OLYMPUS®

Your Vision, Our Future

CONFOCAL LASER SCANNING
BIOLOGICAL MICROSCOPE

FV1000

FLUOVIEW

Next Generation Live Cell Imaging System

Simultaneous Laser Light Stimulation and Imaging

The FV1000 incorporates 2 laser scanners in a single compact design for simultaneous confocal fluorescence observation and independent laser light stimulation. Synchronization of these two functions ensures that cellular reactions that occur during or immediately following stimulation are not overlooked, and makes the FV1000 the most suitable microscope for FRAP, FLIP and photo activation.

SIM Scanner System Captures Reactions During Laser Stimulation

SIM SCANNER
FRAP

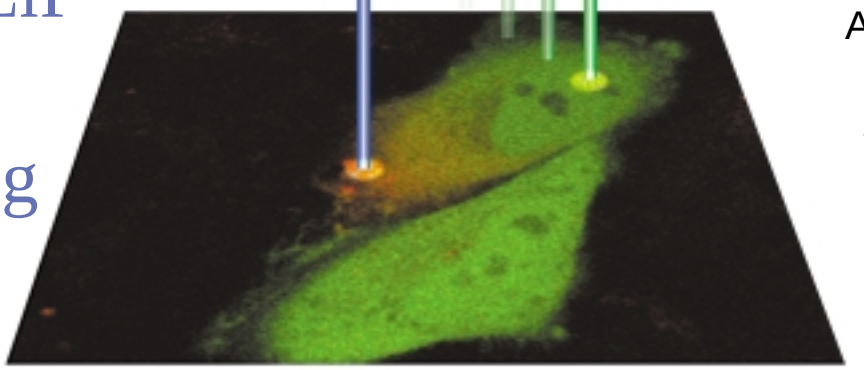
LIVE CELL

SIM SCANNER
FLIP

SIM SCANNER
Uncaging

Laser for stimulation

Laser for imaging

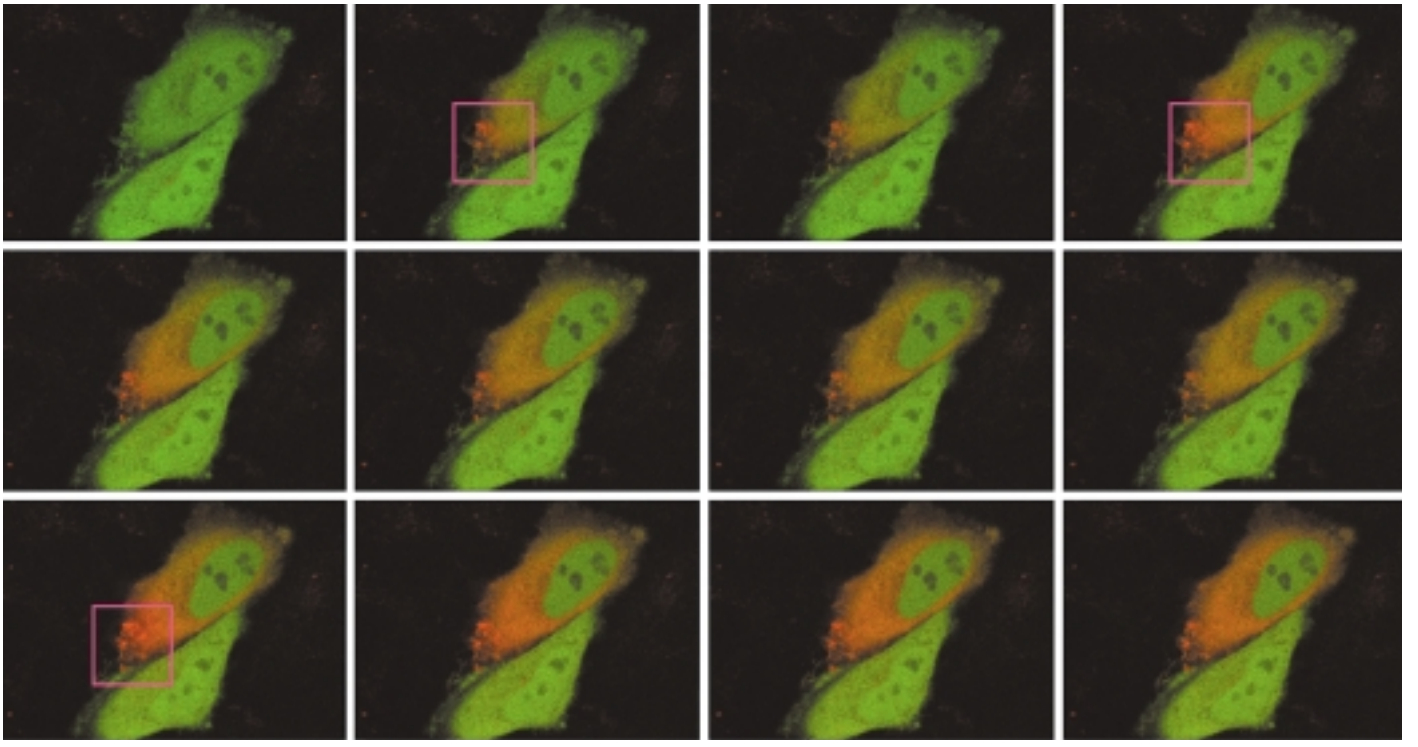


SIM SCANNER
kaede

New Fluorescent Protein Applications

SIM SCANNER
PA-GFP

Protein diffusion in Kaede-expressing HeLa cells can be accurately monitored using the SIM Scanner System. During 405nm laser stimulation within a user-defined region (red box, frames 2, 4, and 9), the fluorescence emission peak of the Kaede protein shifts from green to red. Confocal fluorescence images recorded every 3 seconds using 488/543nm laser excitation demonstrate the spread of the activated Kaede protein (red) throughout the individual HeLa cell.
Data courtesy of: Ms. Ryoko Ando and Dr. Atsushi Miyawaki
RIKEN Brain Science Institute Laboratory for Cell Function Dynamics



Brighter, faster, more precise

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

Opening New Frontiers

Live Cell Imaging System

FV1000

High Sensitivity

- Efficient fluorescence detection via ion deposition filters
- Newly developed, high-sensitivity detection system

High Speed

- High-speed imaging, 16 frames/sec at 256 x 256 image format (4000Hz)
- High-speed spectroscopy, 100 nm/msec

High Precision

- 2nm spectral wavelength resolution, linear spectral distribution
- Stable excitation light via advanced laser intensity feedback system

Laser Light Stimulation

- Simultaneous laser stimulation and imaging via SIM (SIMultaneous) Scanner, with wavelengths ranging from UV to IR.

FV1000 configuration with BX61 microscope

FV1000 configuration with IX81 microscope

Sensitivity, Speed and Precision that Redefine Basic Performance

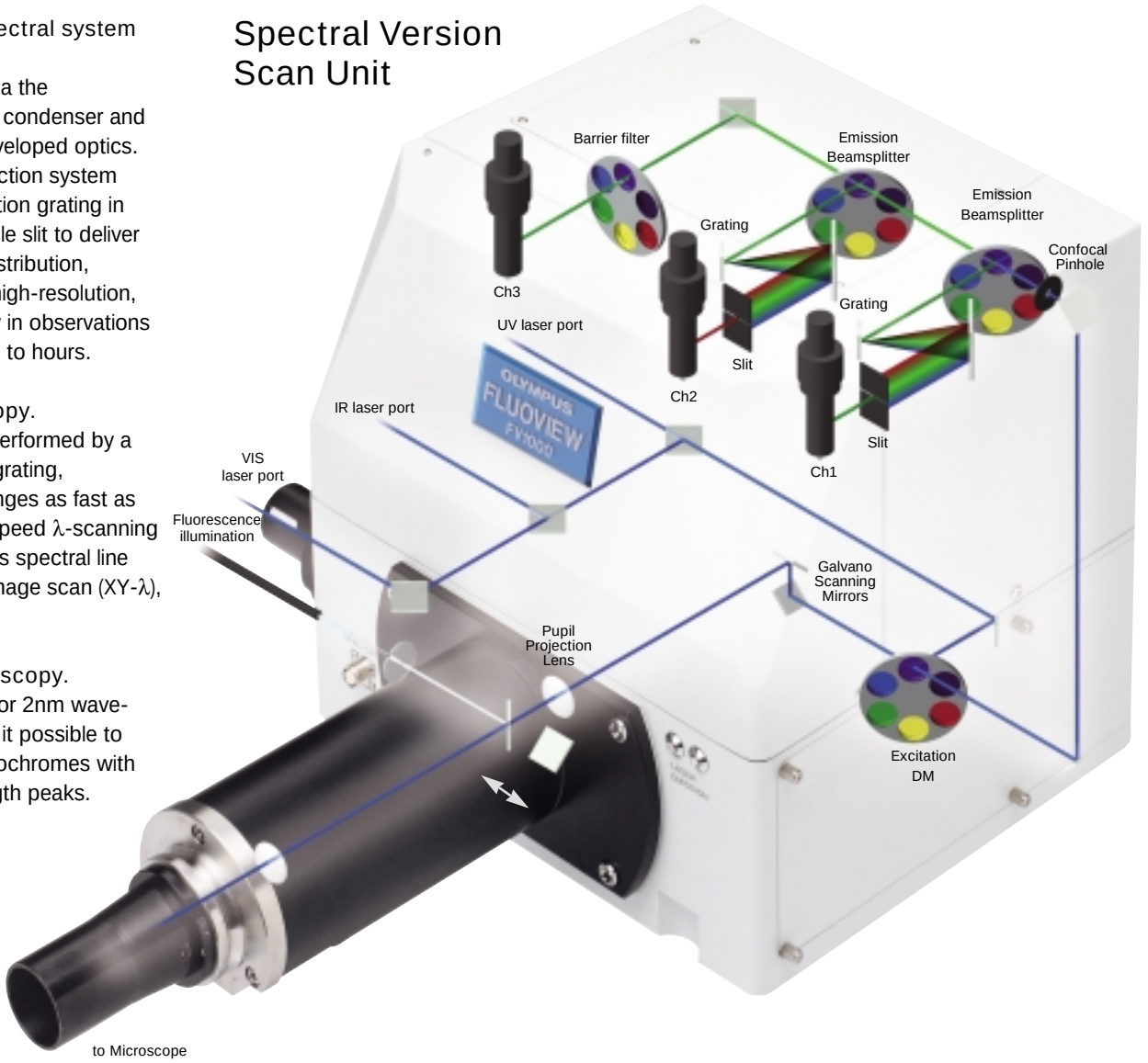
A unique spectral system — sensitive, fast and accurate

Original optical and spectral system design.
The FV1000 is adapted via the microscope fluorescence condenser and incorporates specially developed optics. The original spectral detection system uses a high speed diffraction grating in combination with a variable slit to deliver superior linear spectral distribution, enabling high-precision, high-resolution, high-speed spectroscopy in observations ranging from milliseconds to hours.

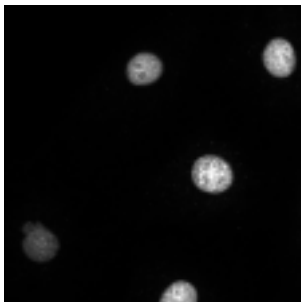
High-speed spectroscopy.
Wavelength selection is performed by a galvanometer diffraction grating, enabling wavelength changes as fast as 100 nm/msec and high-speed λ-scanning image acquisition, such as spectral line scan (X-λ) and spectral image scan (XY-λ), with 1 nm step setting.

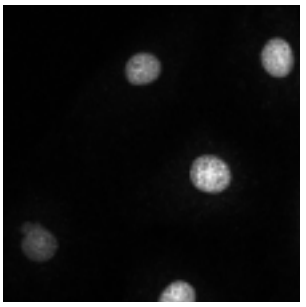
High-precision spectroscopy.
The FV1000 offers superior 2nm wavelength resolution, making it possible to clearly separate two fluorochromes with similar emission wavelength peaks.

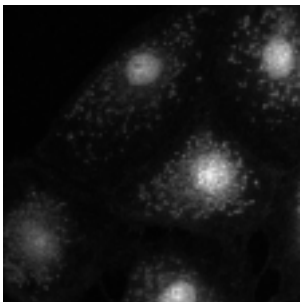
Spectral Version
Scan Unit

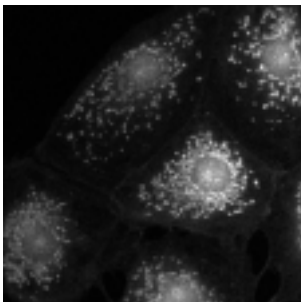


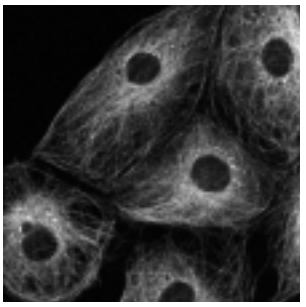
Excitation DM488/543/633 comparison


495nm-515nm

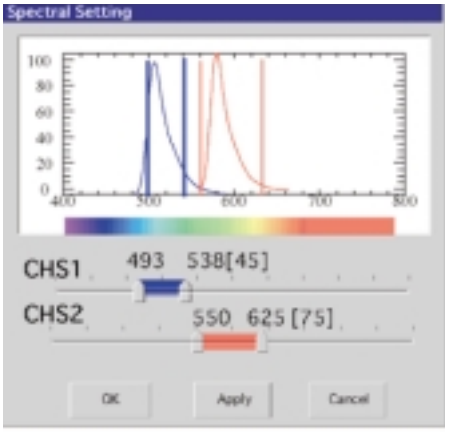

565nm-585nm


575nm-595nm


595nm-615nm


675nm-695nm

Variable wavelength setting.
The ideal emission bandwidth can be automatically set for each fluorochrome using the on-line dye database. In addition, an intuitive menu allows manual setting of the spectral detector's variable bandwidth, permitting the fluorescence detection bandwidth to be optimized for the emission characteristics of each fluorescent dye.



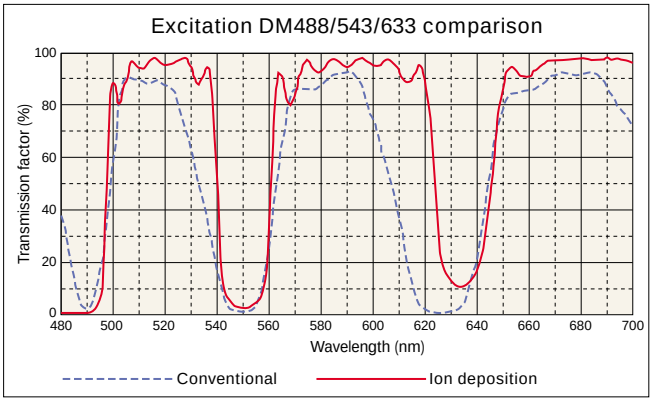
High Sensitivity Detection System.
The FV1000 features two user-selectable detection modes, a low noise analog integration mode and an original hybrid photon counting detection mode.

XYλ series, selected images
XYλ image series of PtK2 cells, triple stained with YOYO-1 for nuclei, MitoTracker Red for mitochondria, and Cy5-conjugated anti-tubulin.
Spectral acquisition ranged from 495nm to 700nm, in 10nm increments with 20nm bandwidth.

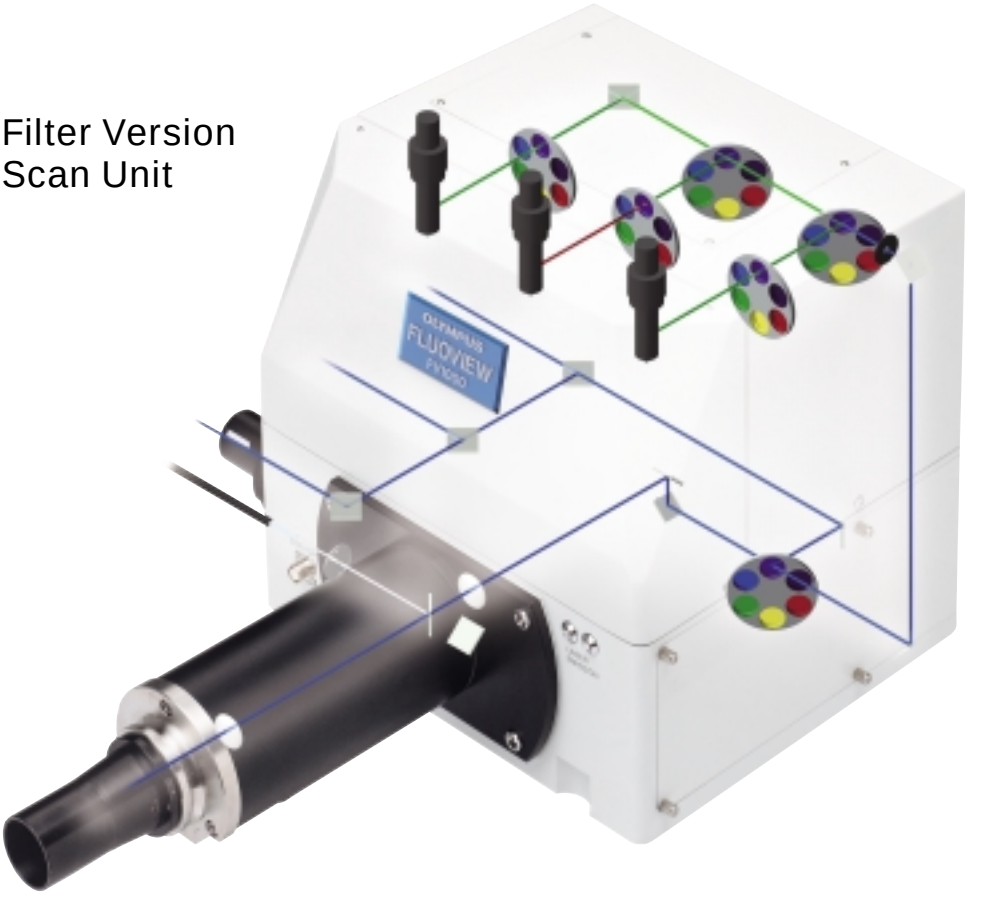
Highly-sensitive filter detection system minimizes cell damage

Ion deposition filters for increased sensitivity and full wavelength coverage.
All FV1000 filters feature an ion sputter coating applied by ion deposition technology. This original Olympus technology provides filters with superior curve steepness and wavelength band transmission compared to conventional vacuum deposition.
The resulting extended emission bandwidths and increased transmission efficiency enable observation with reduced excitation laser intensity, minimizing photo damage to living cells.

Highly-sensitive detection.
A pupil projection lens, highly sensitive photomultiplier and low-noise analog processing circuit are integrated within the optical system, providing enhanced performance sensitivity with a high S/N ratio.



Filter Version Scan Unit

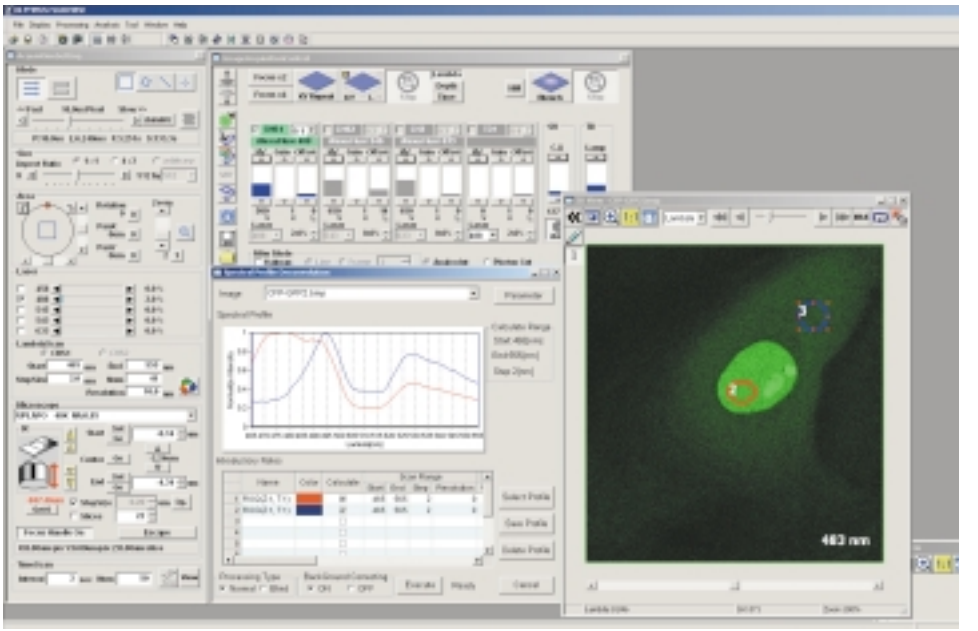


Intuitive Operation Makes Experiments Easy to Design, Easy to Execute

Intuitive software.
Spectral un-mixing for complete assurance.

Intuitive, easy-to-understand GUI.
The operator can easily make on-line adjustments to the acquisition parameters (such as scan speed, optical zoom, scan area rotation, photomultiplier voltage, gain, and offset) while monitoring the image resolution. Automatic contrast mode makes image acquisition very easy.

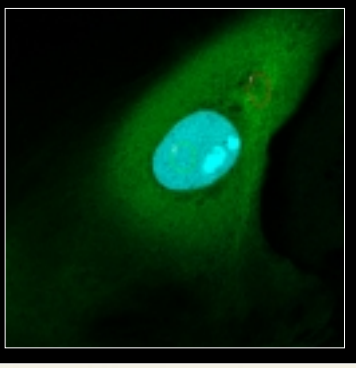
Spectral Un-Mixing.
For separating fluorochromes with similar emission profiles, two modes are provided: Normal mode, which requires the input of the fluorochrome profile, and Blind mode, which automatically separates the emission profiles based on information derived from the specimen image alone. With superior 2 nm wavelength resolution, fast and accurate spectral un-mixing can be easily performed, even for fluo-rochromes without a known fluorescence profile, such as autofluorescence.

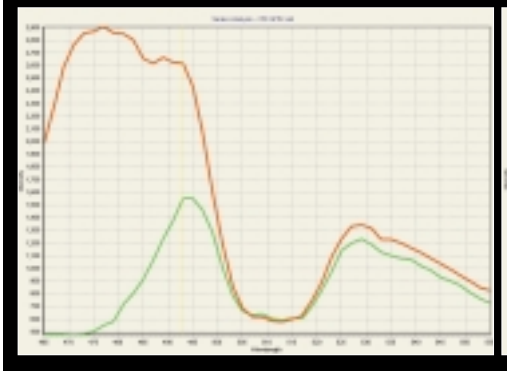


Spectral characteristics of CFP and GFP are measured using two regions of interest to designate the CFP profile (red) and the GFP profile (blue), and then used in the Normal mode for spectral un-mixing of the two fluorochromes.

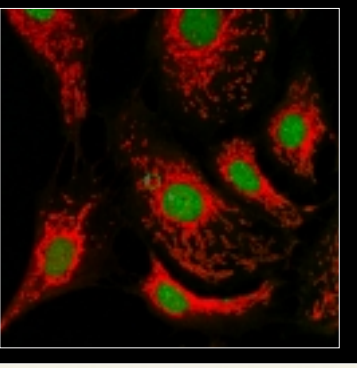
Un-Mixing images

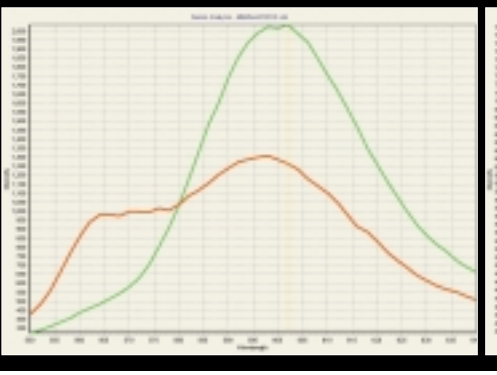
CFP (nucleus) — GFP (tubulin) HeLa cell
XYλ acquisition conditions
Wavelength detection range: 465nm~565nm in 2nm steps
Excitation wavelength: 457nm



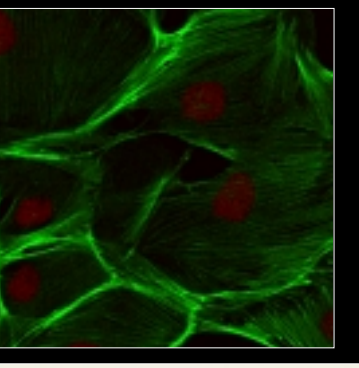


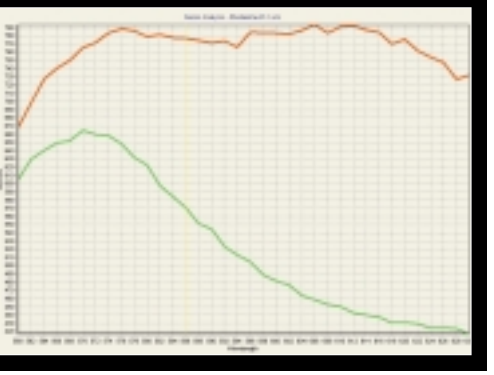
MitoTracker (mitochondria) — POPO-3 (nucleus)
XYλ acquisition conditions
Wavelength detection range: 550nm~640nm in 2nm steps
Excitation wavelength: 543nm



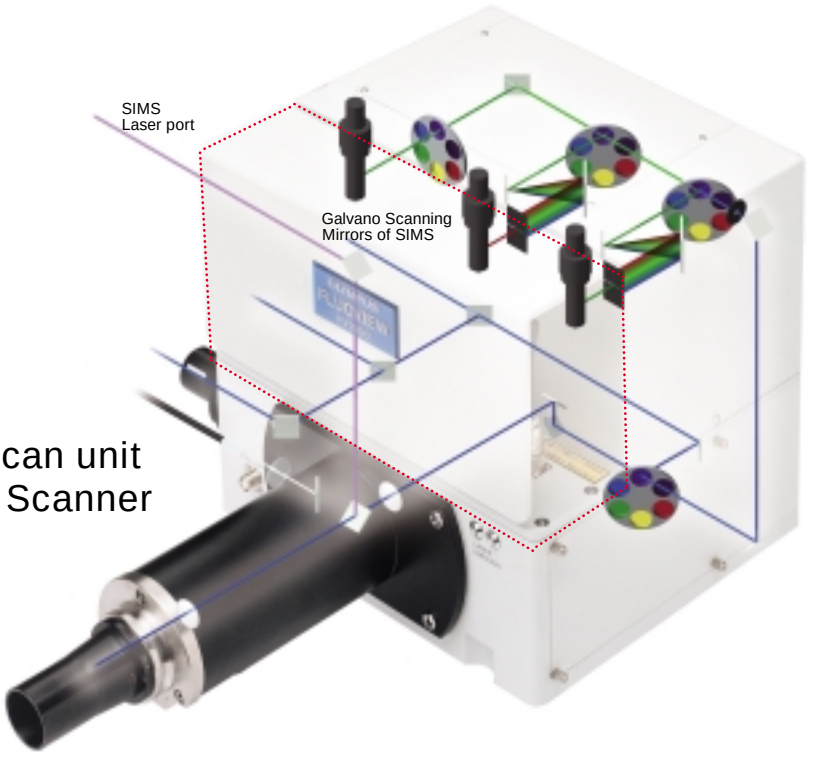


Rhodamine-Phalloidin (actin) — PI (nucleus)
XYλ acquisition conditions
Wavelength detection range: 560nm~630nm in 2nm steps
Excitation wavelength: 543nm



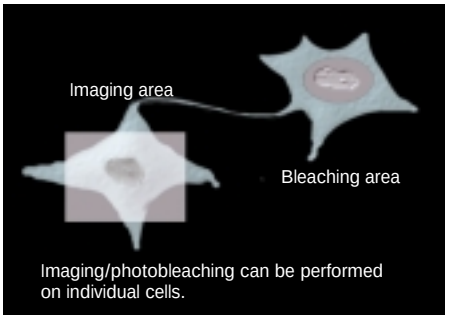


SIM (SIMultaneous) Scanner Syste
Synchronizes laser light stimulatic
confocal imaging (option)



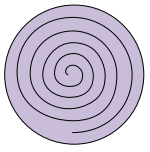
FV1000 scan unit
with SIM Scanner

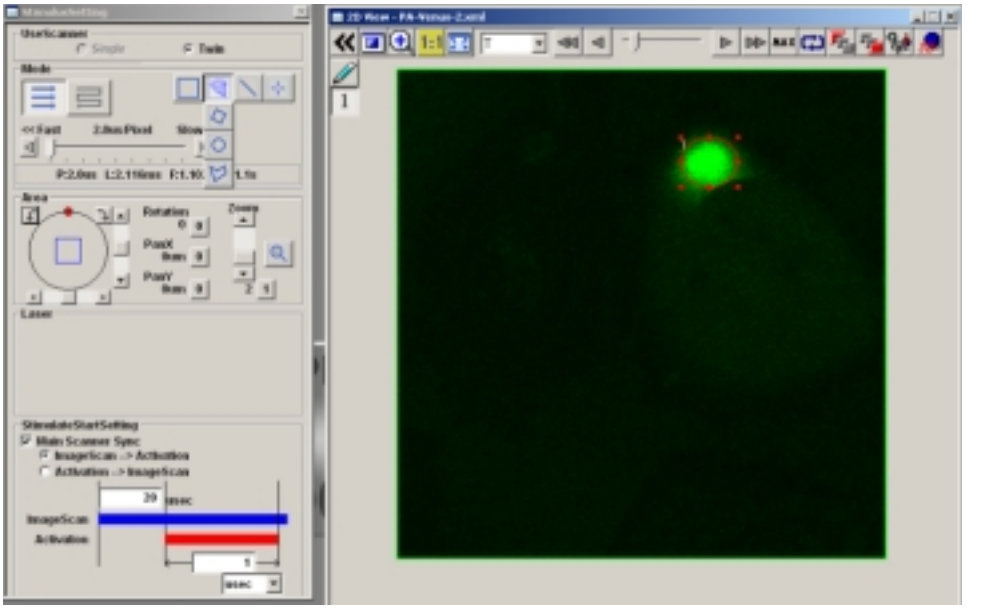
Sure capture of reactions immediately following light stimulation.
In addition to the primary scanner for confocal observation, the FV1000 incorporates a compact 2nd scanner dedicated for laser light stimulation. With 2 synchronized scanners, confocal image observation is no longer interrupted during laser light stimulation or laser manipulation, and fluorescence changes that occur during or immediately following laser stimulation are no longer overlooked. This unique capability offers distinct advantages for a variety of applications, such as FLIP, FRAP, photo activation, photo conversion, uncaging, etc.



Imaging/photobleaching can be performed on individual cells.

Tornado scanning for highly efficient bleaching.
For photobleaching applications, conventional raster scanning tends to be slow and often results in inadequate bleaching. However, with the patent-pending Tornado Scan, fast and efficient bleaching is achieved by the SIM Scanner through a unique circular scanning mode that significantly reduces the necessary laser exposure time.


Tornado scanning



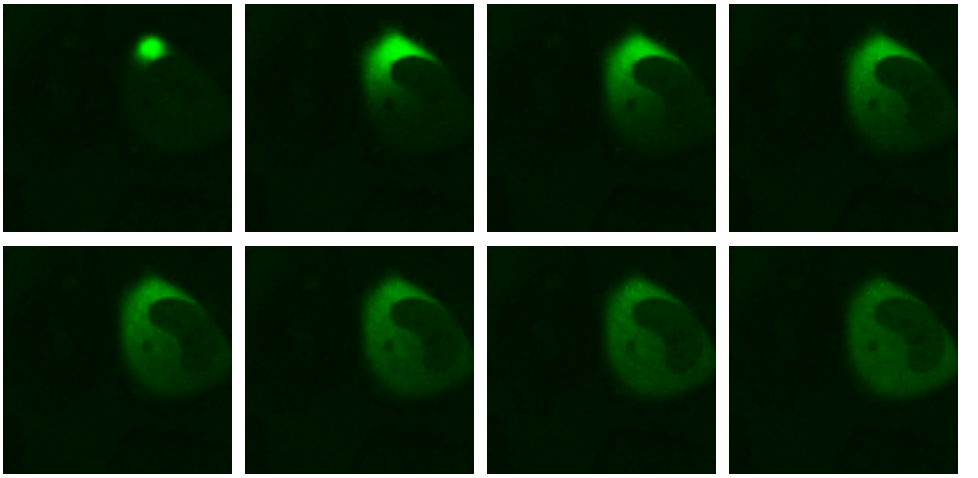


Photo activation of PA-GFP expressing HeLa cells. SIMS stimulation (red ROI) using the 405nm diode laser, releasing GFP within the cell. GFP images were simultaneously acquired at 1 second intervals using the 488nm laser.

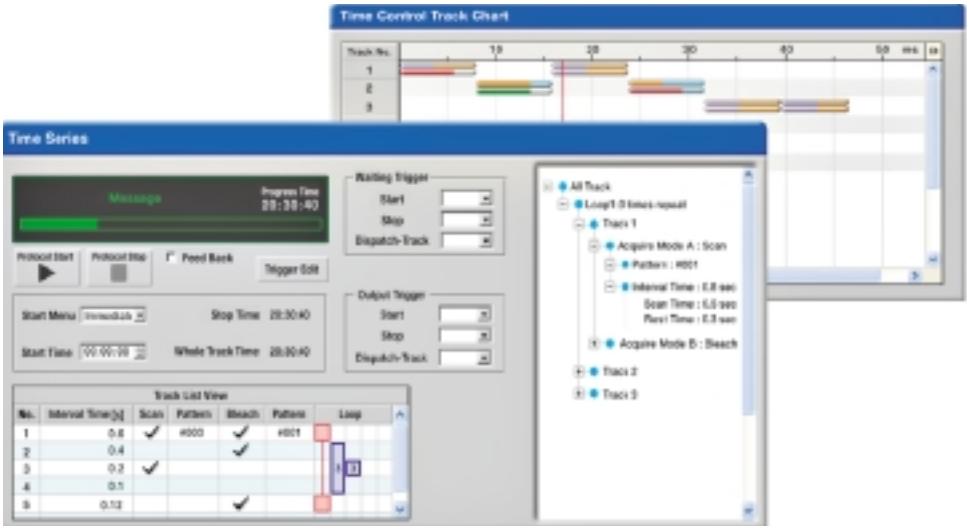
Data courtesy of Dr. Atsushi Miyawaki, Dr. Takeharu Nagai, Dr. Takayuki Miyauchi
RIKEN Brain Science Institute Laboratory for Cell Function Dynamics

High-speed, high-precision detection with time-lapse experiments

16 frames/sec. 4000 lines/sec.
The incorporation of highly stable galvano-meter scanning mirrors and an Olympus original acceleration circuit enables high-speed imaging (16 frames per second at 256x256 image format). Observation of rapid changes within living cells is now is possible with significantly increased time resolution.

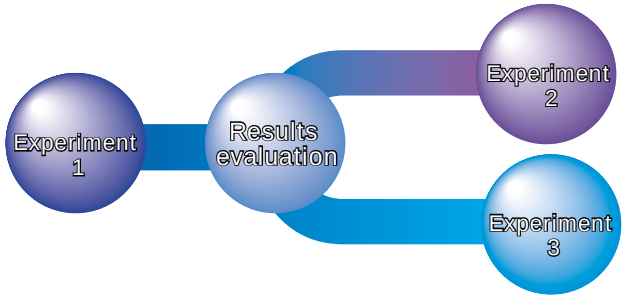
High-precision time-lapse imaging.
The FV1000 employs an external real time controller for high-precision time-lapse imaging of live cells. Data acquisition is controlled in microsecond increments, ensuring far more accurate and reliable timing.
In addition, laser light intensity is monitored and fed back to the AOTF, helping to stabilize the excitation light during time-lapse imaging.

Intuitive change of experiment conditions, even while scanning.
Pre-programmed image acquisition conditions (PMT sensitivity, laser intensity, time interval, channel settings, etc) can be changed while observation is in progress, making it possible to respond quickly to various cellular changes. On-line ROI plot data and real time ratio images can also be displayed.



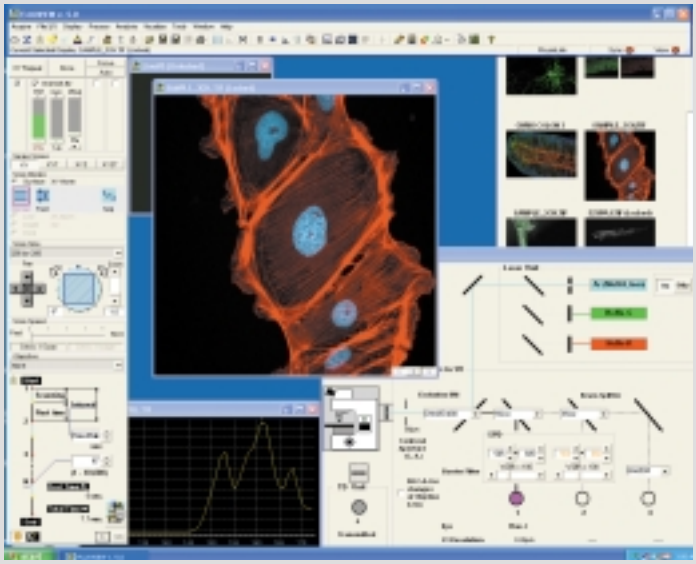
Independent designation of stimulation and scanning.
Any region of interest can be specified for stimulation and scanning independently, with unrestricted control of variations in timing, duration and intensity.

Pre-programming of experiment flow.
Experiments systematically programmed with a time series controller can be separated and freely combined, enabling the next set of experimental conditions to be selected after analysis of the prior experiment.



FV300/FV500/FV1000 SOFTWARE

Newly-developed FV300/FV500 Application Software may be used on FV1000 confocal microscope systems.
Improving on the FV300/500 software platform, the Basic Software adopts the menu bar method as a new function to enhance ease of use. Functional panel and image windows are separately displayed to avoid image acquisition functions being hidden below the image display. Any image can be displayed freely, with each image shown in an independent window. The Basic Software is suitable for both the filter and spectral version of the FV1000, and includes support for a dual monitor display as well as the single monitor display used by the FV300/500 systems. Full functionality is available for the filter-based FV1000 detection system. However, advanced features such as the SIM Scanner, spectral λ-scanning and spectral un-mixing are not supported by the Basic software.

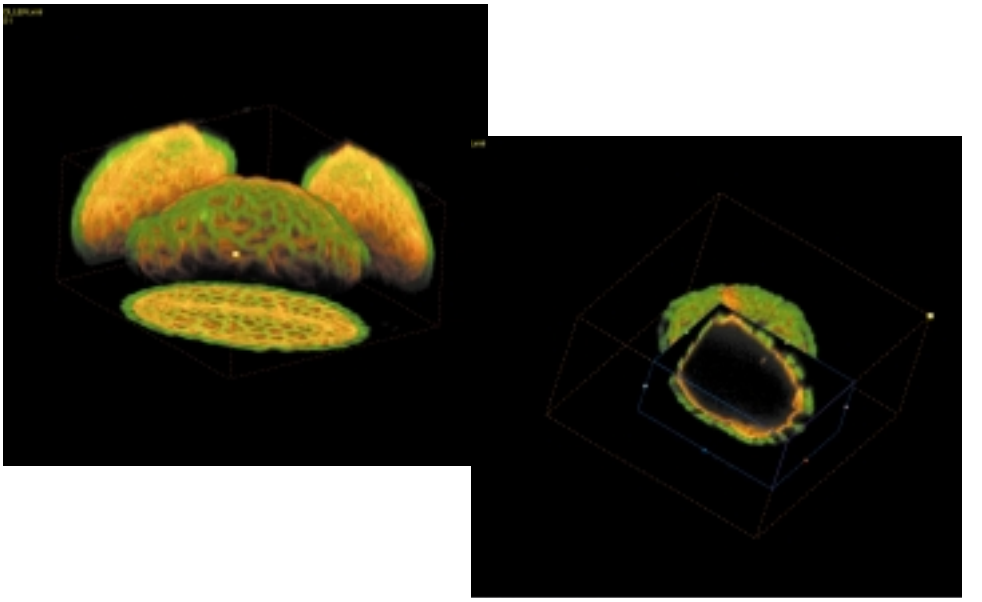


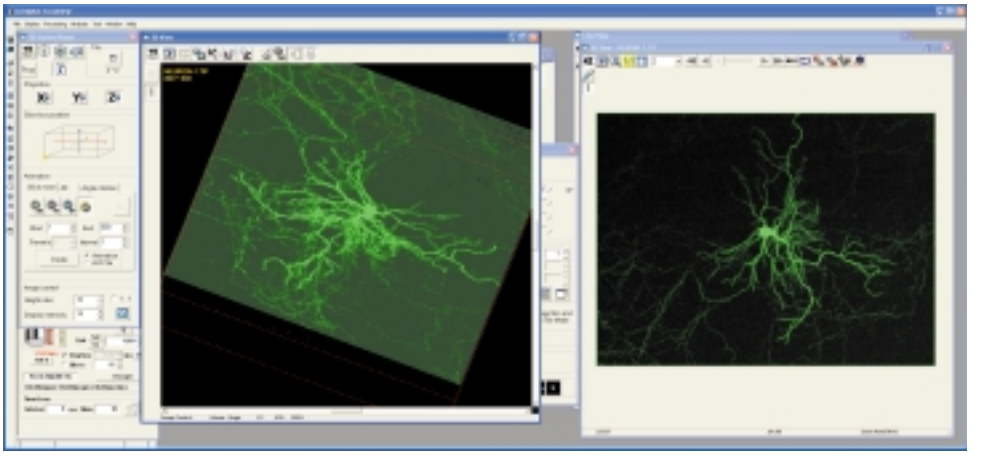
Single monitor image

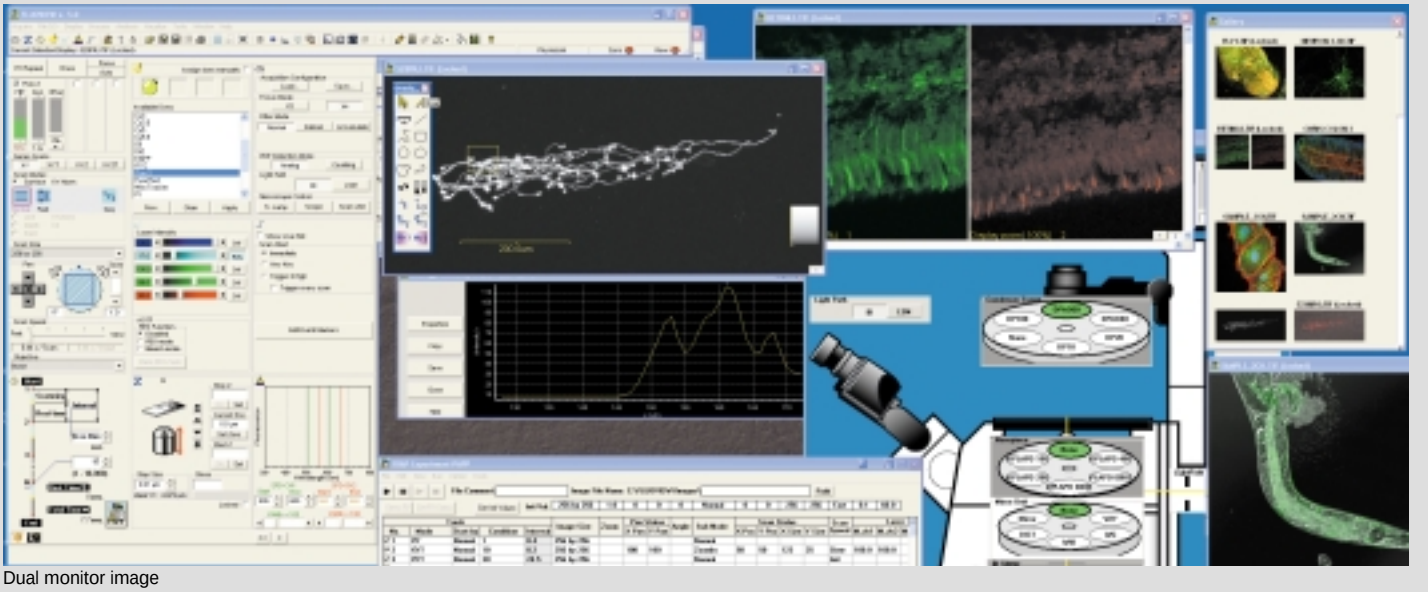
Intuitive on-line observation of desired region as a 2D or 3D image

3D-2D coupled display.
During 4-dimensional XYZT data acquisition or 3D rendering, a separate 2D image of any designated cross section can be obtained. This enables the scanning and image processing conditions to be changed in real time, i.e. while scanning. Any region can be freely observed by changing from 2D to 3D and from 3D to 2D dynamically.

Interactive volume rendering 3D.
Images may be rotated in any direction to facilitate observation.



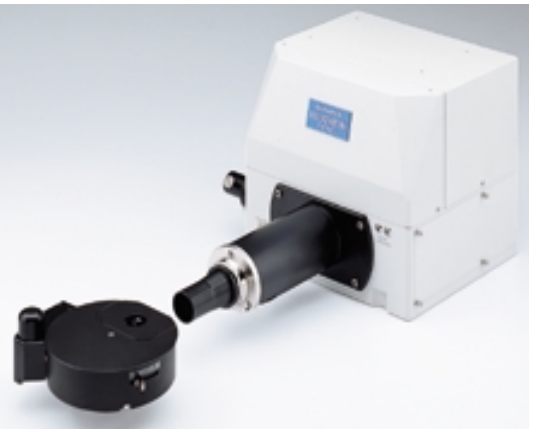




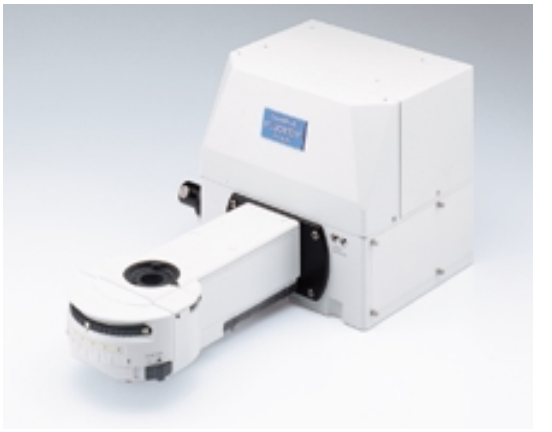
Dual monitor image

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 as well as a laser power monitoring and feedback control system are implemented.



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

A variety of laser systems are available. Ultra-fast and flexible laser intensity modulation is standard with FV1000. Use of a laser intensity monitoring and feedback control system in the scan unit ensures stable excitation intensity during long time-lapse observations.

Laser combiner (with mounted laser heads)
3 visible light lasers (multi–line Ar laser, HeNe-G and HeNe-R) can be mounted on the laser combiner with implemented AOTF module and driver.

LD405/LD440 laser
The Olympus-made 405nm and 440nm laser diodes feature a unique modulation function. High-speed shutter and laser intensity modulation are synchronized to the visible light AOTF system.

Illumination units

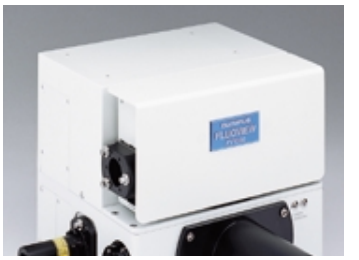
Conventional illumination modules are designed for long-duration time-lapse experiments. Since light is introduced through fiber delivery systems, no heat is transferred to the microscope.

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

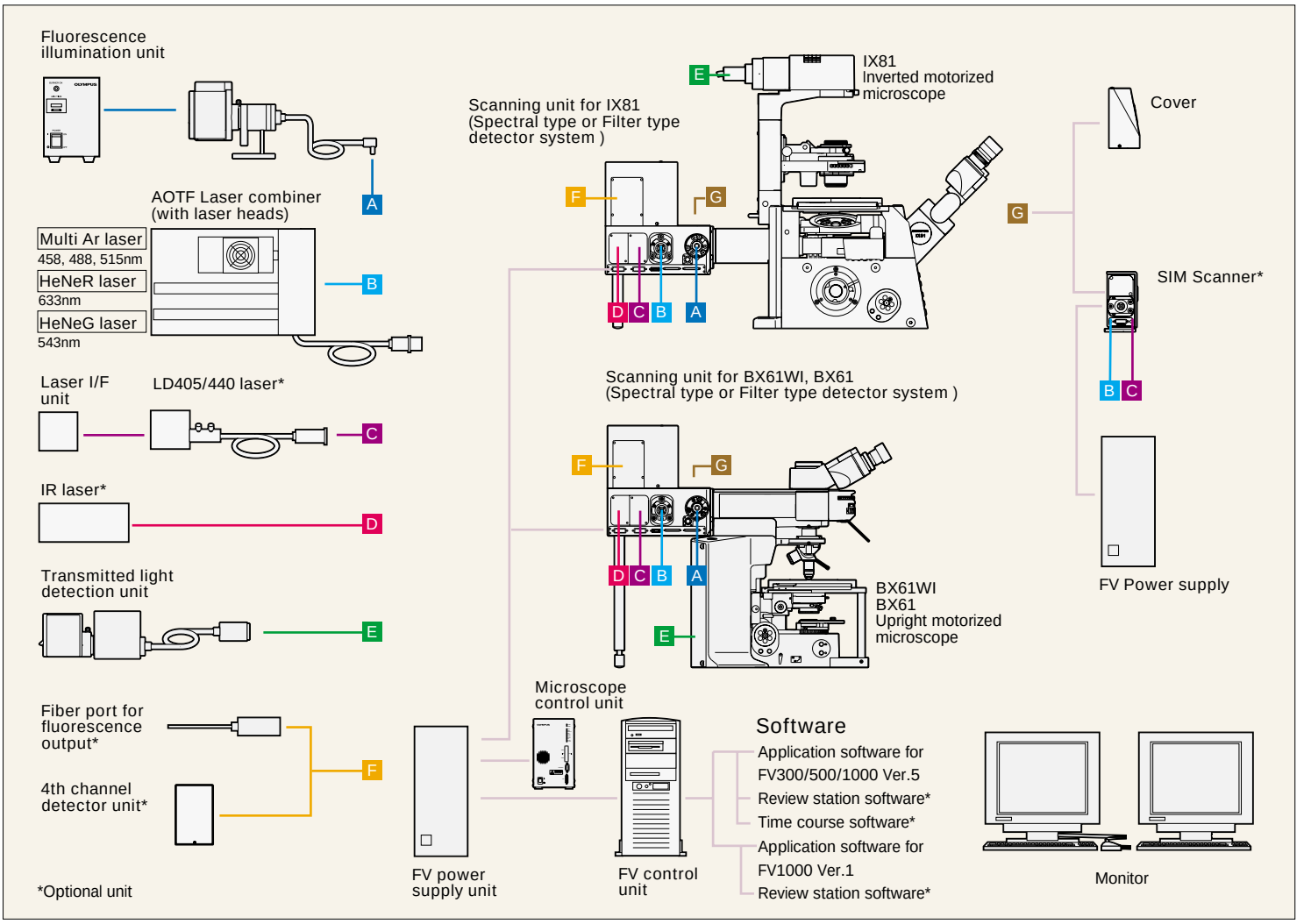
4th channel detector unit
Attaches to the optional port of either the filter or spectral type scanning unit and is used as a 4th confocal fluorescence detection channel. This is a filter-based fluorescence detection unit.



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.

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).

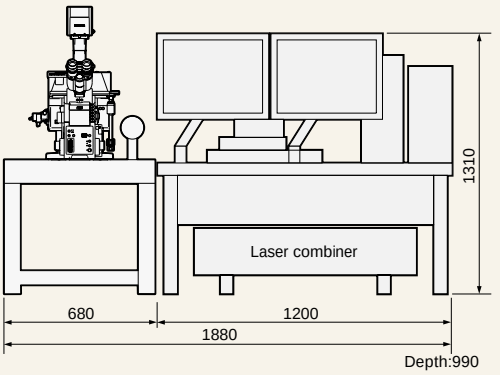
FV1000 SYSTEM DIAGRAM



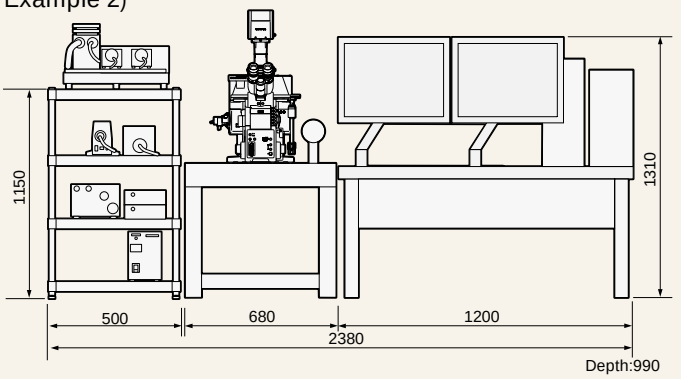
Recommended FV1000 system setup arrangements (IX81, BX61, BX61WI)

(unit: mm)

Example 1)



Example 2)



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	None
Fluorescence illumination unit	Main unit 180(W) x 320(D) x 235(H) Power supply 150(W) x 295(D) x 200(H)	6.7 4.6	100V/200V 260VA
Transmitted light detection unit	170(W) x 330(D) x 130(H)	5.9	Non (Microscope control unit)
Microscope control unit	125(W) x 330(D) x 215(H)	5.8	100-120/220-240V 350VA
FV Power Supply unit	180(W) x 328(D) x 424(H)	7.5	100-120/220-240V 400VA
FV Control unit (Computer)	180(W) x 420(D) x 360(H)	11.2	100-120/220-240V 300VA
Monitor	415(W) x 216(D) x 435(H)	6.6	100-120/220-240V 100VA
Laser combiner (with Laser heads)	483(W) x 800(D) x 279(H)	45	None
Multi Ar laser Power Supply	162(W) x 330(D) x 98(H)	3.2	100-120/220-240V 1500VA
HeNe G laser Power Supply	160(W) x 270(D) x 54(H)	1.4	100-120/220-240V 30VA
HeNe R laser Power Supply	160(W) x 270(D) x 54(H)	1.4	100-120/220-240V 30VA
LD405/440 laser	Laser head 70(W) x 70(D) x 70(H) Power Supply 95(H) x 210(W) x 297(D)	0.5 2.7	100-120/220-240V 100VA

FV1000 Specifications

		Spectral type fluorescence detector	Filter type fluorescence detector
Laser light	Visible light laser	Multi-line Ar laser (457nm, 488nm, 515nm, Total 30m) HeNe(G) laser (543nm, 1mW), HeNe (R) laser(633nm, 10mW)	
	Violet laser (option)	LD laser 405(405nm:25mW) , LD laser440 (440nm:5.3mW).	
	UV laser (option)	UV Argon laser (351nm, 50mW)	
	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	
Scanning and Detection	Scanner module	Standard 3 laser ports, VIS - UV - IR Excitation dichromatic mirror turret, 6 position (Ion deposition DMs and 20/80 half mirror) Dual galvano 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 (Ion deposition filters.)
	Filters	Ion deposition sputtered filters, dichromatic mirrors and barrier filters	
	Scanning method	2 galvano scanning mirror	
	Scanning modes	pixel size:64 x 64 — 4096 x 4096 (128x128 - 2056x2056) Pixel Dwell time: 2 to 200 microsec with unidirectional 0.5 or 1 microsec with fast bidirectional scanning X,Y,T,Z,λ (any combination) 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 0.5x increment (1x — 10x in 0.1increment)	
	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 focussing 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: 1GB or larger, CPU:Pentium 2GHz or higher, Hard disk: 80GB or larger, Media: CD-R/RW FV1000 Special I/F board (built-in PC), Graphic board: ATI RADEON 9200,	
	Power Supply Unit	Galvo control boards, scanning mirrors and gratings Real time controller	Galvo control boards, scanning mirrors
	Monitor	Monitor: SXGA 1280 x1024, dual 19-inch monitors (option: single monitor)	
Optional unit	SIM Scanner	2 Galvano scanning mirrors, pupil projection lens, built-in laser shutter, 1 laser port Fiber introduction of near UV laser diode or visible light laser, Optional: 2nd AOTF laser combiner	
	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 fibre connector (compatible fiber core 100 -125µm)	

Software	FV1000 Version 1 application software (FV10-ASW)	FV300/FV500/FV1000 Version 5 application software (FV10-SW)
Hardware	FV1000	FV300/FV500/FV1000
Display	single monitor or dual monitor display (dual monitor recommended)	
Optional Hardware	SIMS scanner unit, 4ch detector	4ch detector
Optional software	(time coarse acquisition and physiology analysis included)	FV-TCSW (TIEMPO), Multi-point software (XY stage control software for multi area time-lapse)
Image format	OIBOIF image format 8/16 bit gray scale/index color, 24/32/48 bit full color, JPEG/BMP/TIFF/AVI image functions	OLYMPUS Multi TIFF format 8/16 bit gray scale/index color, 24/32/48 bit full color, JPEG/BMP/TIFF/AVI image functions
Image acquisition	Region designation: point, line, free line, clip, clip zoom Real-time image calculation: Kalman filtering, peak detection calculation processing	
	2-dimension: XY, XT and Xλ dimension: XYZ, XYT, XYλ, XZT, XTλ and XZλ 4-dimension: XYZT, XZTλ and XYTλ	Flexible bandwidth selection with spectral detector
Image pixel format	64x64 - 4096x4096	128x128 - 2048x2048
Programmable Time course function	On-time control function,	Protocol processor
Image display	Each image display: Single-channel side-by-side, merge, trimming, live tiling, 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	3D animation, left/right stereo pairs,, red/green stereoscopic images and cross section 3D and 2D sequential operation function
Spectral Un-Mixing	2 Fluorescence spectral un-mixing 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, co-localization	
Others (options)	Off-line review station software	

Mouse Hippocampal Neurons (cover page data)
Expression of the Kaede protein, a green fluorescing protein that transferred upon 405nm exposure, identifies mouse hippocampal neuronal dentrites derived from a single cell body.
Kaede fluorescence in hippocampal neuron cells following 405nm exposure.
Left cell (red) 0.5 sec exposure, Middle cell (yellow) 0.25 sec exposure, Right cell (green) no exposure to the 405nm laser diode excitation.
Data courtesy of: Ms. Ryoko Ando, Dr. Atsushi Miyawaki
RIKEN Brain Science Institute Laboratory for Cell Function Dynamics

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