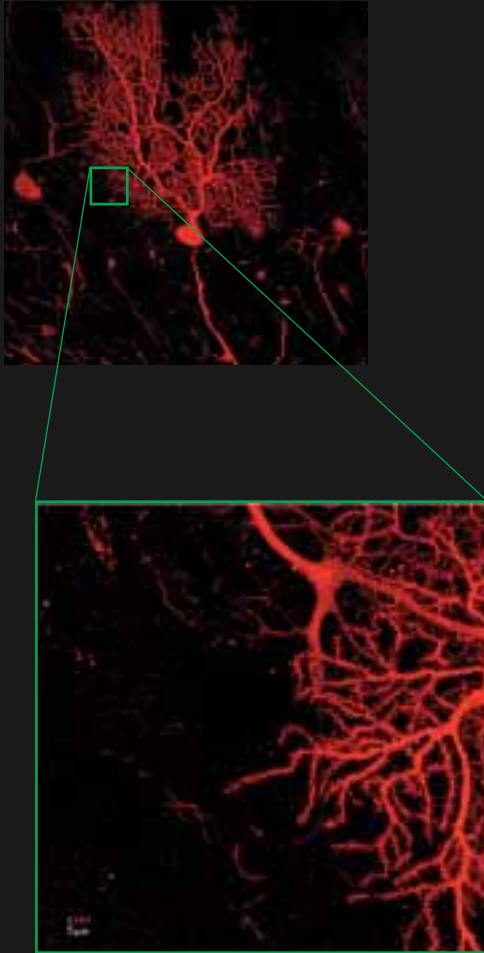
Visualized overall images of Purkinje cell axon, cell body and dendrites. Biotinylated-dextran amine was injected into mouse cerebellar nuclei. After a few days for fixing, they were sliced and anterograte labeled with TexasRed labeled avidin.

Mouse cerebellum climbing fiber
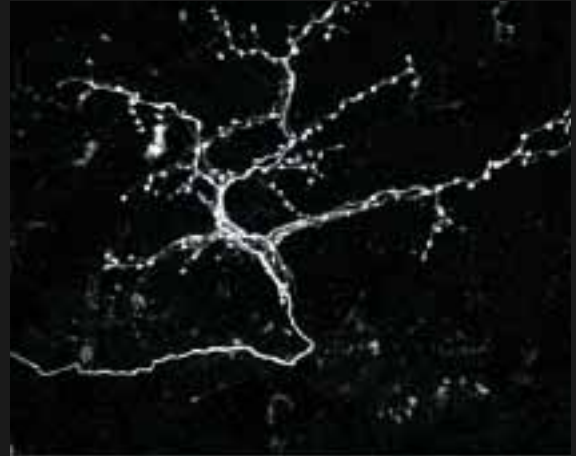
Visualized image of anterograde-labeled climbing fibers and their synapse terminal. Biotinylated-dextran amine (BDA) was injected into the inferior olivary nucleus of a mouse. After a few days for fixing, they were sliced thinly and dyed with TexasRed labeled avidin.

Data courtesy of:
Dr. Tetsuro Kashiwabara, Assistant Professor; and
Dr. Akira Mizoguchi, Professor;
Neuroregenerative medicine course,
Mie University School of Medicine

Rat kidney
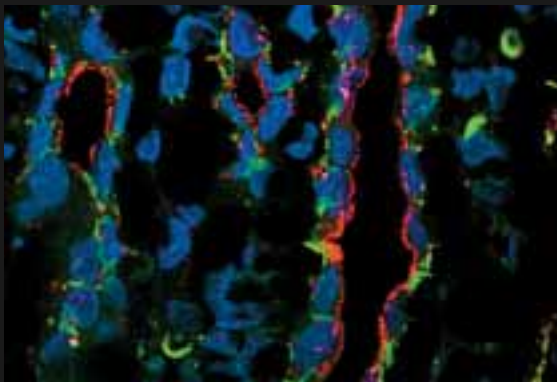
Blue (DAPI): Nuclei
Green (Qdot 525): Aquaporin water channels 3 (aquaporin 3; AQP3)
Red (Qdot 655): Aquaporin water channels 2 (aquaporin 2; AQP2)

Data courtesy of: Dr. Kuniaki Takada
Department of Anatomy and Cell Biology, Gunma University Graduate School of Medicine

Pilidium larva of Micrura alaskensis

Pilidium larva of Micrura alaskensis (Phylum Nemertea) with developing juvenile inside
Courtesy of Dr. Svetlana Maslakova of the University of Washington and Dr. Mikhail V Matz of the Whitney Laboratory for Marine Bioscience, University of Florida.

Mouse cerebellum
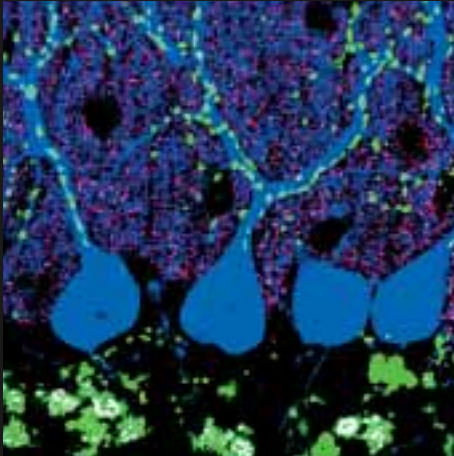
Triple stained image of mouse cerebellum using Calbindin (blue) FITC, VGluT1 (red) CY3, VGluT2 (green) CY5
Purkinje cell, parallel fiber terminal and climbing fiber terminal can be visualized.

Osteoclast induced from rat monocyte in rat kidney
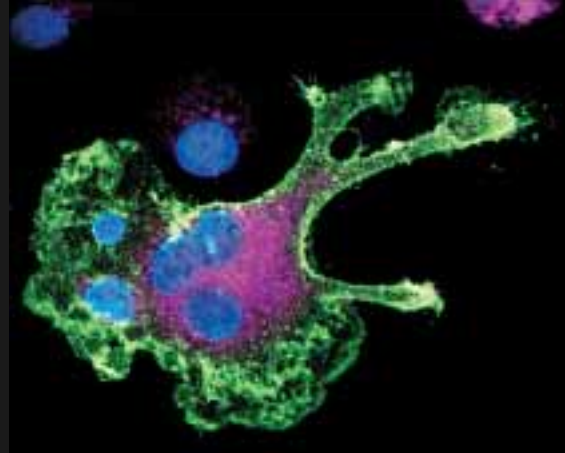
Green (AlexaFluor488): beta3 integrin
Magenta (TexasRed): Osteopontin
Blue (DAPI): Nucleus

Zebrafish

Objective: UPlanApo10X (approx. 300µm thick)
Red (DAPI): Nucleus
Green (AlexaFluor594): Actin

Expression of specific Cre gene in neural stem cell of cerebellum Purkinje cell
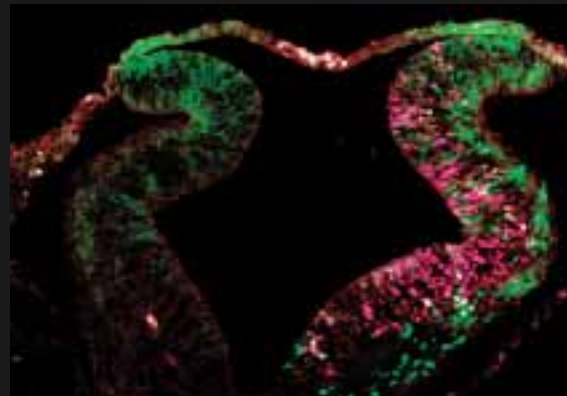
Cre gene is attached to the downstream of promoter of Nestin gene that can induce expression of a specific neural stem cell, and then introduced to the hindbrain of a chick embryo (on the right side of body only), using in ovo electroporation.
Fluorescence labeling: AlexaFluor 488 secondary antibody
EGFP to indicate electroporated side
AlexaFluor 594 secondary antibody to CRE

UNITS

Scanning units

Two types of scanning units, filter-based and spectral detection, are provided. The design is all-in-one, integrating the scanning unit, tube lens and pupil projection lens. Use of the microscope fluorescence illuminator light path ensures that expandability of the microscope itself is not limited. Visible, UV and IR laser introduction ports are provided, as well as a feedback control system.

Scanning unit for IX81 inverted microscope
Dedicated mirror unit cassette is required.

Scanning unit for BX61/BX61WI upright microscopes
Fluorescence illuminator integrated with scanning unit.

Laser systems

The multi-combiner enables combinations with all of the following diode lasers: 405nm, 440nm, 473nm, 559nm and 635nm. The system can also be equipped with conventional Multi-line Ar laser and HeNeG laser.

Double type
The multi-combiner outputs laser light with two fibers. Light can be used both for observation and laser light stimulation.

Single type
Single channel laser output. A0TF is standard equipment.

Illumination units

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.

Fluorescence illumination module
Stand with Mercury lamp house, motorized shutter, and fiber delivery system for conventional fluorescence observation. Light introduction via fiber optic port.

Transmitted light detection module
External transmitted light photomultiplier detector and 100W Halogen conventional illumination, integrated for both laser scanning and conventional transmitted light Nomarski DIC observation. Motorized exchange between transmitted light illumination and laser detection. Simultaneous multi-channel confocal fluorescence image and transmitted DIC acquisition enabled.

Optional upgrade modules for FV1000

4th channel detector unit
Attaches to the optional port of either the filter or spectral type scanning unit and is used as a 4th confocal fluorescence detection channel. This is a filter-based fluorescence detection unit.

SIM Scanner
Second scanner dedicated for laser light stimulation, synchronized to the FV1000 main scanner for simultaneous laser stimulation and confocal image acquisition. Independent fiber optic laser introduction port. Dichromatic mirror within motorized optical port of the scan unit required for introduction of laser into main scanner.

TIRFM unit
Enables control of the necessary volume of excitation light using FV1000 software. This unit enables TIRF observation using the laser light source used with Confocal.

Fiber port for fluorescence output
Confocal fluorescence emission can be introduced via fiber delivery system into external device. Fiber port equipped with FC connector (fiber delivery system not included).

CO₂ incubator to prolong live cell activity

Highly precise incubator control keeps the environment inside a laboratory dish completely stable, at just below 37°C temperature, 90% moisture and 5% CO_2 concentration; in this way, live cell activity can be maintained for approximately two days. A specially designed structure is employed to minimize the focus drift during temperature control. This is the ideal solution for time-lapse experiments under both a confocal laser scanning microscope and a wide field observation. The opening hole located on the top heater is available for the cell injection.

* Not available in some areas

MI-IBC-IF

High grade configuration with a built-in flowmeter for 5% CO_2 and 95% air.
* Built-in stage warming plate
* Objective heater.
* 5% CO_2 supply tube with ø4 outer diameter, ø2 inner diameter and 400mm length.

MI-IBC-IF

FV1000 SYSTEM DIAGRAM

Fluorescence illumination unit

**Scanning unit for IX81
(Spectral type or Filter type detector system)**

IX81
Inverted motorized microscope

Cover *

TIRFM unit *

SIM Scanner*

LD635 laser
635nm

LD559 laser
559nm

HeNeG laser
543nm
Select either laser

Multi Ar laser
548, 488, 515nm

LD473 laser
473nm
Select either laser

LD440 laser*
440nm

LD405 laser*
405nm

**AOTF Laser combiner
(with laser heads)**

**Scanning unit for BX61WI, BX61
(Spectral type or Filter type detector system)**

**BX61WI
BX61**
Upright motorized microscope

FV Power supply *

IR laser*

Transmitted light detection unit

Microscope control unit

Software
ASW: Application software for FV1000 Ver.1
Review station software*
Multi point software*

Fiber port for fluorescence output*

4th channel detector unit*

FV power supply unit

FV control unit

Monitor

*Optional unit

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FLUORESCENCE DYES

351 UV Argon
405 LD
440 LD
458 Multi Argon
488 Argon
488 Multi Argon
515 Multi Argon
543 Green HeNe
568 Krypton / Argon
633 Red HeNe

300 350 400 450 500 550 600 650 700 750 800

indo-1
Hoechst33258
DAPI
ECFP
Alexa488
Cy2
AcridineOrange
AzamiGreen
DiO
FITC
fluo-4
GFP-uv
YOYO1
EGFP
OregonGreen488
fluo-3
Kaede
RhodamineGreen
CalciumGreen
MagnesiumGreen
EYFP
furaRed
SNARF-1
Alexa546
DiI
RhodaminePhalloidin
TRITC
CalciumOrange
Cy3
MagnesiumOrange
rhod-2
Cy35
DsRed2
PI
RhodamineRedX
X-rhod-1
MitoTracker
CalciumCrimson
Alexa568
HcRed1
TexasRed
Alexa594
Alexa633
TOTO3
Alexa647
Cy5
Cy55

MAIN SPECIFICATIONS

		Spectral type fluorescence detector	Filter type fluorescence detector
Laser light	Ultraviolet/Visible light laser	LD lasers: 405nm: 50mW, 440nm: 25mW, 473nm: 15mW, 559nm: 15mW, 635mW, 20mW Multi-line Ar laser (457nm, 488nm, 515nm, Total 30mW), HeNe(G) laser (543nm, 1mW)	
	AOTF laser combiner	Visible light laser platform with implemented AOTF system, Ultra-fast intensity modulation with individual laser lines, additional shutter control Continuously variable (0.1% - 100%, 0.1% increment), REX: Capable of laser intensity adjustment and laser wavelength selection for each region	
	Fiber	Broadband type (400nm-650nm)	
Scanning and Detection	Scanner module	Standard 3 laser ports, VIS - UV - IR Excitation dichromatic mirror turret, 6 position (High performance DMs and 20/80 half mirror), Dual galvanometer mirror scanner (X, Y) Motorized optical port for fluorescence illumination and optional module adaptation, Adaptation to microscope fluorescence condenser	
	Detector module	Standard 3 confocal Channels (3 photomultiplier detectors) Additional optional output port light path available for optional units 6 position beamsplitter turrets with CH1 and CH2 CH1 and CH2 equipped with independent grating and slit for fast and flexible spectral detection Selectable wavelength bandwidth: 1-100nm Wavelength resolution: 2nm. Wavelength switching speed: 100nm/msec CH3 with 6 position barrier filter turret	Standard 3 confocal Channels (3 photomultiplier detectors) Additional optional output port light path available for optional units 6 position beamsplitter turrets with CH1 and CH2 CH1 to CH3 each with 6 position barrier filter turret (High performance filters.)
	Filters	High performance sputtered filters, dichromatic mirrors and barrier filters	
	Scanning method	2 galvanometer scanning mirrors	
	Scanning modes	Pixel size: 64 x 64 — 4096 x 4096 Scanning speed: 512 x 512 (1.1 sec., 1.6 sec., 2.7 sec., 3.3 sec., 3.9 sec., 5.9 sec., 11.3 sec., 27.4 sec., 54.0 sec.) 256 x 256 bidirectional scanning (0.064 sec., 0.129 sec.) 0.5 or 1 microsec dwell time per point for fast bidirectional scanning	
		X,Y,T,Z,λ Line scanning: Straight line with free orientation, free line, Point scanning	X,Y,T,Z Line scanning: Straight line with free orientation, free line, Point scanning
	Photo detection method	2 detection modes: Analog integration and hybrid photon counting	
	Pinhole	Single motorized pinhole pinhole diameter ø50 - 300µm (0.5µm step)	Single motorized pinhole pinhole diameter ø50 - 800µm (0.5µm step)
	Field Number (N.A.)	18	
	Optical Zoom	1x - 50x in 0.1x increment	
	Z-drive	Integrated motorized focus module of the microscope, minimum increment 0.01µm or 10 nm	
	Transmitted light detector unit	Module with integrated external transmitted light photomultiplier detector and 100W Halogen lamp, motorized switching, fiber adaptation to microscope frame	
Microscope	Motorized microscope	Inverted IX81, Upright BX61, Upright focusing nosepiece & fixed stage BX61WI	
	Fluorescence illumination unit	External fluorescence light source with motorized shutter, fiber adaptation to optical port of scan unit Motorized switching between LSM light path and fluorescence illumination	
System Control	PC	PC-AT compatible, OS: Windows XP Professional (English version), Memory: 2GB or larger, CPU:Pentium 3.2GHz or higher, Hard disk: 120GB or larger, Media: DVD Multi Drive, FV1000 Special I/F board (built-in PC), Graphic board: ATI RADEON X700 PRO	
	Power Supply Unit	Galvo control boards, scanning mirrors and gratings, Real time controller	Galvo control boards, scanning mirrors
	Monitor	Monitor: SXGA 1280X1024, dual 19 inch or larger monitors	
Optional unit	SIM Scanner	2 galvanometer scanning mirrors, pupil projection lens, built-in laser shutter, 1 laser port Fiber introduction of near UV diode laser or visible light laser, Optional: 2nd AOTF laser combiner	
	TIRFM unit	Available laser: 405~633nm. Motorized penetration ratio adjustment. Automatic optical setting for TIRFM objectives.	
	4th CH detector	Module with photomultiplier detector, barrier filter turret, beamsplitter turret mounted with 3rd CH light path	
	Fiber port for fluorescence	Output port equipped with FC fiber connector (compatible fiber core 100 -125µm)	

Software	
Display	Dual monitor display
Optional Hardware	SIM scanner unit, 4ch detector. Fiber port
Image format	OIB/OIF image format 8/16 bit gray scale/index color, 24/32/48 bit color, JPEG/BMP/TIFF/AVI/MOV image functions Olympus multi-tif format
Image acquisition	Region designation: point, line, free line, clip, clip zoom Real-time image calculation: Kalman filtering, photon counting mode 2-dimension: XY, XZ, XT and Xλ 3-dimension: XYZ, XYT, XYλ, XZT, XTλ and XZλ 4-dimension: XYZT, XZTλ and XYTλ
Image pixel format	64 x 64 — 4096 x 4096
Programmable Time Controller function	Time Controller function
Image display	Each image display: Single-channel side-by-side, merge, cropping, live tiling, live tile, series (Z/T/λ), LUT: individual color setting, pseudo-color, comment: graphic and text input
3D visualization and observation	Interactive volume rendering Free orientation of cross section display 3D animation, left/right stereo pairs, red/green stereoscopic images and cross section 3D and 2D sequential operation function
Spectral unmixing	2 Fluorescence spectral unmixing modes (normal and blind mode)
Image processing	Individual filter: average, low-pass, Sobel, Median, *Prewitt, 2D Laplacian, edge enhancement etc. Calculations: inter-image, mathematical and logical, DIC background leveling
Image analysis	Fluorescence intensity, area and perimeter measurement, time-lapse measurement
Statistical processing	2D data histogram display, colocalization
Others (options)	Off-line review station software

Objectives for BX and IX (using U-UCD8A, IX-LWUCDA and U-DICTS)

| Description	N.A.	W.D.	Cover glass thickness	Immersion	Correction ring	Condenser for BX U-UCD8A optical element	Condenser for IX IX-LWUCDA optical element	U-DICTS position
UPLSAPO 4x	0.16	13	—					
UPLSAPO 10x	0.40	3.1	0.17			U-DIC10	IX2-DIC10	normal
UPLAPO 10xO3	0.40	0.24	0.17	Oil		U-DP10	IX-DP10	normal
UPLAPO 10xW3	0.40	0.43	0.17	Water		U-DP10	IX-DP10	normal
UPLSAPO 20x	0.75	0.6	0.17			U-DIC20	IX2-DIC20	normal
UPLSAPO 20xO	0.85	0.17	—	Oil		U-DIC20	IX2-DIC20	normal
UPLSAPO 40x	0.90	0.2	0.11-0.23		○	U-DIC40	IX2-DIC40	normal
UPLFLN 40xO	1.30	0.2	0.17	Oil		U-DIC40	IX2-DIC40	BFP1
PLAPON 60xOTIRFM	1.45	0.10	0.17	Oil		U-DIC60	IX2-DIC60	BFP1
UPLSAPO 60xO	1.35	0.15	0.17	Oil		U-DIC60	IX2-DIC60	BFP1
UPLSAPO 60xW	1.20	0.28	0.15-0.2	Water	○	U-DIC60	IX2-DIC60	normal
UPLSAPO 100xO	1.40	0.12	0.17	Oil		U-DIC100	IX2-DIC100	normal

Objectives for fixed stage upright microscopes (using WI-UCD, WI-DICTHRA)

Objective	N.A.	W.D.	DIC prism	Revolving nosepiece
MPlan5x	0.10	19.60	—	WI-SSNP, WI-SRE2
UMPlanFl10xW	0.30	3.30	U-LDPW10H	WI-SSNP, WI-SRE2
UMPlanFl20xW	0.50	3.30	U-LDPW20H	WI-SSNP, WI-SRE2
LUMPlanFl40xW	0.80	3.30	U-LDPW40H	WI-SSNP, WI-SRE2
LUMPlanFl60xW	0.90	2.00	U-LDPW60H	WI-SSNP, WI-SRE2
LUMPlanFl40xW/IR2	0.80	3.30	U-LDPW40H	WI-SSNP, WI-SRE2
LUMPlanFl60xW/IR2	0.90	2.00	U-LDPW60H	WI-SSNP, WI-SRE2
LUMPlanFl100xW	1.00	1.50	U-LDPW60H	WI-SSNP, WI-SRE2
XLUMPlanFl20xW	0.95*	2.00	U-LDPXLU20 HR	WI-SNPXLU

* Note: These conditions are not met in confocal microscopy

Recommended FV1000 system setup (IX81, BX61, BX61WI)

(unit: mm)
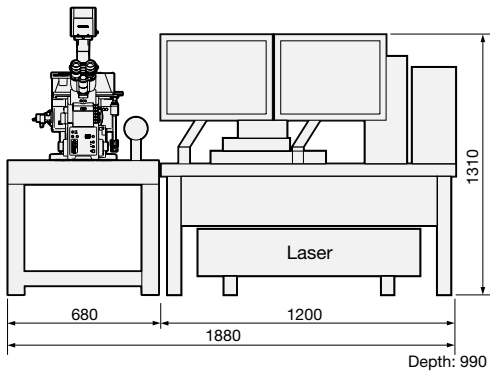
Dimensions, weight and power consumption

	Dimensions (mm)	Weight (kg)	Power consumption
Microscope with scan unit	BX system 320 (W) x 580 (D) x 565 (H) IX system 350 (W) x 750 (D) x 640 (H)	41 51	—
Fluorescence illumination unit	Lamp 180 (W) x 320 (D) x 235 (H) Power supply 150 (W) x 295 (D) x 200 (H)	6.7 4.6	100-120V/200-240V 260VA
Transmitted light detection unit	170 (W) x 330 (D) x 130 (H)	5.9	—
Microscope control unit	125 (W) x 330 (D) x 215 (H)	5.8	100-120V/200-240V 350VA
FV Power supply unit	180 (W) x 328 (D) x 424 (H)	7.5	100-120V/200-240V 400VA
FV control unit (PC)	180 (W) x 420 (D) x 360 (H)	11.2	100-120V/200-240V 300VA
Monitor (dual)	415 (W) x 216 (D) x 435 (H)	6.6	100-120V/200-240V 100VA
Power supply unit for laser combiner	210 (W) x 300 (D) X 100 (H)	4.0	100-120V/200-240V 400VA
Laser combiner (with Ar laser heads)	514 (W) x 504 (D) X 236 (H)	45	—
Laser combiner (without Ar laser heads)	514 (W) x 504 (D) X 236 (H)	40	—
LD559 laser power supply	200 (W) x 330 (D) x 52 (H)	1.2	100-120V/200-240V 30VA
Multi Ar laser power supply	162 (W) x 330 (D) x 98 (H)	3.2	100-120V/200-240V 1500VA
HeNe G laser Power Supply	160 (W) x 270 (D) x 54 (H)	1.4	100-120V/200-240V 30VA

FLUOVIEW website

www.olympusfluoview.com

Image on cover page:
Immunolabeling of a transgenic mouse retina showing the major retinal cells types

Courtesy of:
Dr. Rachel Wong
Mr. Josh Morgan
Dept. Biological Structure, University of Washington, Seattle.

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