



# A1

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Confocal Microscope

# A1R/A1 – the ultimate confocal microscope



# Bring Imaging to Life!

Capturing high-quality images of cells and molecular events at high speed, Nikon's superior A1 confocal laser microscope series, with ground breaking technology, enables you to bring your imaging aspirations to life.

## **A1 with high performance and A1R with additional high-speed resonant scanner**

The new A1 series dramatically improves confocal performance and ease of operation. The A1R with a hybrid scanner supports advanced research methods using photo activation fluorescence protein. The new ergonomic user-friendly design facilitates live-cell work and a huge array of new imaging strategies.



## Dynamics

A high-speed resonant scanner allows imaging of intracellular dynamics at 30 frames per second (fps). Moreover, image acquisition of 420 fps is also possible.

## Interaction

Simultaneous imaging and photo activation with the proprietary hybrid scanner reveal intermolecular interaction. Analysis software\* for FRAP and FRET is provided as standard.

## Spectrum

Fast spectral image acquisition for 32 channels at a maximum of 24 fps (512 x 32 pixels) is possible. New real-time spectral unmixing and the V-filtering functions expand the range of use of spectral images.

## Image Quality

Fluorescence efficiency is increased by 30 percent, and S/N ratio of images is also increased. With diverse new technologies such as the VAAS pinhole unit, superior image quality has been achieved.

\*Will be available from January 2011

## A1R's hybrid scanner for ultrahigh-speed imaging and photo activation

A1R incorporates two independent galvano scanning systems: high-speed resonant and high-resolution non-resonant. This allows ultrafast imaging and photo activation imaging required to unveil cell dynamics and interaction.

### Ultrahigh-speed imaging

#### At 420 fps (512 x 32 pixels), the world's fastest image capture

A resonant scanner with ultrahigh resonance frequency of 7.8 kHz is simultaneously mounted with a non-resonant scanner that is capable of high-resolution (4096 x 4096 pixels) image capture.

1D scanning	15,600 lps
2D scanning	420 fps (512 x 32 pixels)
Full frame scanning	30 fps (512 x 512 pixels)



### Stable, high-speed imaging

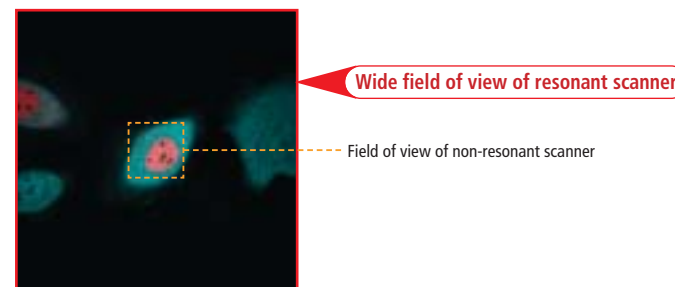
The Nikon original optical clock generation method is used for high-speed imaging with a resonant scanner. Stable clock pulses are generated optically, offering images that have neither flicker nor distortion even at the highest speed.

### High-speed data transfer with fiber-optic communication

The fiber-optic communication data transfer system can transfer data at a maximum of four giga bps—40 times faster than the conventional method. This allows transfer of image data (512 x 512) in five modes at more than 30 frames per second.

### High-speed imaging of wide field of view

When a non-resonant scanner is being used for high-speed image acquisition, the field of view of the scanned image is reduced to avoid overheating of the scanner (motor). Resonant scanners do not suffer overheating. Therefore, the field of view of the scanned area is approximately five times larger.

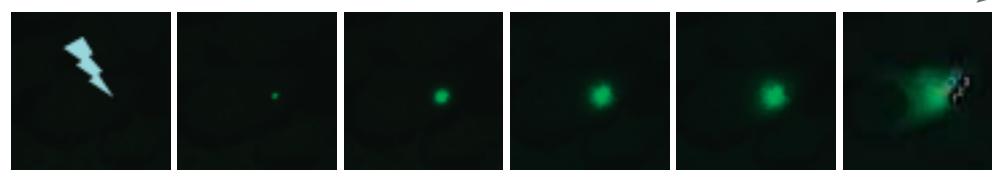


### High-speed photo activation imaging

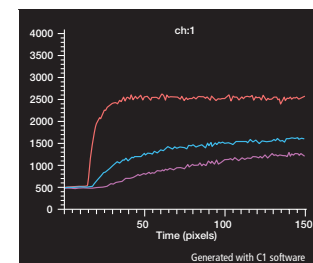
#### Simultaneous photo activation and imaging

Simultaneous photo activation and fluorescence imaging is conducted using non-resonant and resonant scanners. Because the resonant scanner can capture images at 30 fps, image acquisition of high-speed biological processes after photo activation is possible.

#### High-speed imaging of photo activation



Imaged at video rate (30 fps) while photo activating the target area with a 405 nm laser



Points within the cell and changes of fluorescence intensity (From the point closer to the activated point: red, blue, violet)

### Optical path in the A1R

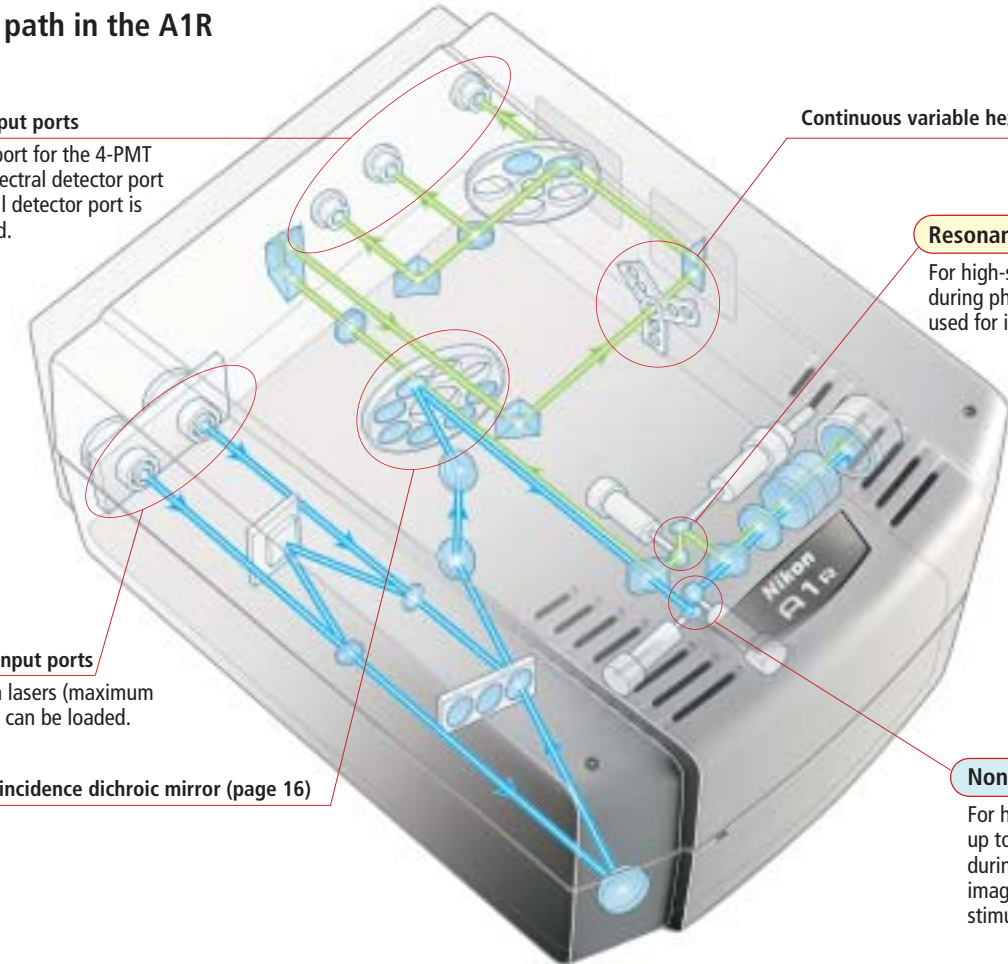
#### Optical output ports

A detector port for the 4-PMT detector, spectral detector port and optional detector port is incorporated.

#### Excitation input ports

Up to seven lasers (maximum nine colors) can be loaded.

#### Low-angle incidence dichroic mirror (page 16)



#### Continuous variable hexagonal pinhole (page 16)

#### Resonant scanner

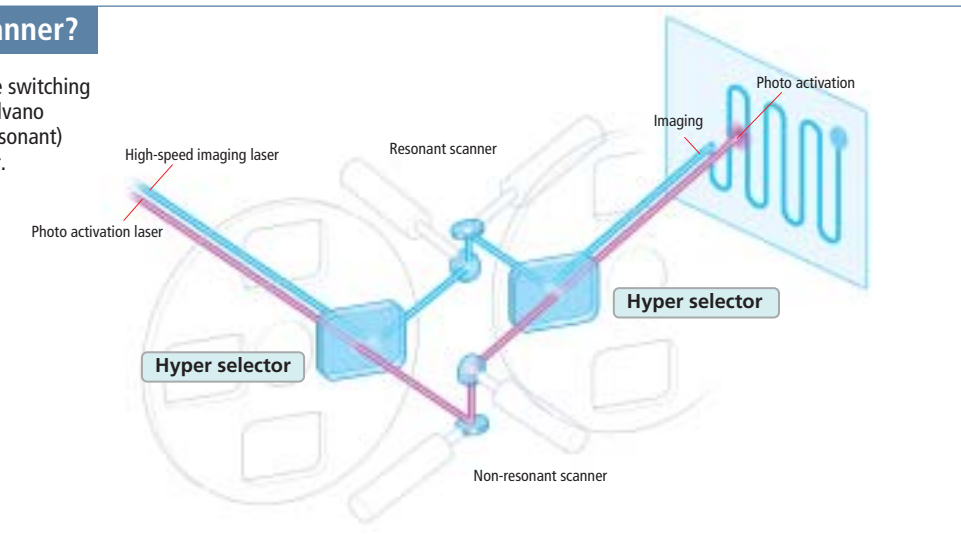
For high-speed imaging of 420 fps, during photo activation imaging, it is used for image capture.

#### Non-resonant scanner

For high-resolution imaging up to 4096 x 4096 pixels, during photo activation imaging, it is used for stimulation.

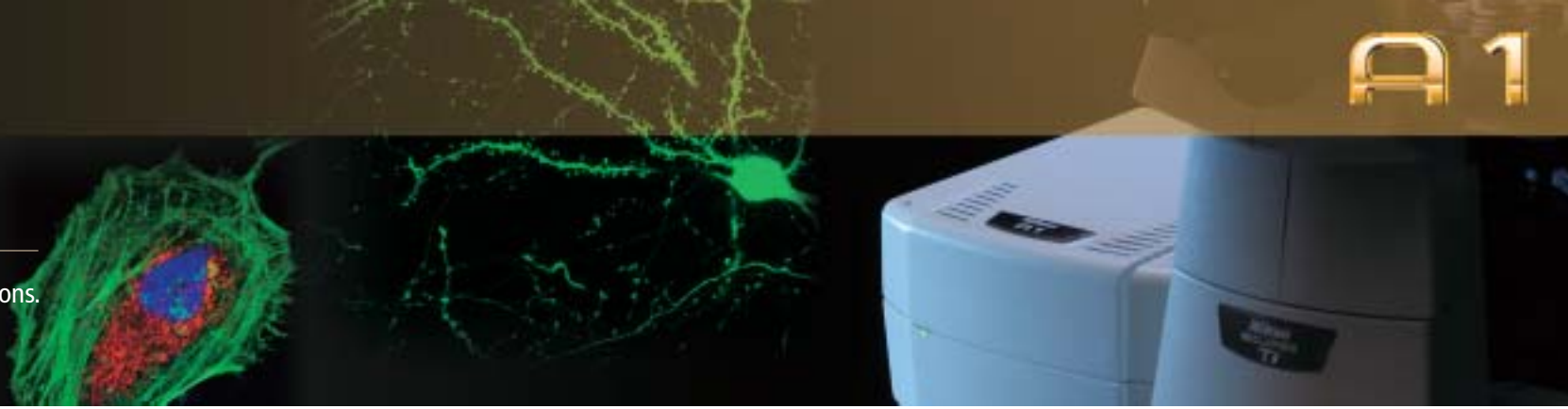
### What is a hybrid scanner?

This mechanism allows flexible switching or simultaneous use of two galvano scanners (resonant and non-resonant) with high-speed hyper selector.



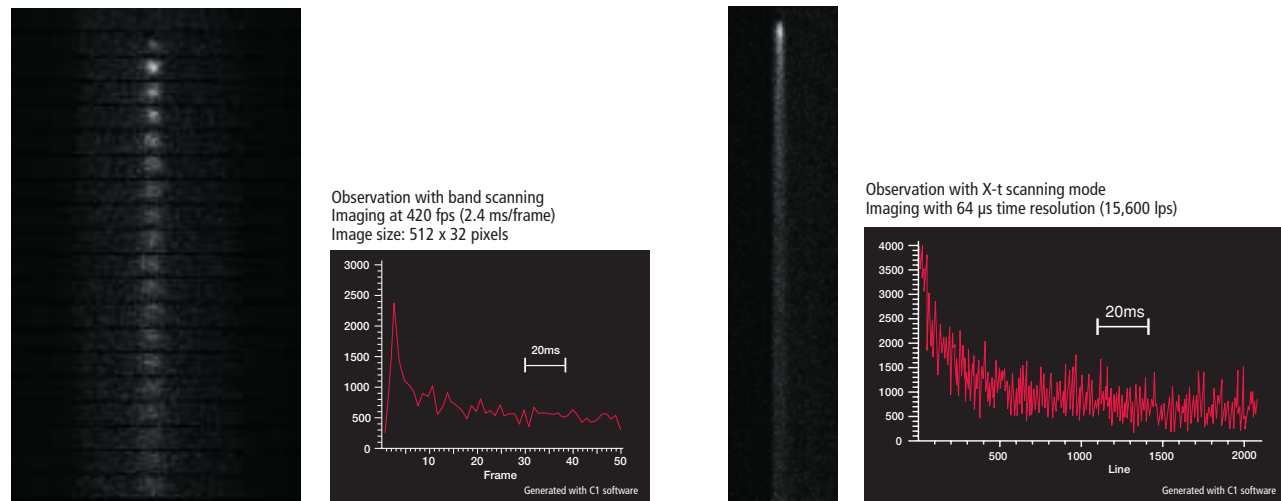
## Applications with a hybrid scanner

The A1R's high-speed imaging and hybrid scanners allow advanced imaging of cell dynamics and molecular interactions.



### Ultrahigh-speed imaging and photo activation

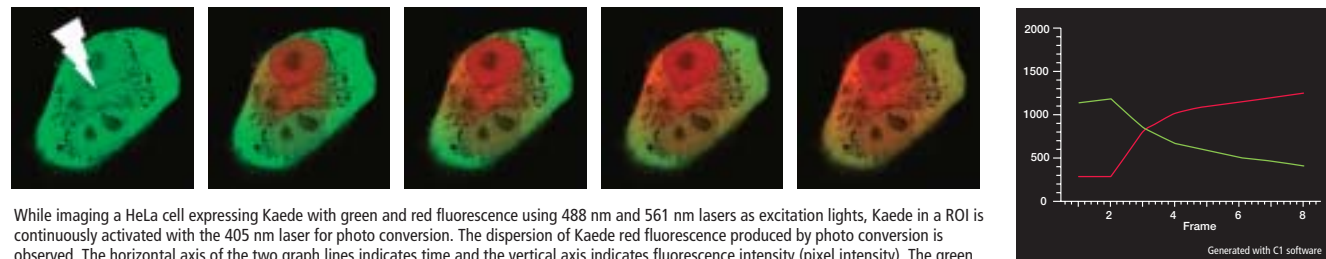
High-speed imaging (high time resolution imaging) from a video rate of 30 fps (33 ms time resolution) to 420 fps (2.4 ms time resolution) is possible. In addition, X-t scanning mode enables ultrahigh-speed imaging of dynamics with 64  $\mu$ s time resolution. Simultaneous photo activation during such high-speed imaging is also possible.



HeLa cells expressing PA-GFP are excited with 488 nm laser light. Directly after photo-activation (using 405 nm laser light) of a region of interest, the green emission (shown in grayscale) generated by photo-activated PA-GFP is detected and the subsequent distribution of the photo-activated protein is recorded at high speed. Please note that photo-activation (with the 405 nm laser) and image acquisition (with the 488 nm laser) is performed simultaneously. Both XYt and Xt recordings are displayed. Graphs show fluorescence intensity (vertical) versus time (horizontal).  
Activation laser wavelength: 405 nm, Imaging laser wavelength: 488 nm  
Photos courtesy of: Dr. Tomoki Matsuda and Prof. Takeharu Nagai, Research Institute for Electronic Science, Hokkaido University

### Kaede photo conversion fluorescence protein

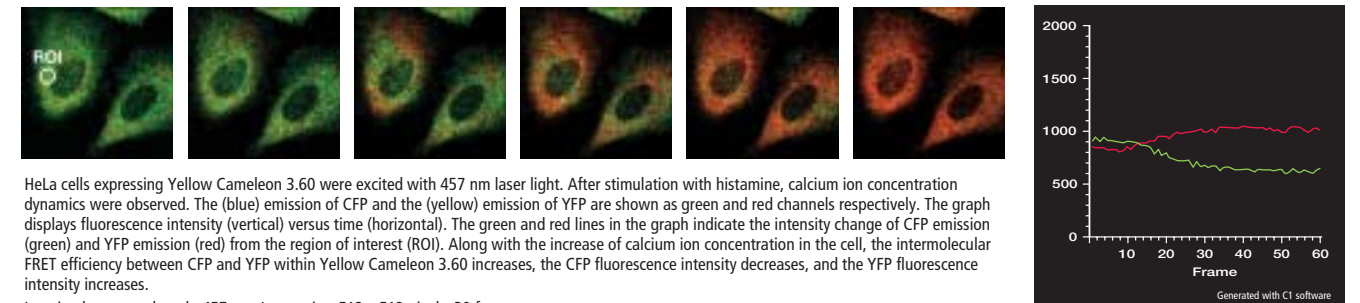
Kaede changes fluorescence colors irreversibly from green to red due to fluorescence spectral conversion when it is exposed to light with a spectrum from ultraviolet to violet.



While imaging a HeLa cell expressing Kaede with green and red fluorescence using 488 nm and 561 nm lasers as excitation lights, Kaede in a ROI is continuously activated with the 405 nm laser for photo conversion. The dispersion of Kaede red fluorescence produced by photo conversion is observed. The horizontal axis of the two graph lines indicates time and the vertical axis indicates fluorescence intensity (pixel intensity). The green line and red line in the graph respectively indicate intensity change of Kaede green and red fluorescence in the ROI.  
Activation laser wavelength: 405 nm, Imaging laser wavelength: 488 nm/561 nm, Image size: 512 x 512 pixels, 1 fps  
Photos courtesy of: Dr. Tomoki Matsuda and Prof. Takeharu Nagai, Research Institute for Electronic Science, Hokkaido University

### FRET (Förster Resonance Energy Transfer)

FRET is a physical phenomenon that occurs when there are at least two fluorescent molecules within a range of approximately 10 nm. When the emission spectrum of a fluorescent molecule overlaps with the absorption spectrum of another fluorescent molecule and the electric dipole directions of the two molecules correspond, then radiationless energy transfer from a donor molecule to an acceptor molecule may occur.

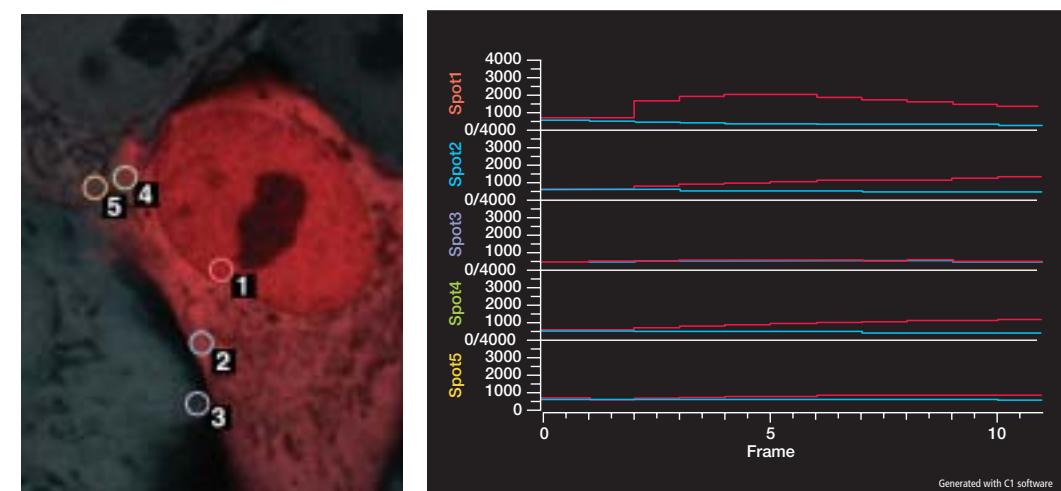


HeLa cells expressing Yellow Cameleon 3.60 were excited with 457 nm laser light. After stimulation with histamine, calcium ion concentration dynamics were observed. The (blue) emission of CFP and the (yellow) emission of YFP are shown as green and red channels respectively. The graph displays fluorescence intensity (vertical) versus time (horizontal). The green and red lines in the graph indicate the intensity change of CFP emission (green) and YFP emission (red) from the region of interest (ROI). Along with the increase of calcium ion concentration in the cell, the intermolecular FRET efficiency between CFP and YFP within Yellow Cameleon 3.60 increases, the CFP fluorescence intensity decreases, and the YFP fluorescence intensity increases.

Imaging laser wavelength: 457 nm, Image size: 512 x 512 pixels, 30 fps  
Photos courtesy of: Dr. Kenta Saito and Prof. Takeharu Nagai, Research Institute for Electronic Science, Hokkaido University

### Photo conversion protein: Phamret (Photo Activation-mediated Resonance Energy Transfer)

Photo conversion protein Phamret is a fusion protein of the CFP variant and the PA-GFP variant. When the PA-GFP variant is activated with violet to ultraviolet light, it changes light blue fluorescence to green fluorescence due to intermolecular FRET from CFP to PA-GFP.



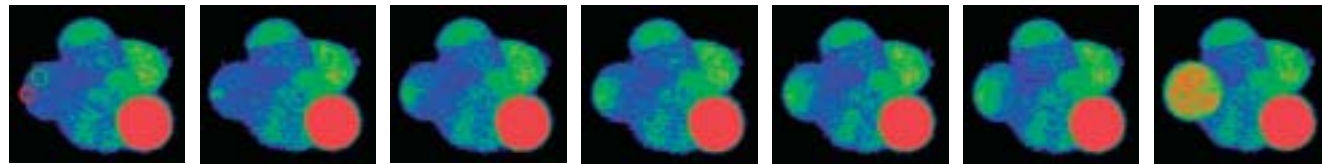
While imaging a HeLa cell expressing Phamret with light blue and green fluorescence using 457 nm laser as excitation light, the PA-GFP variant in an ROI is continuously activated with the 405 nm laser. The activated part observed in light blue fluorescence (shown in monochrome in the images) emits green fluorescence (shown in red in the images). And the dispersion of Phamret indicated by this green (shown in red in the images) is observed.

Activation laser wavelength: 405 nm, Imaging laser wavelength: 457 nm, Image size: 512 x 512 pixels, 1 fps  
Photos courtesy of: Dr. Tomoki Matsuda and Prof. Takeharu Nagai, Research Institute for Electronic Science, Hokkaido University

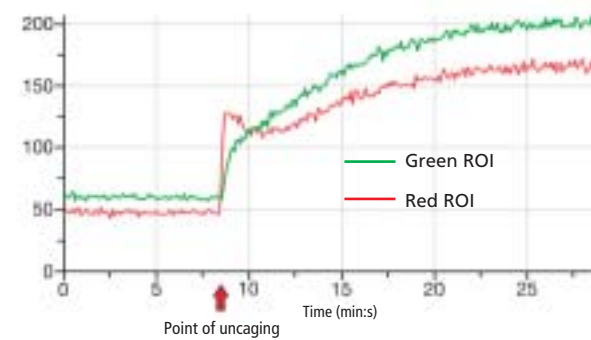
The graph indicates the changes of fluorescence intensity in each ROI. The blue line indicates the changes of fluorescence intensity of the CFP variant and the red line indicates the changes of fluorescence intensity of the PA-GFP variant.

### Caged compounds

Caged compounds are biologically active molecules that have been rendered functionally inert and can be instantly reactivated by near-ultraviolet light exposure. By controlling the light exposure, functionalized molecule expression in active form is possible in targeted intercellular sites with high spatial and time resolution.



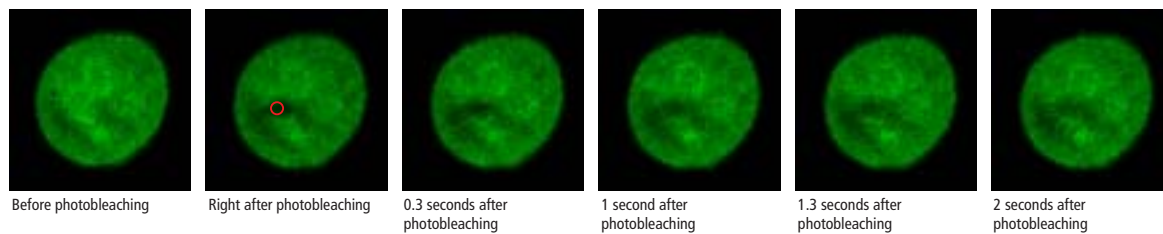
While imaging a human embryonic kidney (HEK) cell loaded with Caged Calcium and Fluo-4 using 488 nm laser at 120 fps, the red ROI is uncaged with the 408 nm laser. The graphs indicate intensity change of the red and green ROIs which were uncaged at the indicated point. Photos courtesy of: Dr. Chien-Yuan Pan, Dept. of Life Science, National Taiwan University



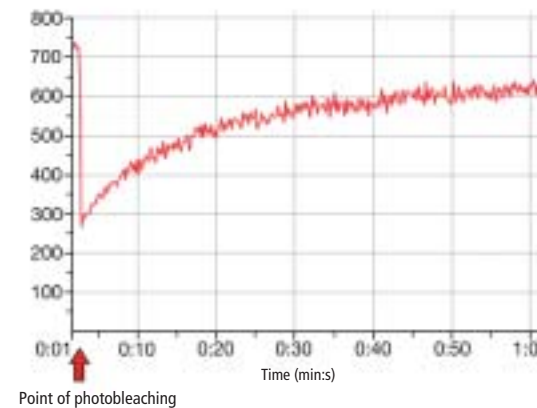
### FRAP (Fluorescence Recovery After Photo-bleaching)

After bleaching fluorescence dyes within the ROI by strong laser exposure, the recovery process of fluorescence over time is observed in order for the molecule diffusion rate to be analyzed.

A1R's hybrid scanner allows high-speed imaging of fluorescence recovery during bleaching at user-defined area.

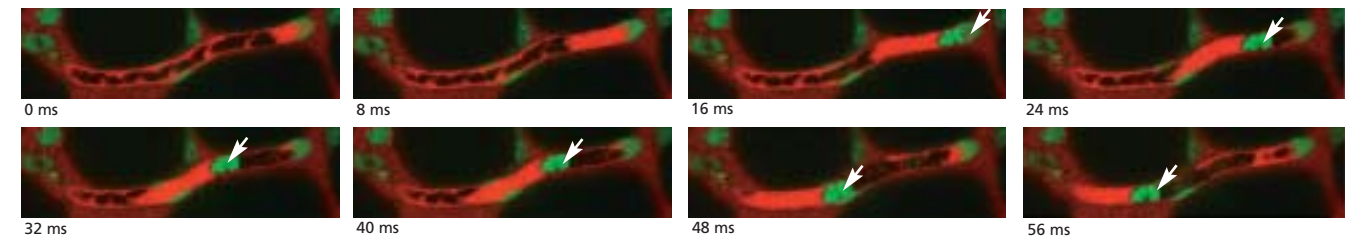


HeLa cell expressing CREB-GFP is bleached during imaging of FRAP observation. Red ROI is photobleached with a 408 nm laser during imaging with a 488 nm laser at 30 fps at room temperature. The graph indicates the intensity change of the red ROI, which is bleached at the indicated point. Photos courtesy of: Dr. Hiroshi Kimura, Associate Professor, Graduate School of Frontier Biosciences, Osaka University



### In vivo imaging

Imaging dynamic status of fluorescence labeled agents and intravital substances in live organisms under good physiological conditions is possible.



Mouse blood vessel administered Tetramethyl Rhodamine and Acridine Orange and observed at 120 fps (8 ms/frame). Red: blood vessel, Green: nucleus. Tile images displayed every 8 ms. The arrows indicate white blood cell flow in the vessel. Photos courtesy of: Dr. Satoshi Nishimura, Department of Cardiovascular Medicine, the University of Tokyo, Nano-Bioengineering Education Program, the University of Tokyo, PRESTO, Japan Science and Technology Agency

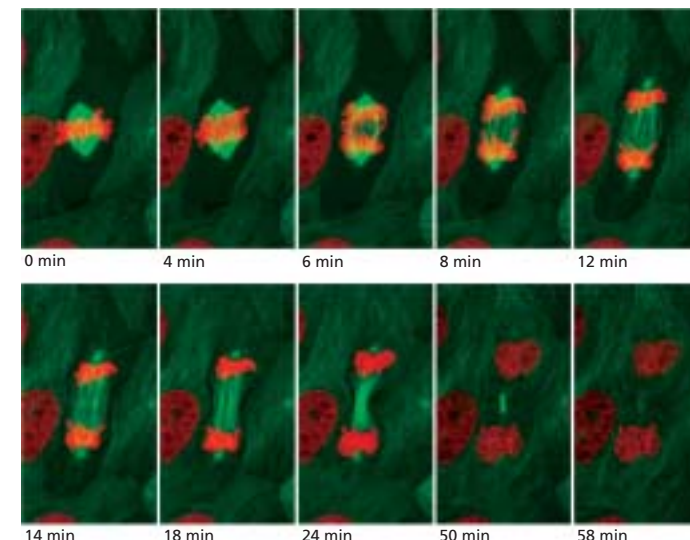


Zebra fish expressing DsRed in red blood cells. The red blood cell flow, indicated by the arrows, is simultaneously observed with DIC and confocal images at 60 fps (16 ms/frame). Photos courtesy of: Dr. Yung-Jen Chuang, Assistant Professor, Institute of Bioinformatics and Structural Biology & Department of Life Science, National Tsing Hua University



### Multicolor imaging

Standard four-channel detector eliminates the necessity for an additional fluorescence detector after purchase and allows easy imaging of a specimen labeled with four probes.



Z series projection of XYZ images of LLC-PK1 cell expressing EGFP- $\alpha$ -tubulin (green) and Histone H2B-mCherry (red) captured every 2 min. Photos courtesy of: Dr. Keiju Kamijo, Department of Stem Cell Biology and Histology, Tohoku University Graduate School of Medicine

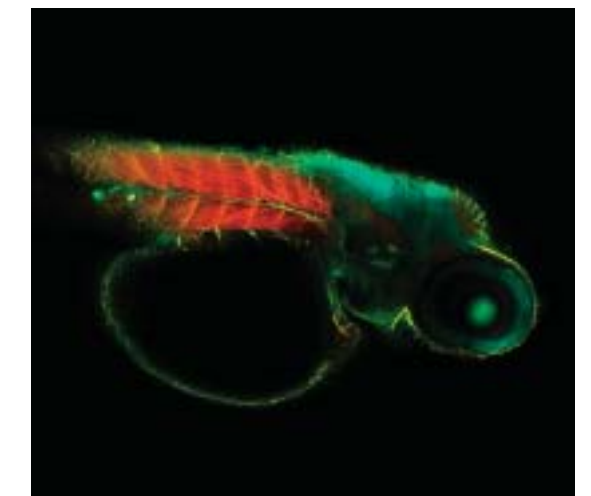


Image of a zebra fish labeled with four probes. Nucleus (blue): Hoechst33342, Pupil (green): GFP, Nerve (yellow): Alexa555, Muscle (red): Alexa647. Photographed with the cooperation of: Dr. Kazuki Horikawa and Prof. Takeharu Nagai, Research Institute for Electronic Science, Hokkaido University

# Applications

## Calcium sparks

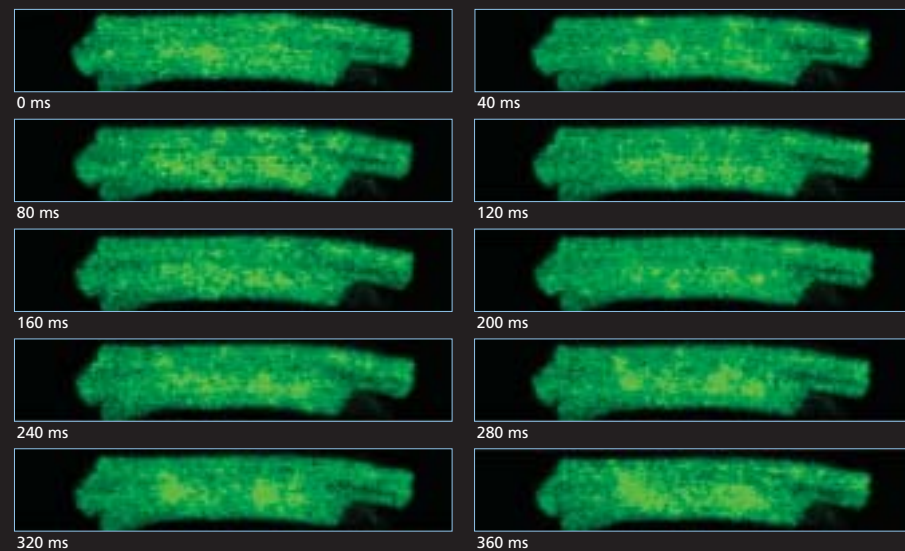
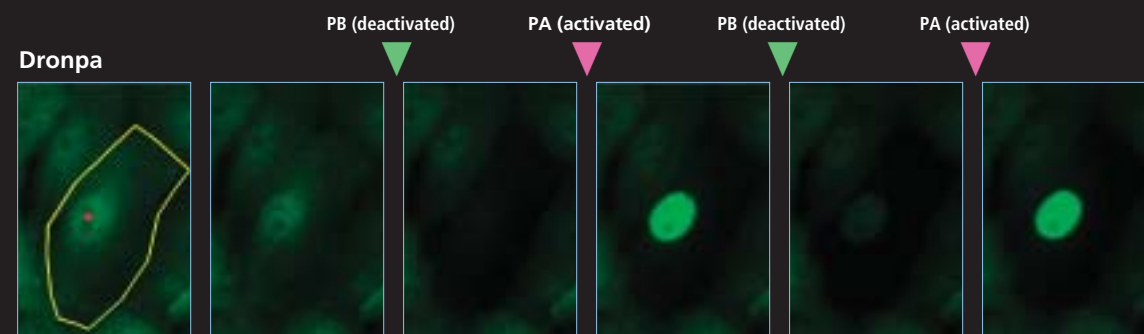


Image of calcium sparks in mouse's isolated cardiomyocyte loaded with calcium indicator captured at 230 fps (4 ms/frame)  
Tile image of 10 images displayed every 40 ms

Photos courtesy of: Dr. Heping Cheng, Institute of Molecular Medicine, Peking University



Dronpa-Green is photochromic fluorescence protein that loses fluorescence when exposed to blue-green light (488 nm), and fluoresces again when exposed to violet light (408 nm). At the point of PB (PhotoBleach), the yellow ROI—a whole LLC-PK1 cell, stable Dronpa-Green expression strain—was exposed to 488 nm intense light to deactivate its fluorescence, and at the point of PA (Photoactivation), the red ROI—a part of the nucleus—was exposed to 408 nm light to activate the fluorescence. Excitation by weak 488 nm light allows dynamic observation of molecules that are labeled with green fluorescence. PB and PA can be repeated.

Photos courtesy of: Dr. Keiju Kamijo, Department of Stem Cell Biology and Histology, Tohoku University Graduate School of Medicine

## PA-GFP



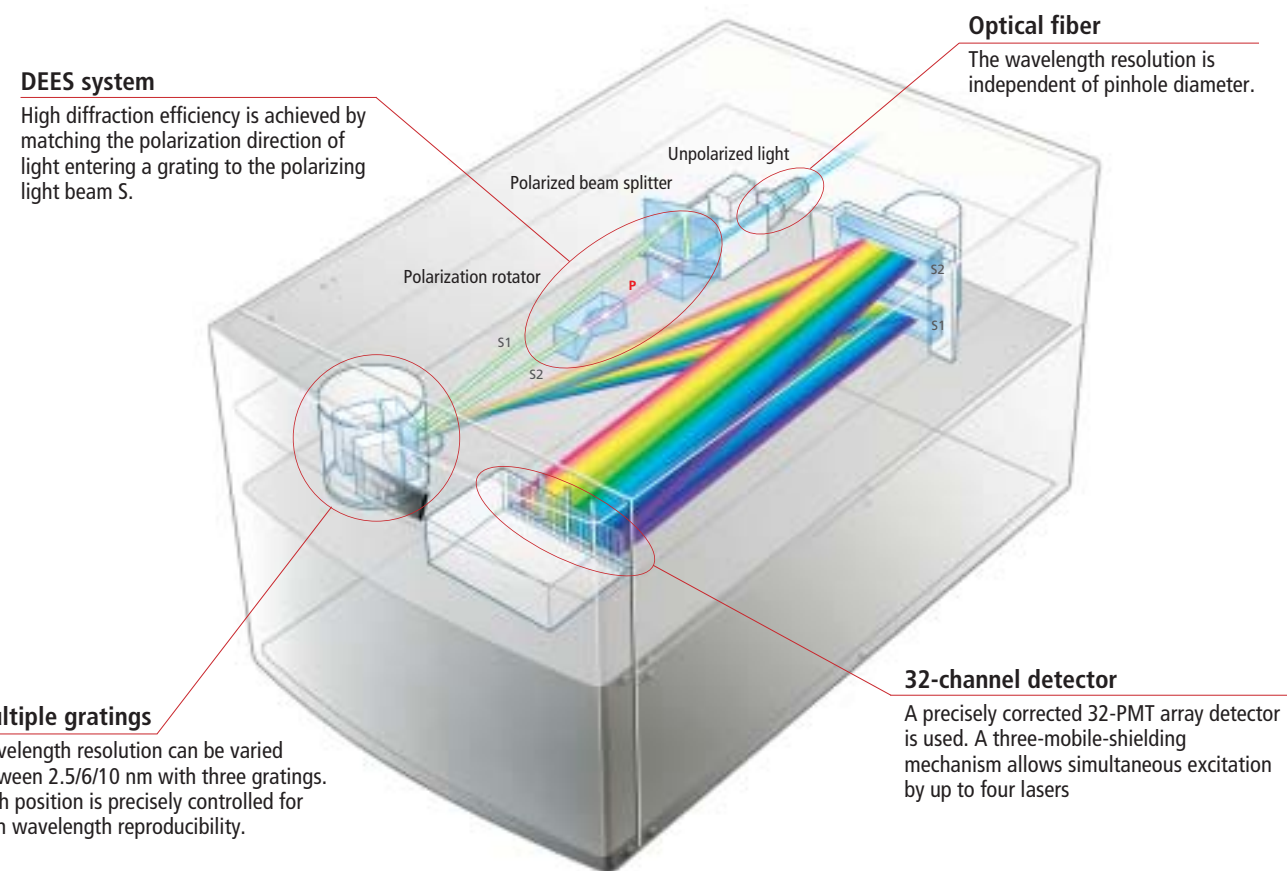
Red ROI in HeLa cell expressing PA-GFP was photoactivated for 1 second with a 405 nm laser while imaging in resonant mode at 30 fps with 488 nm laser. DIC images were captured simultaneously.

Photos courtesy of: Dr. Hiroshi Kimura, Associate Professor, Graduate School of Frontier Biosciences, Osaka University

# Spectrum

## Enhanced spectral detector

Nikon's original spectral performance is even further enhanced in the A1 series, allowing high-speed spectral acquisition with a single scan. In addition, new functions including a V-filtering function are incorporated.



### DEES system

High diffraction efficiency is achieved by matching the polarization direction of light entering a grating to the polarizing light beam S.

### Optical fiber

The wavelength resolution is independent of pinhole diameter.

### Multiple gratings

Wavelength resolution can be varied between 2.5/6/10 nm with three gratings. Each position is precisely controlled for high wavelength reproducibility.

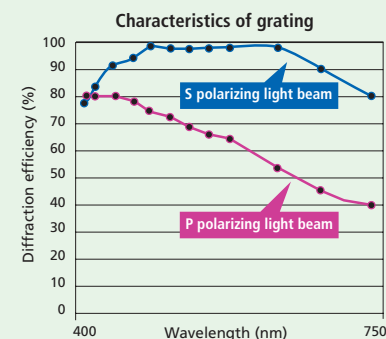
### 32-channel detector

A precisely corrected 32-PMT array detector is used. A three-mobile-shielding mechanism allows simultaneous excitation by up to four lasers

## High-quality spectral data acquisition

### Diffraction Efficiency Enhancement System (DEES)

With the DEES, unpolarized fluorescence light emitted by the specimen is separated into two polarizing light beams P and S by a polarizing beam splitter. Then, P is converted by a polarization rotator into S, which has higher diffraction efficiency than P, achieving vastly increased overall diffraction efficiency.



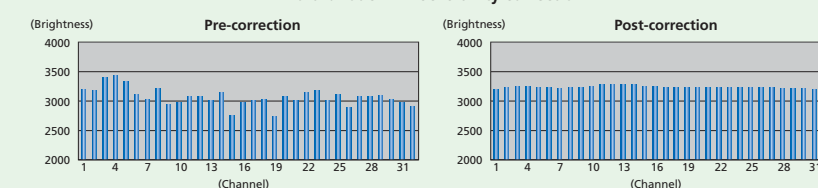
### High-efficiency fluorescence transmission technology

The ends of the fluorescence fibers and detector surfaces use a proprietary anti-reflective coating to reduce signal loss to a minimum, achieving high optical transmission.

### Accurate, reliable spectral data: three correction techniques

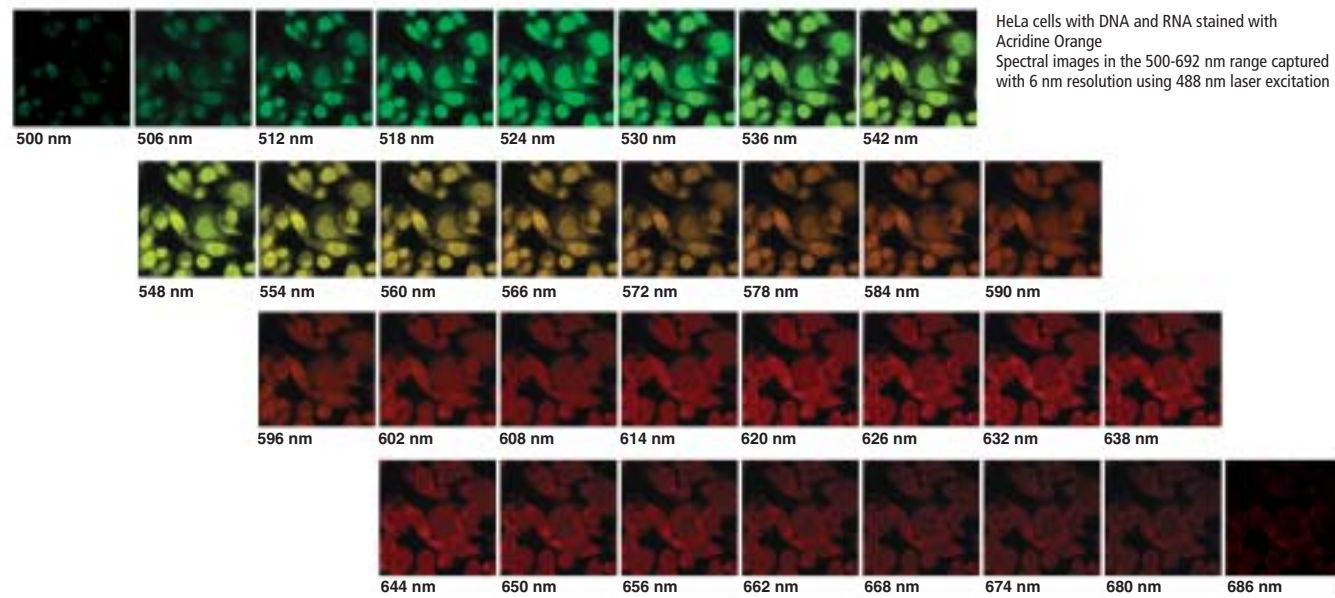
Three correction techniques allow for the acquisition of accurate spectra: interchannel sensitivity correction, which adjusts offset and sensitivity of each channel; spectral sensitivity correction, which adjusts diffraction grating spectral efficiency and detector spectral sensitivity; and correction of spectral transmission of optical devices in scanning heads and microscopes.

### Multi-anode PMT sensitivity correction



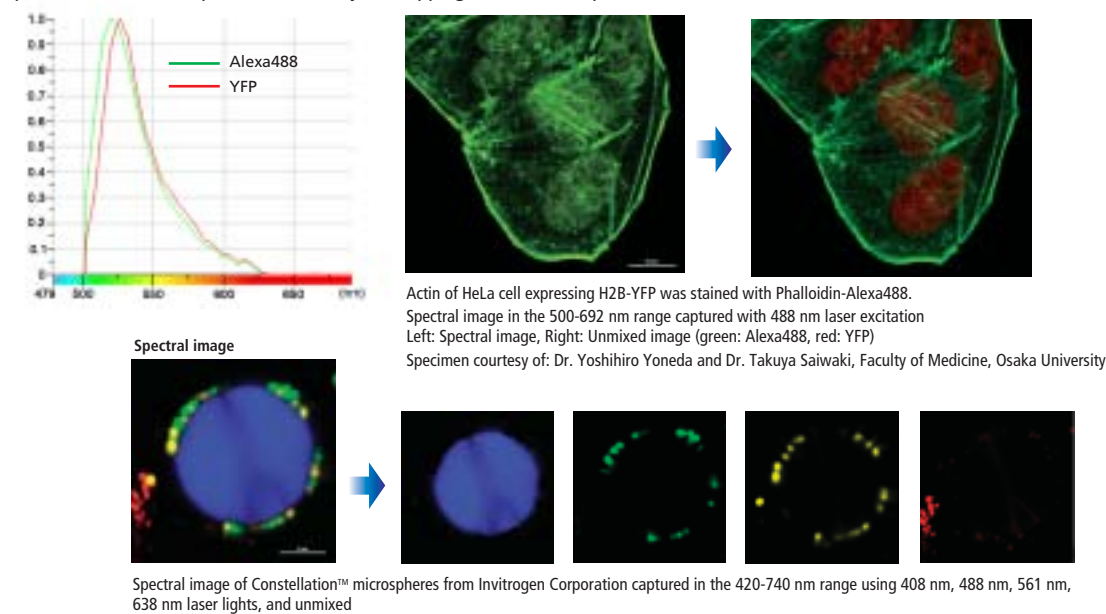
### Fast 32-channel imaging at 24 fps

New signal processing technology and high-speed AD conversion circuit allow acquisition of a 32-channel spectral image (512 x 512 pixels) in 0.6 second. Moreover, acquisition of 512 x 32 pixels images at 24 frames per second is achieved.



### Accurate spectral unmixing

Accurate spectral unmixing, which has a high reputation among users of the previous model, the C1si, provides maximum performance in the separation of closely overlapping fluorescence spectra and the elimination of autofluorescence.



### Real time unmixing

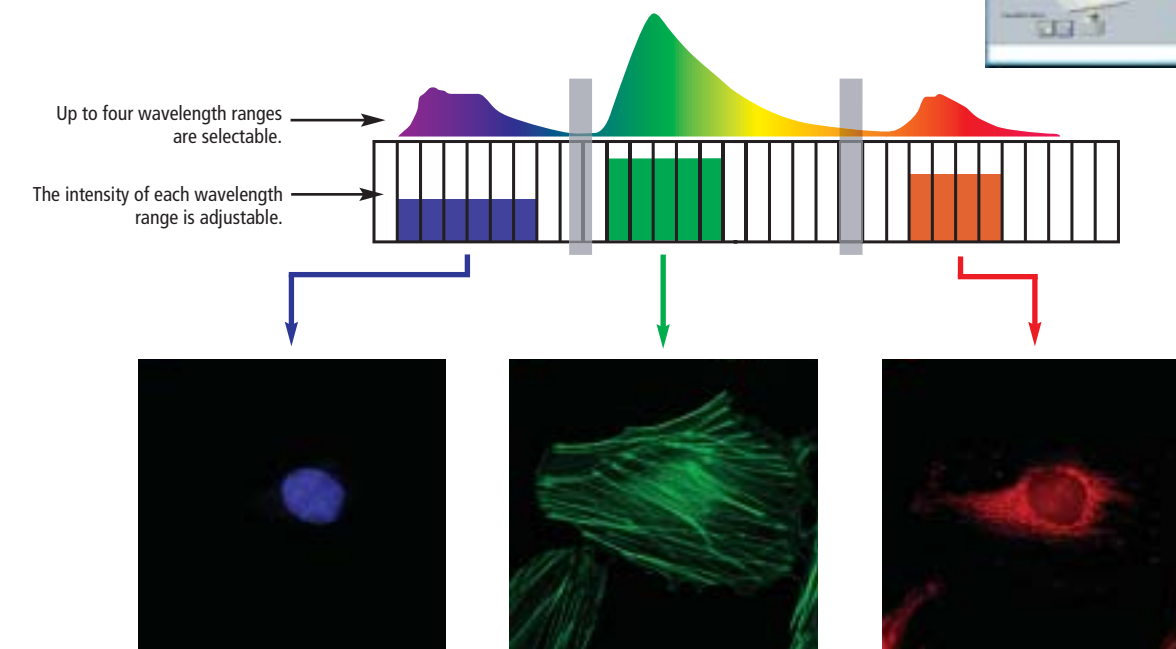
Newly developed algorithms and high-speed data processing enable real time unmixing during image acquisition, which used to be processed after spectral imaging. This is highly effective for FRET analysis, since probes with adjacent spectra such as CFP and YFP, GFP and YFP that were difficult to unmix in real time can be easily unmixed.

### Simultaneous excitation of four lasers

Three user-defined laser shields allow simultaneous use of four lasers selected from a maximum of nine colors, enabling broader band spectral imaging.

### Filter-less intensity adjustment is possible with V-filtering function

Desired spectral ranges that match the spectrum of the fluorescence probe in use can be selected from 32 channels and combined to perform the filtering function. By specifying the most appropriate wavelength range, image acquisition with the optimal intensity of each probe is possible in FRET and colocalization. Up to four wavelength ranges can be simultaneously selected. The sensitivity of each range can be individually adjusted, which supports applications using various probe combinations.

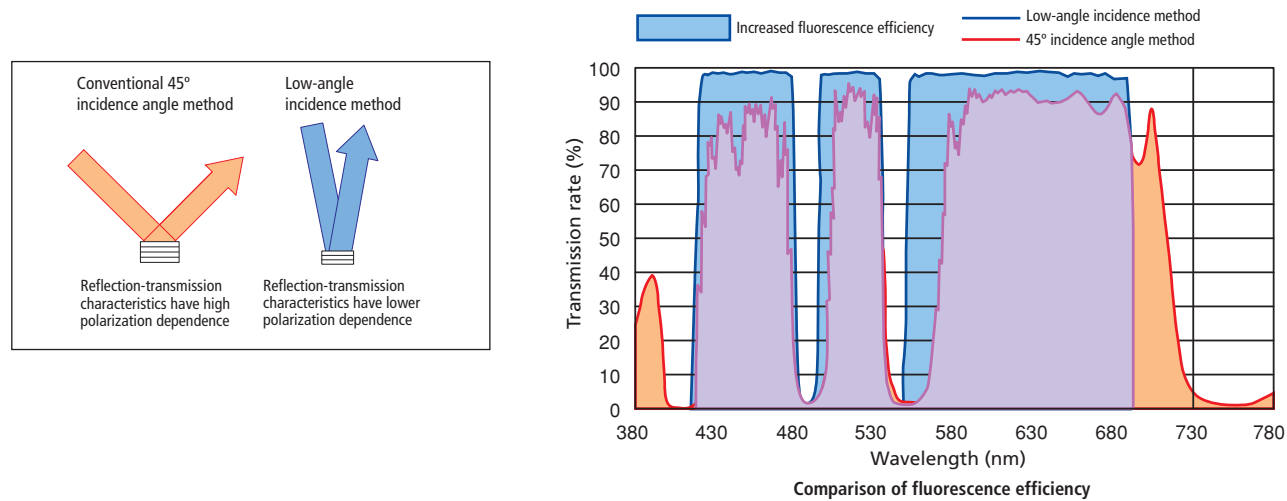


## Key Nikon innovations for improving image quality

The best image quality is achieved by an increased light sensitivity resulting from comprehensive technological innovations in electronics, optics and software.

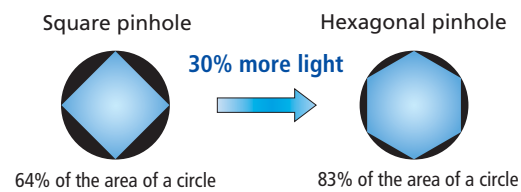
### Low-angle incidence dichroic mirror creates a 30% increase in fluorescence efficiency

With the A1 series, the industry's first low-angle incidence method is utilized on the dichroic mirrors and a 30% increase of fluorescence efficiency is realized.



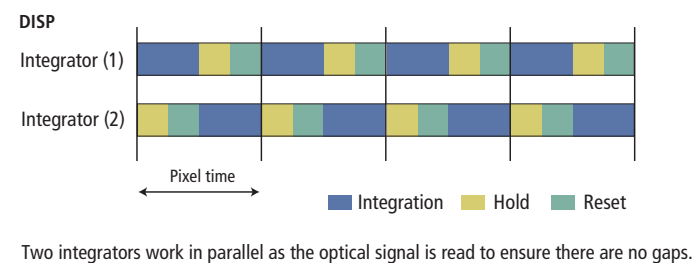
### Brighter images with continuous variable hexagonal pinhole

Instead of a continuous variable square pinhole, the industry's first hexagonal pinhole is employed. Higher brightness, equivalent to that of an ideal circular pinhole is achieved while maintaining the confocality.



### DISP improves electrical efficiency

Nikon original DISP (Dual Integration Signal Processing) technology has been implemented in the image processing circuitry to improve electrical efficiency, preventing signal loss while the digitizer processes pixel data and resets. The signal is monitored for the entire pixel time resulting in an extremely high S/N ratio.



### VAAS pinhole unit transcends the existing concept of a confocal microscope

It is widely recognized that reducing the pinhole size to eliminate flare light from the non-focal plane causes darker images. The VAAS (Virtual Adaptable Aperture System) provides a new confocal microscopy that can eliminate flare while retaining image brightness.

#### Effects

- 1 Acquisition of brighter images with less flare is possible.
- 2 Different sectionings (slice thickness) can be simulated via deconvolution after image acquisition.
- 3 Images of both focal plane and non-focal plane can be acquired with a single scan, boosting speed and reducing damage to live cells.

#### Principle and features

#### Conventional confocal microscope

Small pinholes reduce flare but darken images, while large pinholes brighten images but increase flare.

Light that passes through the pinhole is detected.

— In-focus signal

#### VAAS pinhole unit

By deconvolving the light that passes through the pinhole and the light that doesn't pass through the pinhole, flare can be eliminated while using a large pinhole.

Light that passes through the pinhole is detected.

— In-focus signal  
— Out-of-focus signal

#### Line profile

Intensities of both images are nearly equal.

VAAS images contain less background noise.

Mixture of 10- $\mu$ m and 0.1- $\mu$ m beads

Fluorescence of 10- $\mu$ m beads is detected with the light from the non-focal plane, which leaks through large pinholes in conventional confocal imaging. VAAS pinhole unit creates the image without light leaking from the non-focal plane.

Conventional confocal image (1 AU)

Acute brain slice from pThy1-EYFP transgenic mouse

Photos courtesy of: Dr. Yasushi Okada, Cell Biology, Medical Dept. of Graduate School, the University of Tokyo

Conventional image captured with 1.5 AU pinhole

Differential VAAS image

## Increased flexibility and ease of use

Control software NIS-Elements C features easy operation and diverse analysis functions. Combined with a remote controller and other hardware, it provides a comprehensive operational environment.



### NIS-Elements C



Detailed operability based on the analysis of every possible confocal microscope operation pattern ensures an intuitive interface and operation, satisfying both beginners and experienced confocal users. By taking advantage of the hybrid scanner, the software enables a complicated sequence of experiments such as photo activation to be carried out with simple-to-use settings.

#### Simple image acquisition

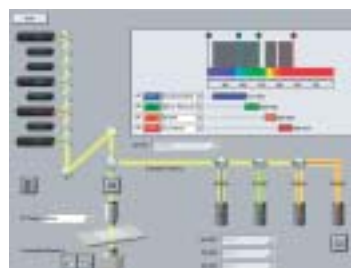
##### • Basic operation

Parameters for basic image acquisition are integrated in a single window, allowing simple image acquisition.



##### • Optical setting

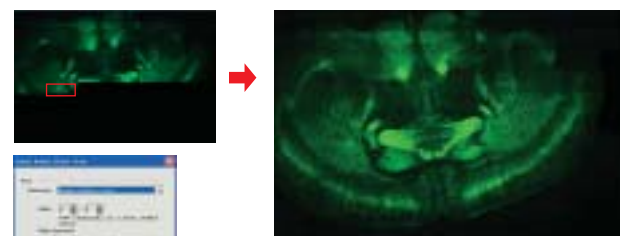
By simply selecting a fluorescence probe, an appropriate filter and laser wavelength are set automatically. Microscope setup is also conducted automatically.



#### Diverse application

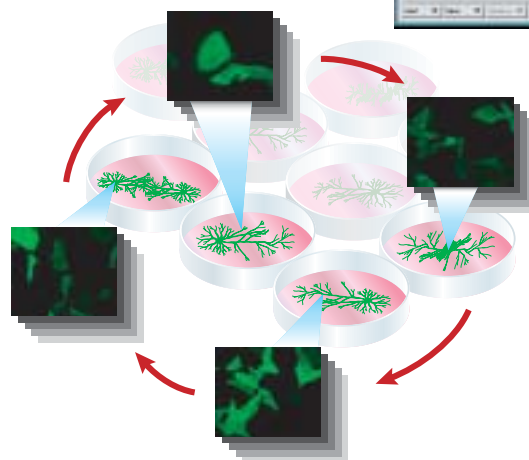
##### • Large imaging (image tiling)

Images of adjacent fields that are continuously captured with the motorized stage are automatically stitched to produce a whole high-resolution image of the tissue.



##### • Multidimensional image acquisition

Acquisition of images with a free combination of multidimensional parameters including X, Y, Z, t,  $\lambda$  (wavelength), and multipoint is possible.



##### • Parameter setting for photo activation

Timing and imaging parameters for photo activation are set intuitively.



#### Reliable analysis functions

- Real-time ratio display
- Deconvolution
- High-speed 3D rendering
- Multidimensional image display (nD Viewer)
- Synchronized display of multidimensional images (View synchronizer)
- Diverse measurement and statistical processing
- Powerful image database function
- Colocalization and FRET



#### User-friendly hardware

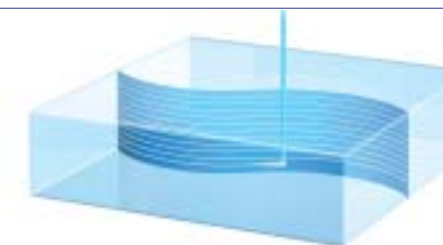
##### 4-channel detector unit with changeable filters

In combination with four lasers, simultaneous observation of four fluorescence labels is possible as standard. Each of three filter wheels can mount six filter cubes that are commonly used for a microscope. They are easily changeable by users, combining modularity, flexibility with user-friendliness.



##### Spline Z scans for real-time display of cross-sectional images\*

High-speed image acquisition in the Z direction as well as the XY direction is possible. By using the piezo motorized Z stage, an arbitrary vertical cross-sectional view can be achieved in real time without acquiring a 3D image.

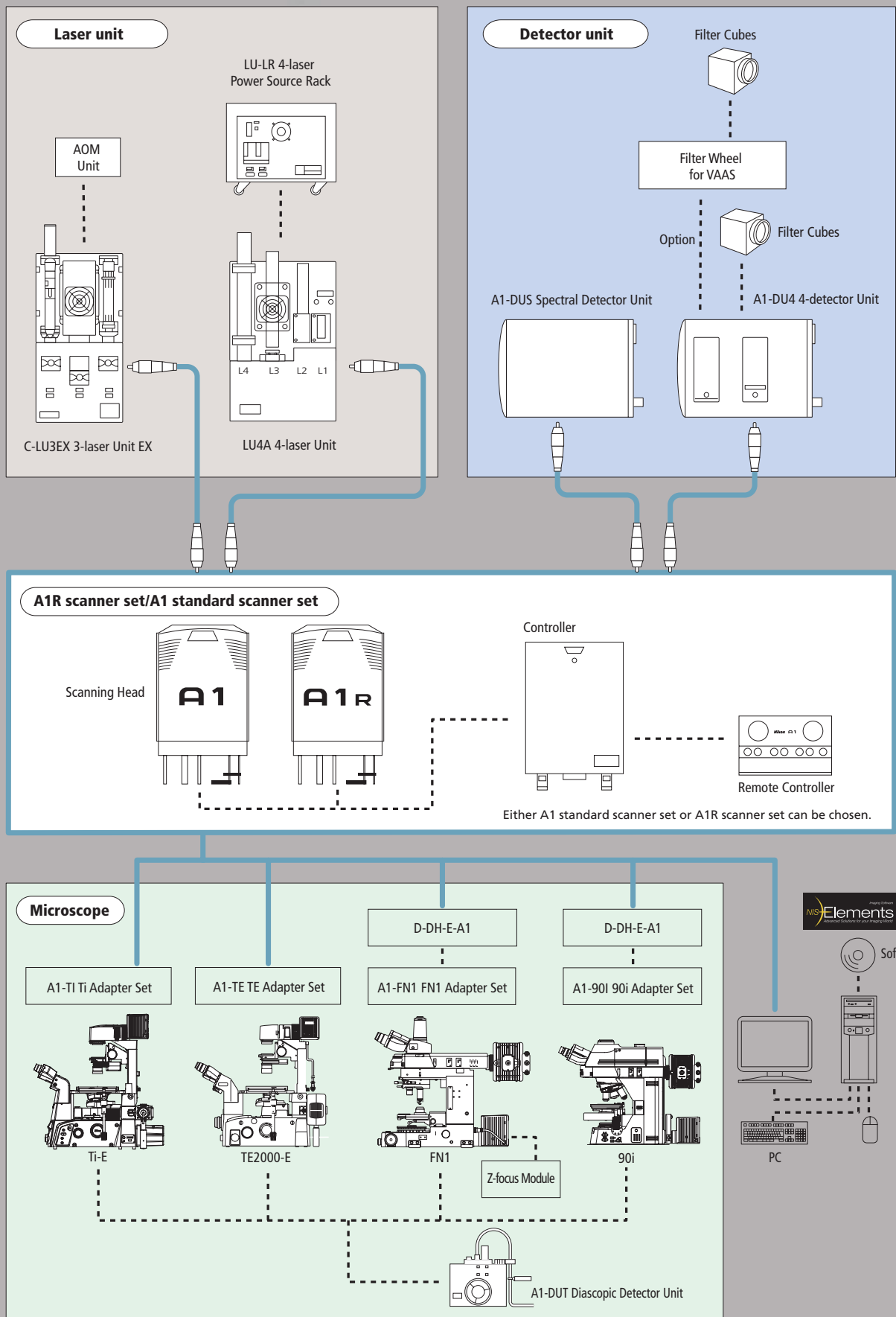


##### Easy operation by remote controller

The remote controller allows the regulation of major settings of laser, detector, and scanner with simple operation using push buttons and dials.



# System diagram



## System components



A1 Scanning Head



4-detector Unit



A1R Scanning Head



Spectral Detector Unit



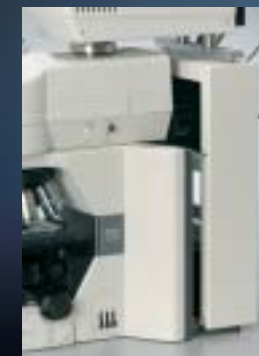
4-laser Unit



Diascopic Detector Unit



4-laser Power Source Rack



90i Adapter Set



3-laser Unit EX

## Recommended objective lenses

CFI Plan Apochromat 10x	NA 0.45, W.D. 4.0 mm
CFI Plan Apochromat 20xVC	NA 0.75, W.D. 1.0 mm
CFI Plan Apochromat 40xC	NA 0.95, W.D. 0.14 mm
CFI Plan Apochromat VC 60xWI	NA 1.20, W.D. 0.27 mm
CFI Apochromat TIRF 60x Oil	NA 1.49, W.D. 0.13 mm
CFI Apochromat TIRF 100x Oil	NA 1.49, W.D. 0.12 mm
CFI Plan Fluor 10x	NA 0.30, W.D. 16.0 mm
CFI Plan Fluor 20x	NA 0.50, W.D. 2.1 mm
CFI Plan Fluor 40x	NA 0.75, W.D. 0.66 mm
<b>NEW</b> CFI Apochromat LWD 40xWI λS	NA 1.15, W.D. 0.60 mm
<b>NEW</b> CFI Apochromat 40xWI λS	NA 1.25, W.D. 0.18 mm
<b>NEW</b> CFI Apochromat 60xH λS	NA 1.4, W.D. 0.14 mm
<b>NEW</b> CFI Plan Apochromat IR 60xWI	NA 1.27, W.D. 0.17 mm

## Recommended filters

Excitation laser	Channel 1	Channel 2	Channel 3	Channel 4
405/488/561/638	450/50	525/50	595/50	700/75
405/488/543/638	450/50	515/30	585/65	700/75
457/514	482/35	540/30	—	—

For filters other than the above, please consult your local Nikon representative.



## Diverse peripherals and systems for pursuit of live cell imaging



### CFI Plan Apochromat VC series objectives

The VC lens corrects aberrations up to the viewfield periphery and eliminates shading, providing uniform high resolution throughout the viewfield. Axial chromatic aberration is corrected up to 405 nm (h line), making this series perfect for confocal observations and photo activation with a semiconductor laser. The frequently used 20x objective has been added recently to this series.

CFI Plan Apo VC 100x Oil, NA 1.40  
CFI Plan Apo VC 60x Oil, NA 1.40  
CFI Plan Apo VC 60x WI, NA 1.20  
CFI Plan Apo VC 20x, NA 0.75

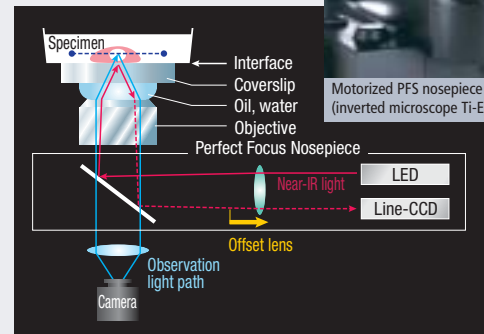
### Confocal microscope with Perfect Focus System

With the inverted microscopes Ti-E, an automatic focus maintenance mechanism—Perfect Focus System (PFS) can be used. It continuously corrects focus drift during long time-lapse observation and when adding reagents.

\*Use with glass bottom dish is recommended.



#### Concept of the Perfect Focus System



The diagram shows the case when an immersion type objective is used. A dry type objective is also available.

### Motorized stages

Motorized stage makes multipoint observation easy. It allows multipoint XYt (4D), multipoint XYZ (4D), multipoint XYZt (5D) and multipoint XYZtλ (6D, including spectral information) observations. By using the standard motorized stage or motorized XY stage equipped with a linear encoder with enhanced positioning repeatability in combination with the optional motorized piezo Z stage with high-speed Z-direction scanning capability, high-speed line Z scans\* are possible.



Standard motorized XY stage



Motorized Piezo Z stage

### Multi-mode imaging system—A1 with TIRF system

The laser TIRF system and the confocal microscope system A1 series can be mounted simultaneously on the inverted microscope Ti-E. The laser TIRF system incorporates an epi-fluorescence module. Switching and adjusting of alignment is easy between the two light sources.

By combining the observations of single molecules with laser TIRF and the sectioning capabilities of the A1, this system allows for multi-perspective cellular analysis.



CFI Apochromat TIRF 60x Oil, NA 1.49 (left)  
CFI Apochromat TIRF 100x Oil, NA 1.49 (right)



### Stage incubation system INU series

Temperature of the stage, water bath, cover, and objective lens is controlled, allowing living cells to be maintained for a long period. A transparent glass heater prevents condensation, and focus drift due to heat expansion on the stage surface is prevented, making this system ideal for lengthy time-lapse imaging applications.

Manufactured by Tokai Hit Co., Ltd.



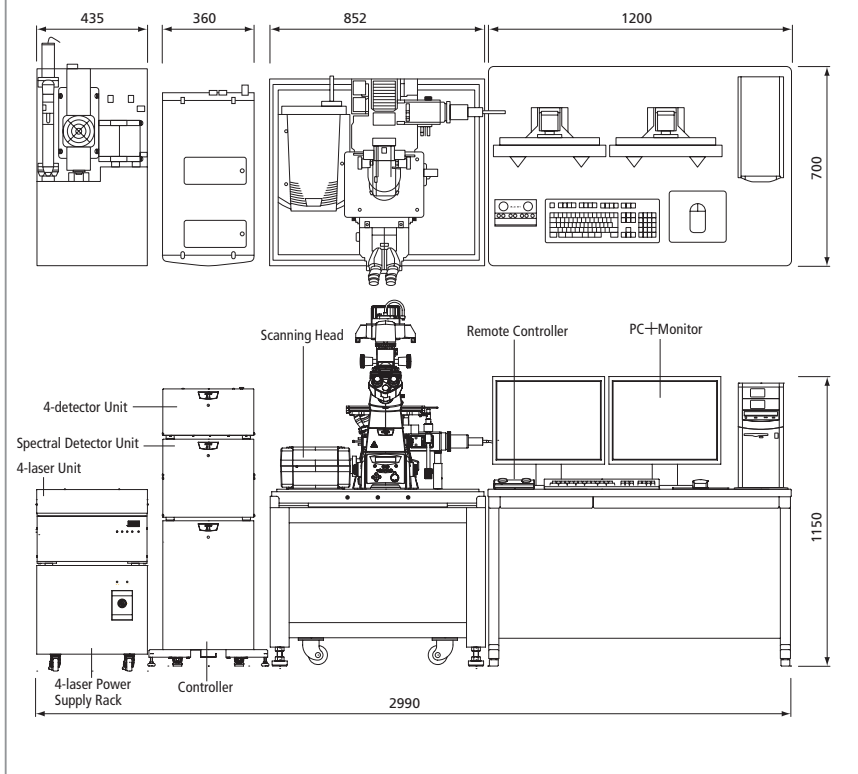
## Specifications

Input/output port		2 laser input ports 4 signal output ports (for 4-PMT detector, spectral detector, VAAS, optional detector)	
Laser	Mountable Laser	Laser diode (405 nm, 36 mW), Ar laser (457 nm/478 nm <sup>**</sup> /488 nm/514 nm, 60 mW max.), Solid-state laser (488 nm, 20 mW), Solid-state laser (561 nm, 25 mW max.), G-HeNe laser (543 nm, 1 mW), Laser diode (638 nm, 10 mW), Laser diode (440 nm, 20 mW)	
	Modulation	Method: AO (Acousto Optical) device or drive current control Control: power control for each wavelength, Return mask, ROI exposure control	
	Laser unit	Standard: LU4A 4-laser unit Optional: C-LU3EX 3-laser unit EX	
Standard fluorescence detector	Wavelength	400-750 nm	
	Detector	4 PMT	
	Filter cube	6 filter cubes commonly used for a microscope mountable on each of three filter wheels Recommended wavelengths: 450/50, 482/35, 525/25, 595/50, 700/75, 540/30, 515/30, 585/65	
Diascopic detector (Option)	Wavelength	485-650 nm	
	Detector	PMT	
Scanning head	Scanning	Standard image acquisition Scanner: non-resonant scanner x2 Pixel size: max. 4096 x 4096 pixels Scanning speed: standard 1 fps (512 x 512 pixels), max. 4 fps (512 x 512 pixels) Zoom: 1-1000x continuously variable Scanning mode: X-Y, XY rotation, Free line*, Line Z High-speed image acquisition Scanner: resonant scanner (X-axis, resonance frequency 7.8 kHz), non-resonant scanner (Y-axis) Pixel size: max. 512 x 512 pixels Scanning speed: 30 fps (512 x 512 pixels) to 420 fps (512 x 32 pixels), 15,600 lines/sec (line speed) Zoom: 7 steps (1x, 1.5x, 2x, 3x, 4x, 6x, 8x) Scanning mode: X-Y, Line Acquisition method: Standard image acquisition, High-speed image acquisition, Simultaneous photo activation and image acquisition	Standard image acquisition Scanner: non-resonant scanner x2 Pixel size: max. 4096 x 4096 pixels Scanning speed: standard 1 fps (512 x 512 pixels), max. 4 fps (512 x 512 pixels) Zoom: 1-1000x continuously variable Scanning mode: X-Y, XY rotation, Free line*, Line Z
		Dichroic mirror	Low-angle incidence method, Position: 8 Standard filter: 405/488, 405/488/561, 405/488/561/638, 405/488/543/638, 457/514, BS20/80 Optional filter: 457, 405/488/543, 457/514/561
Spectral detector (with non-resonant scanner) (Option)	Pinhole	12-256 μm variable (1st image plane)	
	Number of channels	32 channels	
	Corresponding wavelength	400-750 nm	
	Spectral image acquisition speed	4 fps (256 x 256 pixels), 1000 lps	
	Wavelength resolution	2.5 nm, 6 nm, 10 nm Wavelength range variable in 0.25 nm steps	
Unmixing	High-speed unmixing, Precision unmixing		
FOV	Square inscribed in a ø18 mm circle		
Image bit depth	12 bits		
Z step	0.025 μm (with FN1 stepping motor: 0.05 μm)		
Compatible microscopes	ECLIPSE Ti-E inverted microscope, ECLIPSE TE2000-E inverted microscope, ECLIPSE 90i upright microscope, ECLIPSE FN1 fixed stage microscope		
Option	Motorized XY stage, High-speed Z stage, VAAS		
Software	Display/image generation	2D analysis, 3D volume rendering/orthogonal, 4D analysis, spectral unmixing	
	Image format	JPG, TIFF, BMP, GIF, PNG, ND2, JFF, JTF, AVI, ICS/IDS	
	Application	FRAP, FLIP, FRET, photo activation, three-dimensional time-lapse imaging, multipoint time-lapse imaging, colocalization	
Control computer	OS	Microsoft Windows® Vista Business 64-bit SP1 (Japanese version /English version)	
	CPU	Intel Xeon X5570 (2.93 GHz/8 MB/1333 MHz/quad core) or higher	
	Memory	12 GB standard	
	Hard disk	SAS (15,000 rpm), 146 GB or more x2, RAID 0 configuration	
	Data transfer	Dedicated data transfer I/F	
	Monitor	1600 x 1200 or higher resolution, dual monitor configuration recommended	
Recommended installation conditions	Temperature 5 to 35 °C, humidity 65 % (RH) or less (non-condensing) Recommended temperature for spectral detector is 23 ± 5 °C.		

\*Under development \*\*Special 1st DM for 478 is required. Please consult with your local sales representative

# Layout

Unit: mm



## Power source

A1R/A1 system	Confocal system (scanner set, laser unit)	100 VAC	7 A
	Computer unit	100 VAC	14.6 A
Laser	Ar laser (457 nm, 488 nm, 514 nm)	100 VAC	15 A
	Except Ar laser (457 nm, 488 nm, 514 nm)	100 VAC	3 A
Microscope	Inverted microscope Ti-E with HUB-A and epi-fluorescence illuminator	100 VAC	5.3 A

Note: When an air compressor is used with a vibration isolated table, an additional power source of 15 A/100 V is necessary.

## Dimensions and weight

LU4A 4-laser unit	438(W) x 301(H) x 690(D) mm	Approx. 43 kg (without laser)
LU-LR 4-laser power source rack	438(W) x 400(H) x 800(D) mm	Approx. 20 kg (without laser power source)
Scanning head	276(W) x 163(H) x 364(D) mm	Approx. 13 kg
Controller	360(W) x 580(H) x 600(D) mm	Approx. 40 kg
A1-DU4 4-detector unit	360(W) x 199(H) x 593.5(D) mm	Approx. 16 kg (approx. 22 kg with VAAS)



The AOTF incorporated into the 4-laser unit and the AOM optionally incorporated into the 3-laser unit are classified as controlled products (including provisions applicable to controlled technology) under foreign exchange and trade control laws. You must obtain government permission and complete all required procedures before exporting this system.

Specifications and equipment are subject to change without any notice or obligation on the part of the manufacturer. March 2010 ©2008-10 NIKON CORPORATION

**WARNING** TO ENSURE CORRECT USAGE, READ THE CORRESPONDING MANUALS CAREFULLY BEFORE USING YOUR EQUIPMENT.

\* Monitor images are simulated.

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