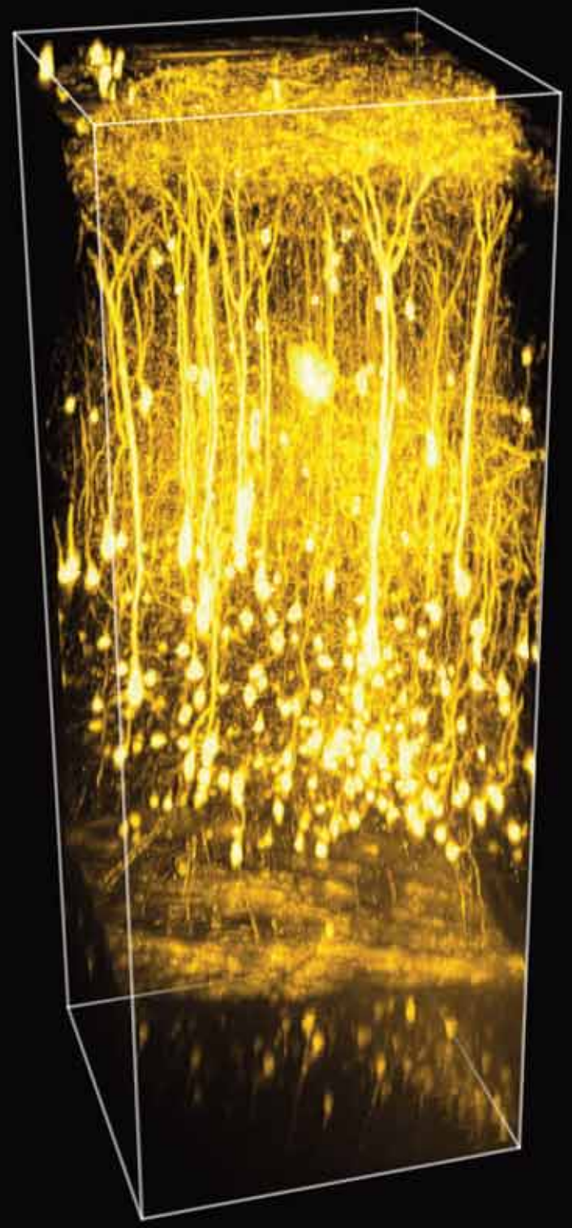
Nikon

A1 MP+

Multiphoton confocal microscope

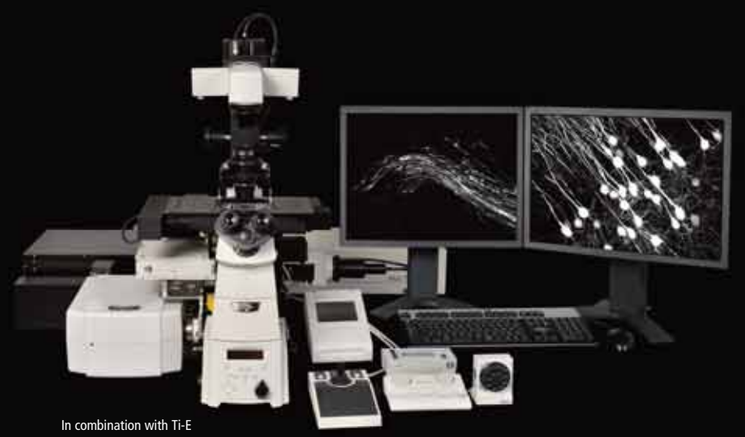


In combination with Ni-E

The A1 MP+/A1R MP+ multiphoton confocal microscopes provide faster and sharper imaging from deeper within living organisms, extending the boundaries of traditional research techniques in biological sciences.

- Ultrahigh-speed imaging up to 420 frames per second (fps) (512 x 32 pixels) with multiphoton imaging using A1R MP+' high efficiency optics and resonant scanner.
- Deep specimen imaging with high-sensitive non-descanned detectors (NDD) located close to the back aperture of the objective lens. Newly developed ultrasensitive gallium arsenide phosphide (GaAsP) NDD allows much deeper *in vivo* imaging of mouse brain over 1.2 mm.
- Auto laser alignment function quickly corrects the IR laser beam shift caused after changing the multiphoton excitation wavelength.
- The IR laser is coupled to the microscope using a compact Incident Optical Unit that contains an acousto-optic modulator and features auto-alignment functions.
- The A1R MP+ system is compatible with a wavelength of 1080 nm.
- Compatible with both upright and inverted microscopes. Provides optimum multiphoton imaging configurations for brain research, other neuroscience applications and *in vivo* imaging of living specimens.

Amazingly deep — A1 MP+/A1R MP+ sharply visualize ultra-deep dynamics within living organisms.



In combination with Ti-E

In combination with FN1

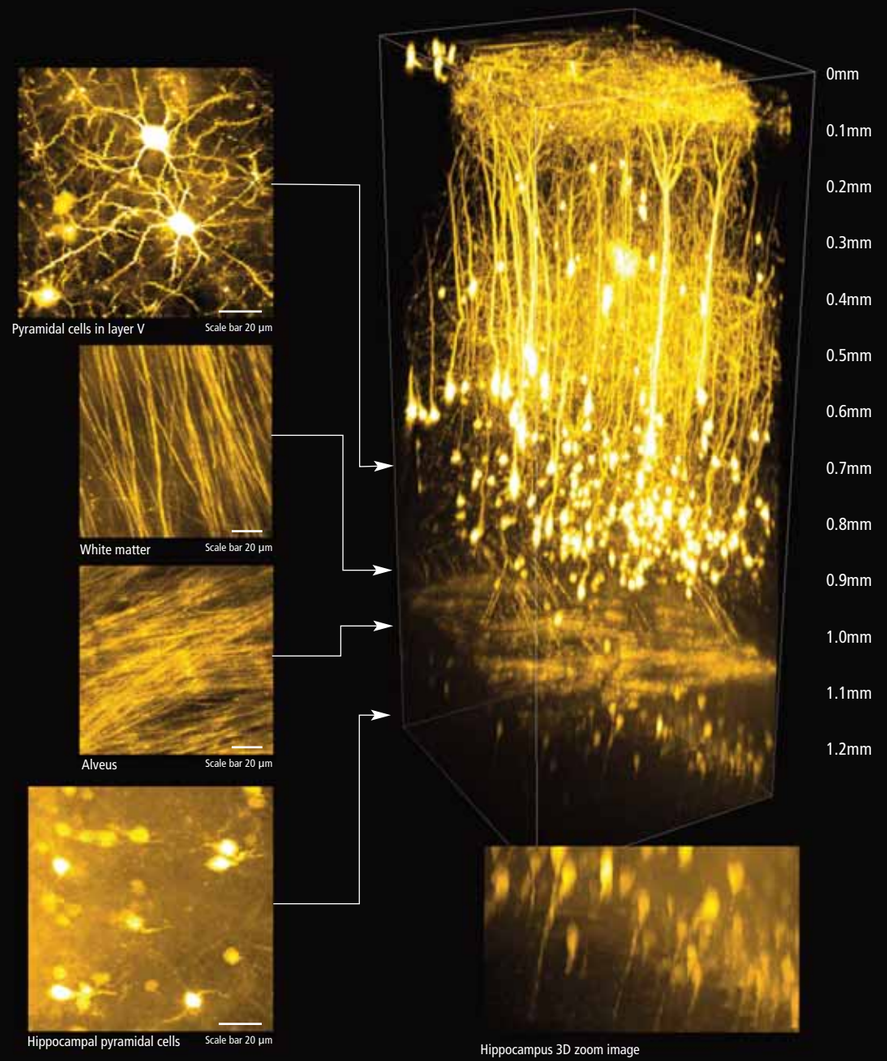
Ultra-deep imaging with the new GaAsP NDD

The new ultrasensitive GaAsP NDD allows clear *in vivo* imaging in deeper areas than ever before and is powerful enough to analyze the mechanisms, such as brain neurons, of living specimens.

GaAsP

Deep brain imaging in *in vivo* mouse

In vivo imaging of an anesthetized YFP-H mouse (4-week-old) via open skull method. Visualization of the entire layer V pyramidal neurons and the deeper hippocampal neurons. Deep imaging achieved for 3-dimensional imaging of hippocampal dendrites over 1.1 mm into the brain.



Captured with episcopic GaAsP NDD and CFI75 Apochromat 25xW MP objective lens (NA 1.10, WD 2.0 mm)

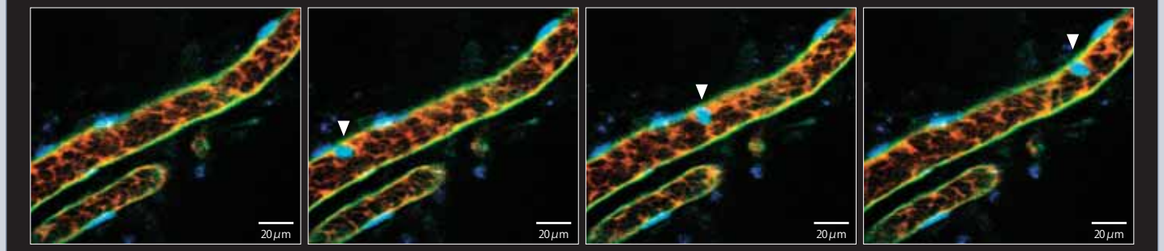
Photographed with the cooperation of:
Dr. Ryosuke Kawakami, Dr. Terumasa Hibi, Dr. Tomomi Nemoto, Research Institute for Electronic Science, Hokkaido University

Fast multiphoton imaging, powerful enough for *in vivo* imaging

The Nikon resonant scanner is capable of high-speed 420-fps imaging, the world's fastest for a multiphoton microscope using point scanning technology. Unique to this design is a resonant scan mirror capable of imaging full fields of view at much higher speeds than traditional galvano scanners. Nikon's optical pixel clock system, which monitors the position of the resonant mirror in real time, adjusts the pixel clock to ensure more stable, geometrically correct and more evenly illuminated imaging even at high speeds. This enables the successful visualization of *in vivo* rapid changes, such as reactions in living organisms, dynamics and cell interactions.

Visualization of intravital microcirculation

Blood cells in blood vessels within a living organism were excited by a femtosecond pulsed IR laser with the A1R MP+' ultrahigh-speed resonant scanner, and their movements were simultaneously captured in three successive fluorescence images at 30 fps (30 msec), with three separate color channels.
The arrowhead indicates the tracking movement of the white blood cell nucleus.
Three fluorescent probes are simultaneously excited and imaged—nucleus (blue), endothelium (green), and plasma (red).
The long-wavelength ultrafast laser in combination with the ultrahigh-speed resonant scanner effectively reduces photodamage and makes time resolved multiphoton imaging of biomolecules possible.



3.46 sec | 3.49 sec | 3.52 sec | 3.55 sec

Image resolution: 512 x 512 pixels, Image acquisition speed: 30 fps, Objective: water immersion objective 60x

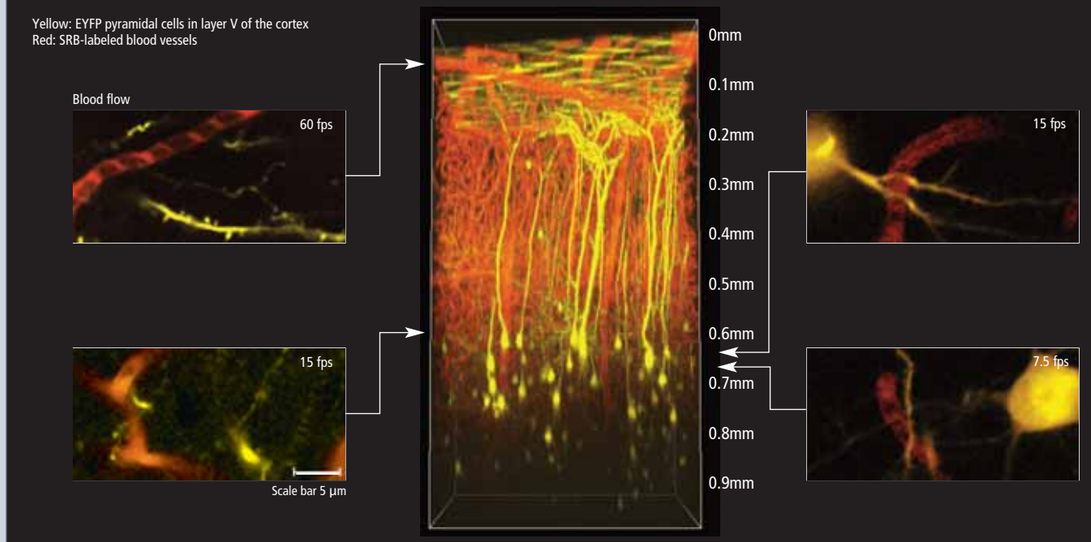
Photographed with the cooperation of: Dr. Satoshi Nishimura, Department of Cardiovascular Medicine, the University of Tokyo, TSBMI, the University of Tokyo, PRESTO, Japan Science and Technology Agency

Mouse brain *in vivo* high-speed imaging

The cerebral cortex of an anesthetized YFP-H mouse (4-week-old) was studied with the open skull method. SRB (Sulforhodamine B) was injected into the tail vein. Using resonant scanning with episcopic GaAsP NDD, blood flow can be imaged at various deep Z positions.

GaAsP

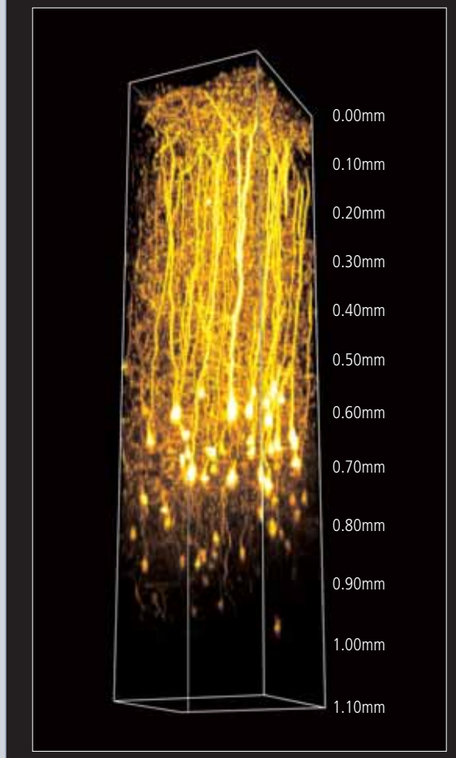
Yellow: EYFP pyramidal cells in layer V of the cortex
Red: SRB-labeled blood vessels

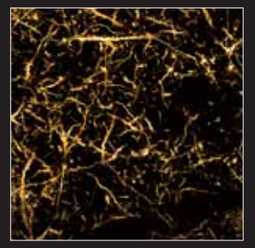


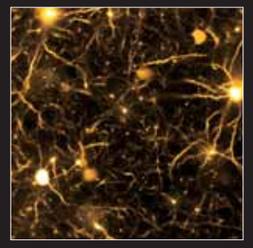
Photographed with the cooperation of:
Dr. Ryosuke Kawakami, Dr. Terumasa Hibi, Dr. Tomomi Nemoto, Research Institute for Electronic Science, Hokkaido University

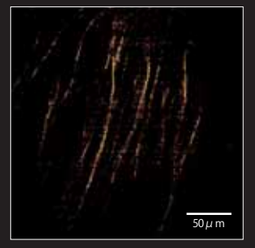
Deep imaging of living specimens with highly efficient standard NDD

In vivo image of deep areas of cerebral cortex of a mouse









The cerebral cortex of an H-line 5-week-old mouse was studied with the open skull method. The entire shape of dendrites of pyramidal cells in layer V expressing EYFP were visualized from the bottom layer into a superficial layer. In addition, the fluorescence signal of white matter in deeper areas was also studied.

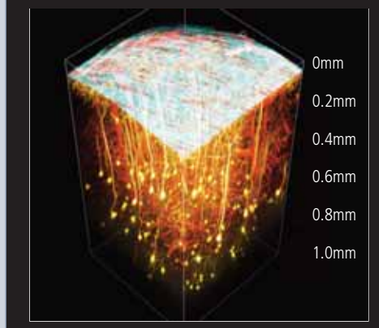
Left) 3D reconstruction image
Right) Z-stack images
Top: dendrites located in superficial layers in the layer V pyramidal cells 25 μm from the surface
Middle: basal dendrites in the layer V pyramidal cells 625 μm from the surface
Bottom: fluorescence from white matter

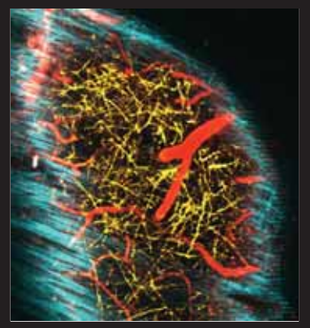
Excitation wavelength: 930 nm
Objective: CFI75 Apochromat 25xW MP (NA 1.10 WD 2.0)

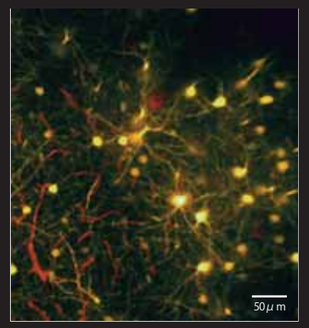
Photographed with the cooperation of:
Dr. Tomomi Nemoto, Research Institute for Electronic Science, Hokkaido University
Dr. Shigenori Nonaka, National Institute for Basic Biology
Dr. Takeshi Imamura, Graduate School of Medicine, Ehime University

Mouse cerebral cortex multi-color imaging

Simultaneous acquisition of three channels in anesthetized YFP-H mouse using IR excitation of 950 nm and imaging Second Harmonic Generation (SHG) and two fluorescence emissions.







Cyan: SHG signal of dura mater
Yellow: EYFP pyramidal neurons in layer V of the cortex
Red: SRB-labeled blood vessels

Photographed with the cooperation of:
Dr. Ryosuke Kawakami, Dr. Terumasa Hibi, Dr. Tomomi Nemoto, Research Institute for Electronic Science, Hokkaido University

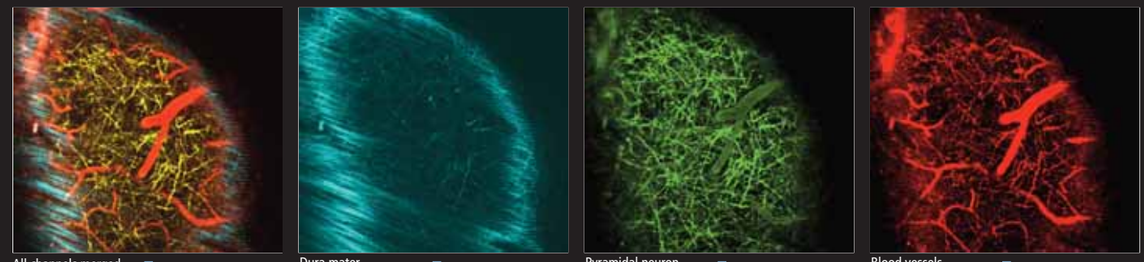
Channel unmixing

With multiphoton excitation, fluorophores have a considerably broader profile of the absorption spectra than with single photon excitation. Therefore simultaneous excitation of multiple fluorophores with single excitation wavelength is possible. Additionally, the wavelength of a pulsed laser for multiphoton excitation can be changed and the user can select a suitable and well-balanced wavelength for the excitation of multiple fluorophores.
A1 MP+/A1R MP+ NDD and channel unmixing technology enables the user to clearly isolate multiple fluorophores and obtain information on the minute structure of a specimen deep within a living organism.

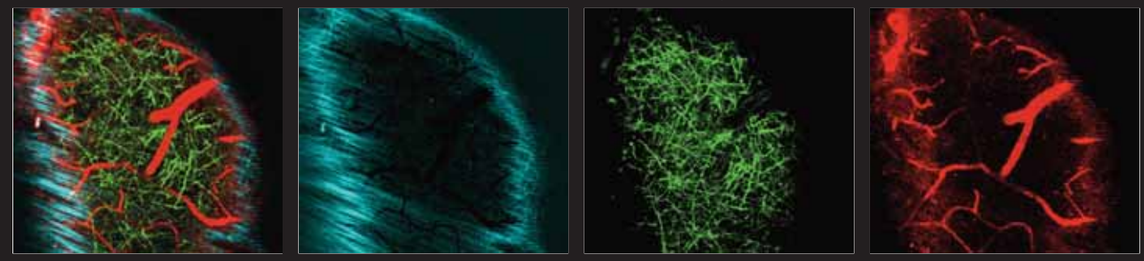
Unmixing with three-color simultaneous excitation

Simultaneous imaging of three colors in anesthetized YFP-H mouse with IR excitation of 950 nm
The upper four images are acquired original data and the lower four images are unmixed images by utilizing the unmixing function. Blood vessels and neurons are clearly separated.

Acquired



Unmixed

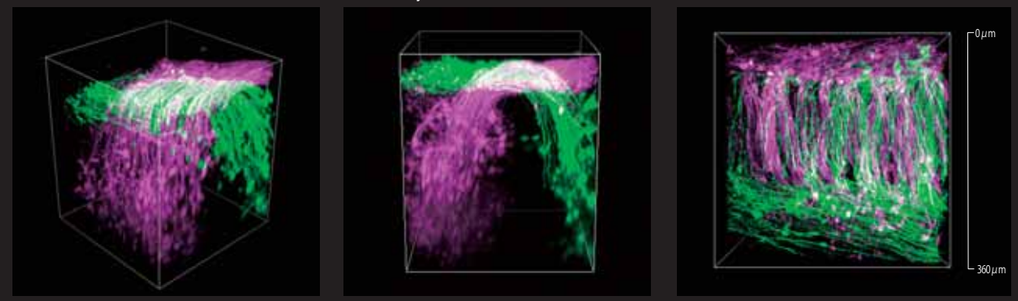


Cyan: SHG signal of dura mater
Yellow: EYFP pyramidal neurons in layer V of the cortex
Red: SRB-labeled blood vessels

Photographed with the cooperation of:
Dr. Ryosuke Kawakami, Dr. Terumasa Hibi, Dr. Tomomi Nemoto, Research Institute for Electronic Science, Hokkaido University

Unmixing with two-color simultaneous excitation

Spinal cord primordia (neural tube) of a 12.5-day-old rat embryo
The entire embryo was cultured for approximately 44 hours after transfection of the right and left nerve cells with eGFP and YFP (Venus) by electroporation.
A cross-sectional slice of spinal cord was embedded in gel and simultaneous excitation of eGFP and YFP was conducted using pulsed IR laser (930 nm).
The image is captured with NDD and processed by the unmixing function.
Observation of interneuron and its commissural axon is clearly achieved.



Photographed with the cooperation of:
Dr. Noriko Osumi, Dr. Masanori Takahashi, Division of Developmental Neuroscience, United Center for Advanced Research and Translational Medicine (ART), Tohoku University Graduate School of Medicine

Multiphoton imaging gallery

Four-color imaging of human colon cancer cells in *in vivo*

Three-dimensional volume rendering of implanted subcutaneous tumor of HCT116 expressing Fucci. The cell cycle of tumor cells and the environment (collagen fiber and vessels) are visualized. Upper right, only collagen fiber and vessels are shown.

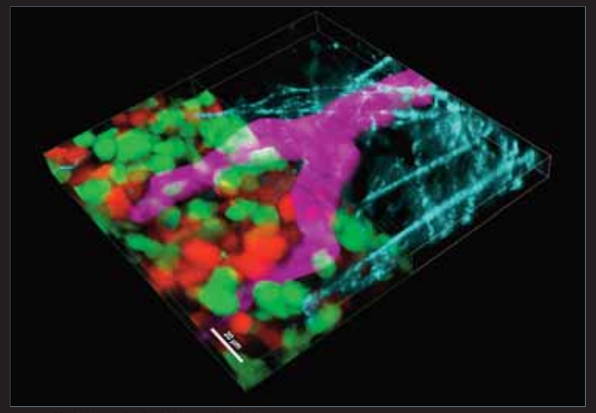


Image width: 156.61 μm, height: 156.61 μm, depth: 22.50 μm

Red: Fucci mKO2/cancer cell
Green: Fucci mAG/cancer cell
Cyan: SHG/collagen fiber
Purple: Qtracker655/neovascular vessels

Objective: CFI Plan Fluor 20xA MI
Excitation Wavelength: 940 nm

Photographed with the cooperation of Dr. Yoshinori Kagawa and
Dr. Masaru Ishii, Immunology Frontier Research Center, Osaka University

Dynamic *in vivo* imaging of granulocytes in live adipose tissues

The epididymal adipose tissue of a LysM-EGFP mouse was observed using intravital multiphoton microscopy.
Granulocytes patrolling around adipocytes were visualized.
Time-lapse images show the movement of the granulocytes.
(arrowhead : granulocyte-A, arrow : granulocyte-B)

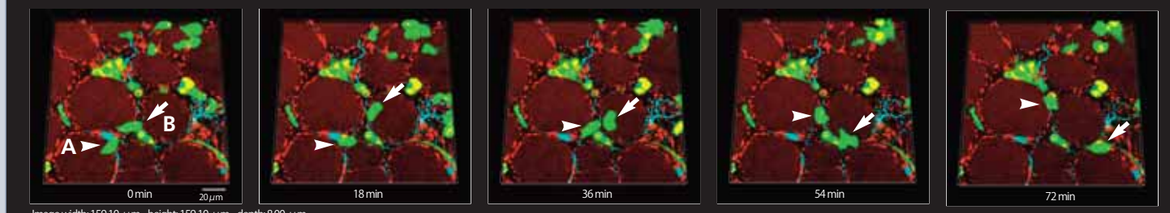


Image width: 159.10 μm, height: 159.10 μm, depth: 8.00 μm

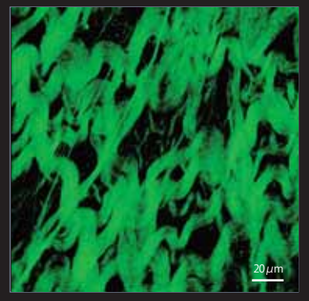
Red: BODIPY /fat droplet
Green: EGFP /granulocyte
Cyan: Hoechst /nucleus and SHG/collagen fiber

Objective: CFI Apochromat LWD 40x WI λS
Excitation Wavelength: 920 nm

Photographed with the cooperation of Junichi Kikuta, Shoko Yasuda and Dr. Masaru Ishii, Laboratory of Cellular Dynamics,
Immunology Frontier Research Center, Osaka University

Knitted stitch structure of colon wall muscle by SHG imaging

NOD/SCID mouse colon wall was observed toward mucosal membrane from serosal membrane side. Knitted stitch structure of colon wall muscle fibers was clearly visualized using SHG. Left, maximum intensity projection calculated from Z stack. Right, three-dimensional volume rendering using depth-code pseudo color.



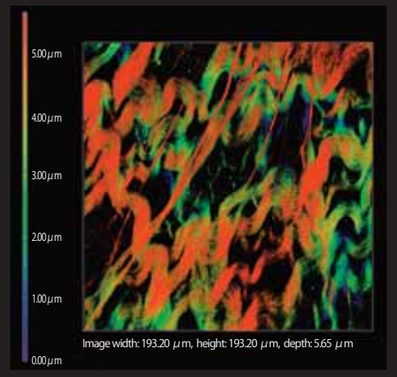


Image width: 193.20 μm, height: 193.20 μm, depth: 5.65 μm

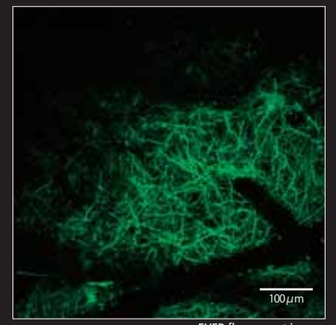
SHG of collagen fiber

Objective: CFI Plan Fluor 20xA MI
Excitation Wavelength: 840 nm

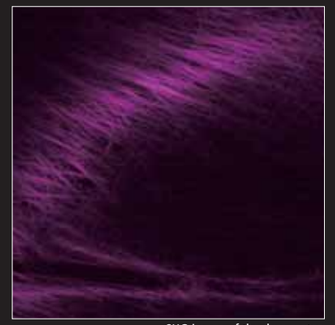
Photographed with the cooperation of Dr. Yoshinori Kagawa and
Dr. Masaru Ishii, Immunology Frontier Research Center, Osaka University

SHG image of the brain surface of a mouse

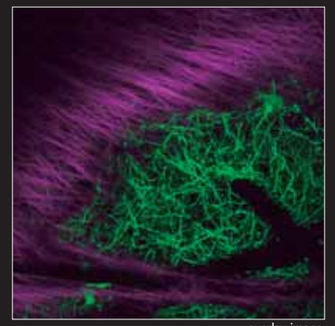
The neocortex of an H-line 5-week-old mouse was studied with the open skull method.
The SHG signals from dura mater and EYFP fluorescence signals were simultaneously acquired using the NDD.



EYFP fluorescent image



SHG image of the dura mater



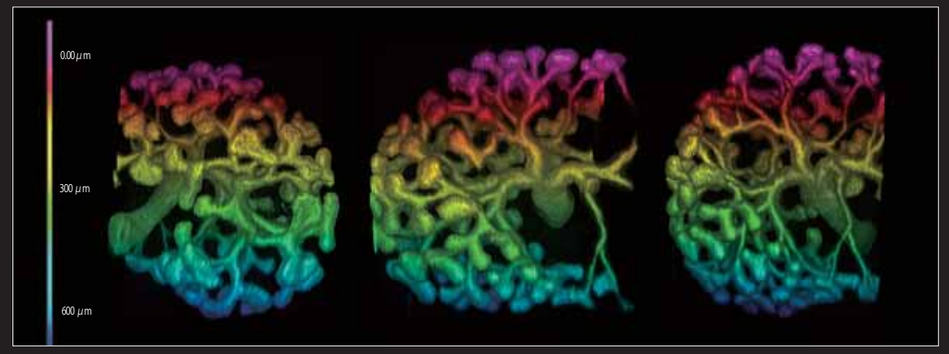
overlay image

Excitation wavelength: 950 nm
Objective: CFI75 Apochromat 25xW MP (NA 1.10 WD 2.0)

Photographed with the cooperation of:
Dr. Takeshi Imamura, Graduate School of Medicine, Ehime University
Dr. Yusuke Oshima, Dr. Shigenori Nonaka, National Institute for Basic Biology
Dr. Terumasa Hibi, Dr. Ryoshuke Kawakami, Dr. Tomomi Nemoto, Research Institute for Electronic Science, Hokkaido University

3D volume rendering images

Three-dimensional volume renderings of a kidney labeled with Hoxb7/myrVenus marker (Chi et al, 2009 Genesis),
using depth-code pseudocolor volume rendering to reference Z depths (pseudocolored by depth - 1 μm step for 550 μm).

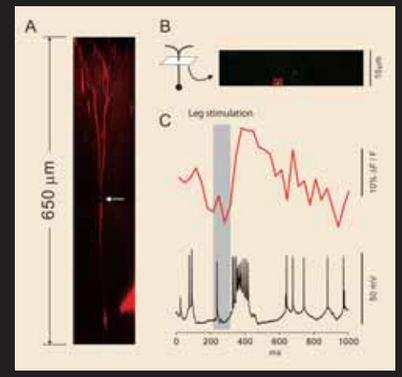


Objective: CFI Apochromat 25xW MP, Scan zoom: 1x, Z step size: 1 μm, IR excitation wavelength: 930 nm
Image resolution: 1024x1024 pixels, Image volume: 460 μm (length) x 460 μm (width) x 600 μm (height)
Photographed with the cooperation of Dr. Frank Costantini and Dr. Liza Pon, Columbia University Medical Center, New York

Ca^{2+} signals from the layer V pyramidal neuron

A: A two-photon image of Alexa 594 fluorescence from the layer V pyramidal neuron of the primary somatosensory area of a mouse rear leg. Alexa 594 and a fluorescent calcium indicator (OBG-1) were loaded to the soma using whole-cell recording.
B: An OBG-1 fluorescence image of the neurodendrites in the X-Y plane (indicated with an arrow in fig. A).
C: The upper graph (red) shows Ca^{2+} signaling of the neurodendrites (indicated with a red box in fig. B) evoked by electrical stimulation of the legs (stimulated during the period indicated with grey in fig. C). The lower graph (black) shows membrane potential changes in the soma.

Photographed with the cooperation of Dr. Satoshi Manita and
Dr. Masanori Murayama, Brain Science Institute (BSI), Riken



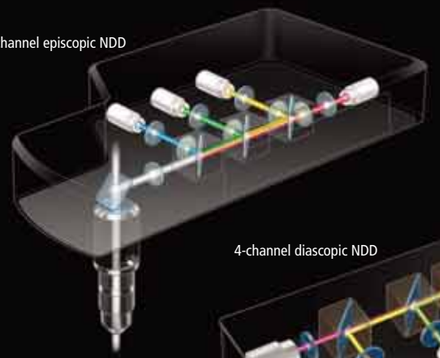
A1 MP+/A1R MP+ achieve the most advanced multiphoton imaging

Standard NDD

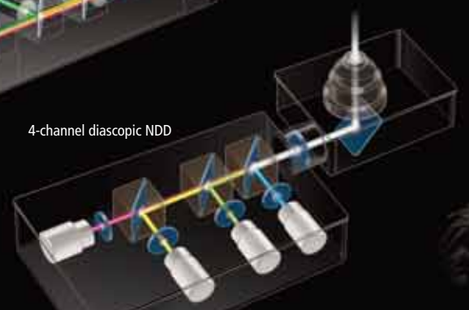
The fluorescence emissions from deep within a specimen are highly scattered in multiphoton excitation, and therefore the conventional detector using a pinhole cannot provide bright fluorescent images. The episcopic NDD in the A1 MP+/A1R MP+ is located close to the back aperture of the objective to detect the maximum amount of scattered emission signals from deep within living specimens. The use of this four-channel detector in combination with special spectral mirrors, together with Nikon's unmixing algorithm, eliminates cross talk between fluorescent probes with highly overlapping emission spectra. Background auto-fluorescence is also eliminated, enabling high-contrast image capture from deep within the specimen.
Using diascopic NDD* together with episcopic NDD, brighter images can be acquired by detecting fluorescence signals from both reflected and transmitted.

*Compatible with Ni-E focusing nosepiece microscope

4-channel episcopic NDD



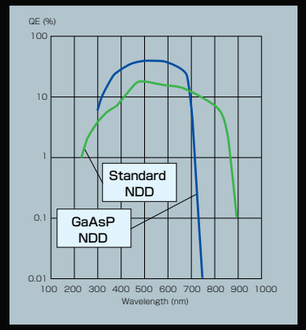
4-channel diascopic NDD

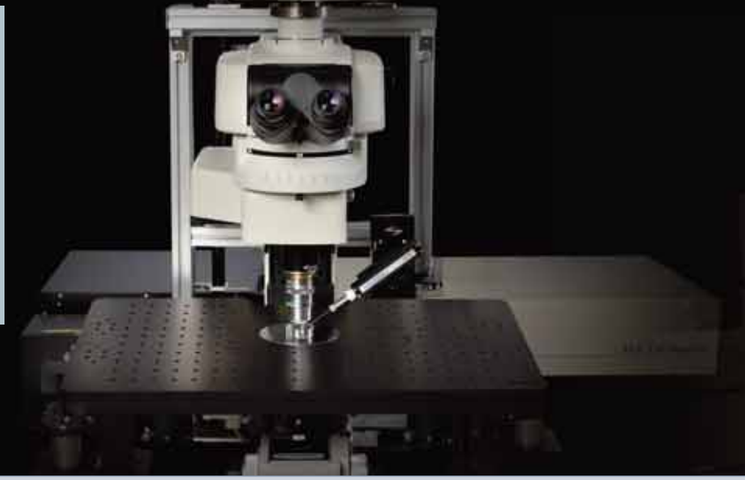


Super High-sensitive GaAsP NDD

The newly developed episcopic GaAsP NDD* has approximately twice the sensitivity of a standard NDD and allows clear imaging of deeper areas of living specimens than ever before. Its ability to acquire bright images enables faster imaging and higher quality Z-stack imaging. Its high sensitivity allows acquisition of fluorescent signals with less laser power, resulting in less photo damage to living specimens.

* Compatible with Ni-E, FN1, Ti-E





Nikon's high-NA objectives are ideal for multiphoton imaging

High-NA objectives have been developed that highly correct chromatic aberrations over a wide wavelength range, from ultraviolet to infrared. Transmission is increased through the use of Nikon's exclusive Nano Crystal Coat technology.
In particular, the CFI Apochromat 25xW MP objective lens provides an industry leading highest numerical aperture of 1.10 while still maintaining a 2.0 mm working distance. It also has a collar that corrects spherical aberrations depending on the depth of the specimen and a 33° manipulator pipette access angle, making it ideal for deep multiphoton imaging and physiology research applications.

Nano Crystal Coat is a Nikon exclusive lens coating technology using an ultralow refractive index nanoparticle thin film originally developed for the semiconductor fabrications industry. The Nano Crystal Coat particle structure dramatically reduces stray reflections and boosts transmission over a wide wavelength range, producing images with higher signal-to-noise (S/N) ratios.

Objectives			
CFI75 Apochromat 25xW MP	NA 1.10	WD 2.0	Nano Crystal Coat
CFI Apochromat LWD 40xWI λS	NA 1.15	WD 0.6	Nano Crystal Coat
CFI Apochromat 40xWI λS	NA 1.25	WD 0.18	Nano Crystal Coat
CFI Plan Apochromat IR 60xWI	NA 1.27	WD 0.17	Nano Crystal Coat

Auto laser alignment when changing multiphoton excitation wavelength

When the multiphoton laser wavelength or group velocity dispersion pre-compensation is changed, the multiphoton laser beam positional pointing at the objective back aperture may also change, resulting in uneven intensity across the image, or a slight misalignment between the IR and visible laser light paths.

Verifying the IR laser beam pointing and setting the alignment has traditionally been difficult. Nikon's A1 MP+ series' auto laser alignment function, housed in the Incident Optical Unit for the multiphoton excitation light path, automatically maximizes IR laser alignments with a single click in NIS-Elements C.

Auto laser alignment with a single click

Two types of scanning head enable high-speed, high-quality imaging

A1 MP+ is equipped with a galvano (non-resonant) scanner for high-resolution imaging. A1R MP+ is a hybrid scanning head that incorporates both galvano and ultrahigh-speed resonant scanners. A1R MP+ allows imaging and photoactivation at ultrafast speeds necessary for revealing cell dynamics and interaction.

High-resolution imaging — A1 MP+ A1R MP+

The A1 MP+/A1R MP+ galvano scanner enables high-resolution imaging of up to 4096 x 4096 pixels. In addition, with the newly developed scanner driving and sampling systems, plus Nikon's unique image correction technology, high-speed acquisition of 10 fps (512 x 512 pixels) is also possible.

1D scanning	5,200 lps (lines per second)
2D scanning	130 fps (512 x 32 pixels)
Full frame scanning	10 fps (512 x 512 pixels)

Ultrafast imaging — A1R MP+

A1R MP+ is a hybrid scanning head equipped with both a galvano scanner and a resonant scanner with an ultrahigh resonance frequency of 7.8 kHz.
It allows ultrafast imaging and photoactivation at 420 fps (512 x 32 pixels), the world's fastest image acquisition.

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

Stable, ultrafast imaging

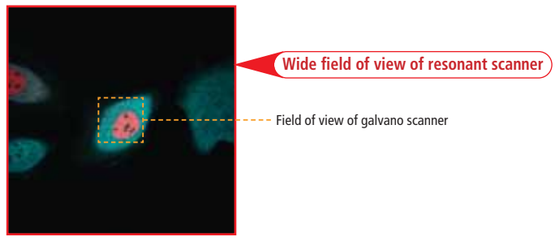
The Nikon original optical clock generation method is used for high-speed imaging with a resonant scanner. Stable clock pulses are generated optically, offering images that have neither flicker nor distortion even at the highest speed.

High-speed data transfer with fiber-optic communication

High-speed data transfer with fiber-optic communication
The fiber-optic communication data transfer system can transfer data at a maximum of 4 Gbps. This allows the transfer of five channels of image data (512 x 512 pixels, 16 bit) at 30 fps.

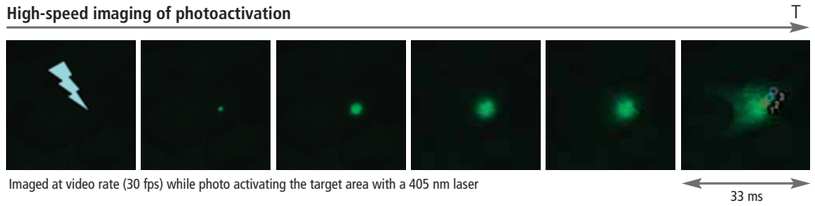
Wide field of view

Resonant scanners do not suffer from overheating of the motor during high-speed image acquisition. Therefore, it is not necessary to reduce the field of view of the scanned image in order to avoid overheating, thus enabling a wide field of view.

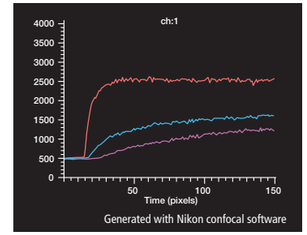


Simultaneous photoactivation and imaging

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

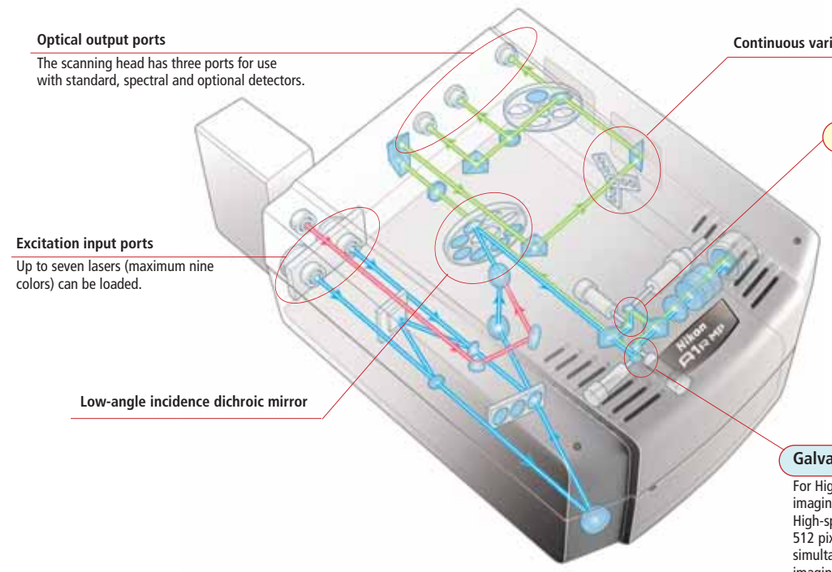


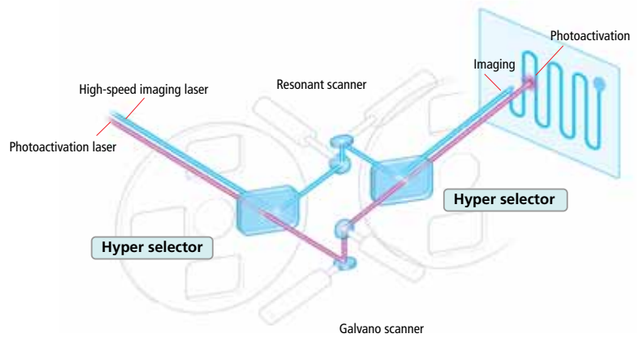
Imaged at video rate (30 fps) while photo activating the target area with a 405 nm laser



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

Optical path in the A1R MP+ scanning head





What is a hybrid scanner?

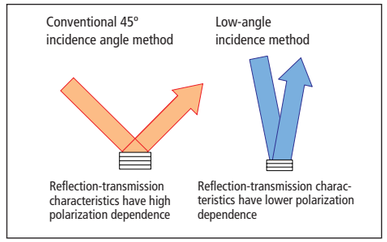
This mechanism allows flexible switching or simultaneous use of two scanners (resonant and galvano) with the use of a hyper selector.

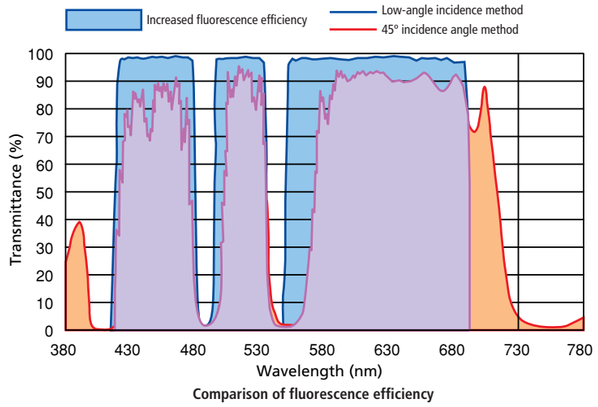
Key Nikon innovations for improving image quality

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

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





Comparison of fluorescence efficiency

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.

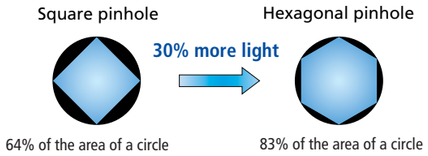
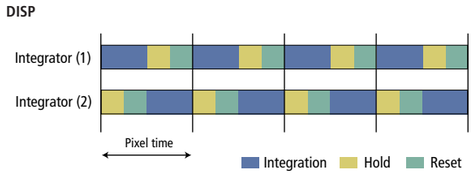


Image of a zebrafish labeled with four probes (captured with galvano scanner)
Nucleus (blue): Hoechst33342, Pupil (green): GFP, Nerve (yellow): Alexa555, Muscle (red): Alexa647
Photographed with the cooperation of: Dr. Kazuki Horikawa and Prof. Takeharu Nagai, Research Institute for Electronic Science,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

Dual integration signal processing

Nikon's original Dual Integration Signal Processing (DISP) 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.



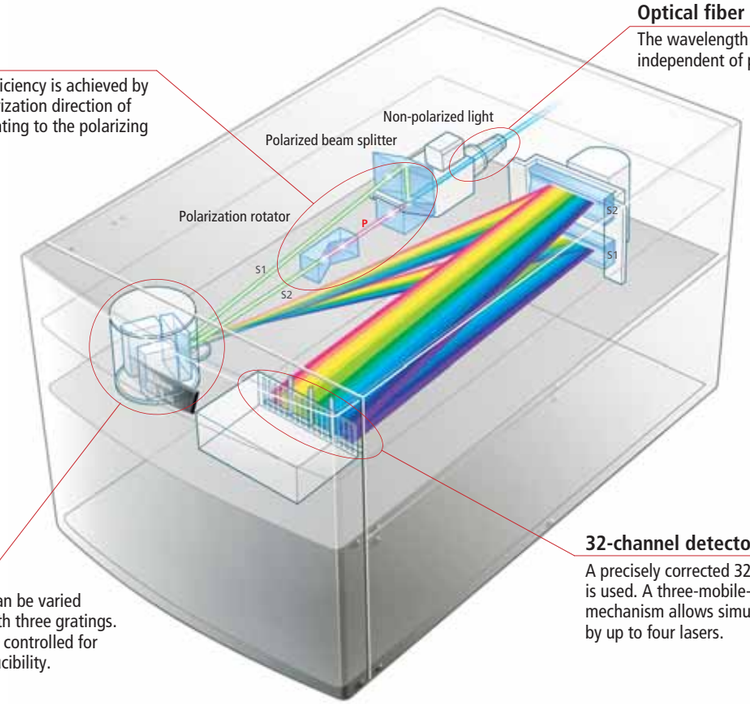
Two integrators work in parallel as the optical signal is read to ensure there are no gaps.

VAAS pinhole unit (option)

It is widely recognized that reducing the pinhole size to eliminate flare light from the non-focal plane causes darker confocal images. With the innovative VAAS pinhole unit, image sharpness can be increased and brightness retained without reducing the pinhole size.

Enhanced spectral detector

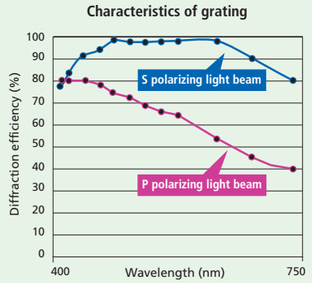
Nikon's original spectral performance is even further enhanced in the A1 MP+ series, allowing high-speed spectral acquisition with a single scan. In addition, advanced functions, including real-time unmixing, are incorporated.



High-quality spectral data acquisition

Diffraction Efficiency Enhancement System (DEES)

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

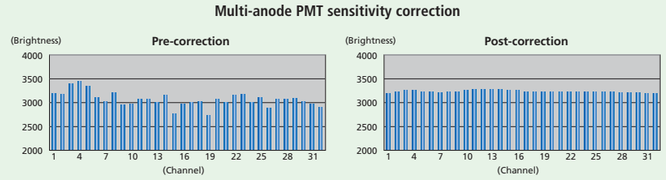


High-efficiency fluorescence transmission technology

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: inter-channel 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.

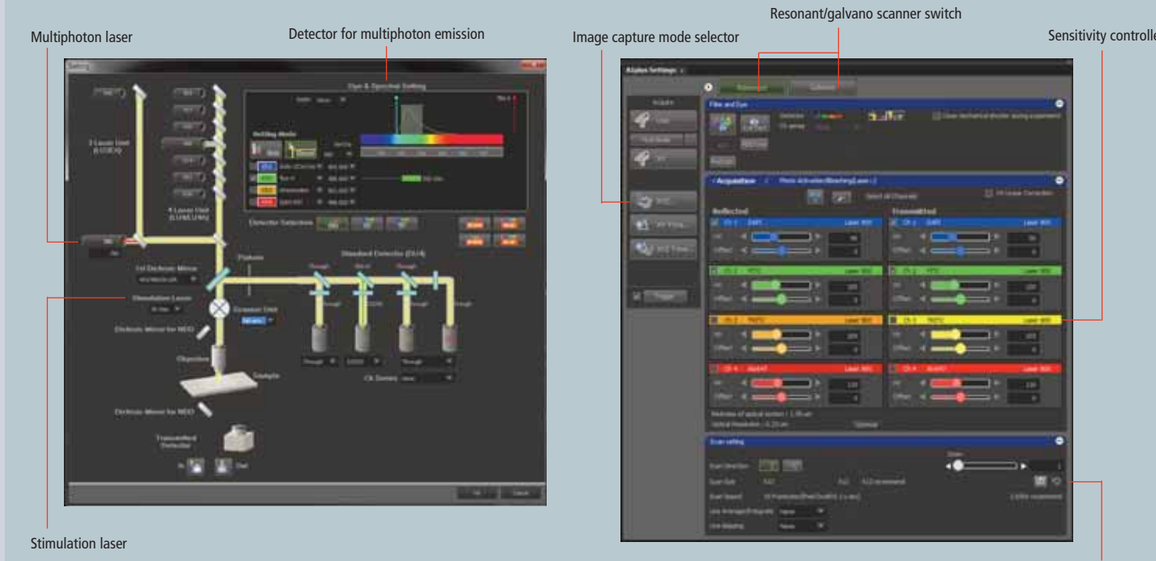


Intuitive, easy-to-use software for multiphoton imaging

NIS-Elements C Acquisition and Analysis software

Simple operations common with Nikon confocal microscopes

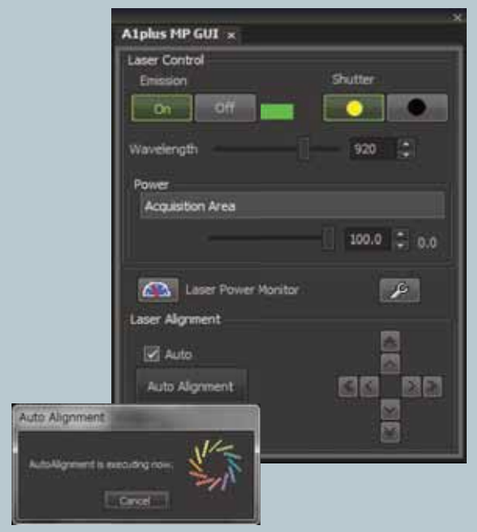
- All necessary operations for image capture are displayed in one window.
- Lasers and detectors for visible laser excitation can be switched simply by selecting fluorescent probe to be used.
- One-touch switching of high speed resonant scanner and high-resolution galvano (non-resonant) scanner
- Simultaneous photoactivation with high speed imaging is possible with visible laser excitation.



Functions for high quality multiphoton imaging

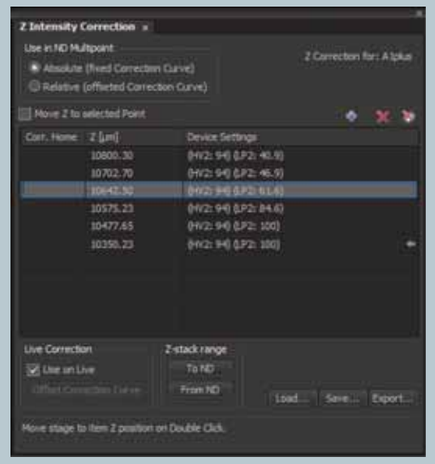
Auto laser alignment function

The IR laser alignment can be quickly optimized with a single click when changing the multiphoton excitation wavelength



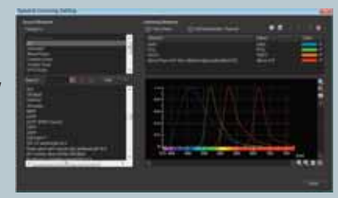
Z-intensity control function

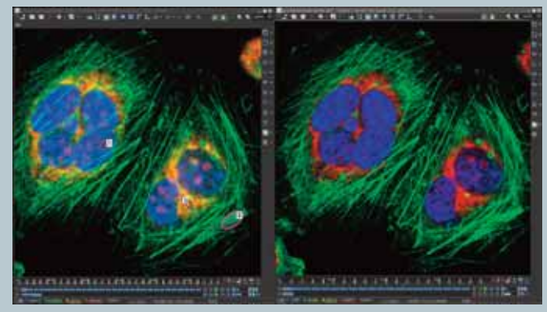
Users can define the laser power and PMT gain to use at different depths in a Z series using the Z intensity control function, so that even when imaging dense and thick specimens, the intensity of the emission is maintained throughout the specimen.



Channel unmixing function

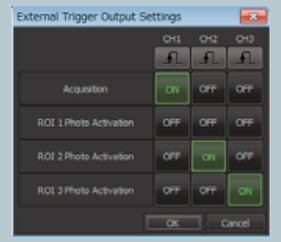
Nikon's channel unmixing allows you to obtain emissions from multiple NDD PMTs simultaneously, using one IR excitation wavelength, and unmix overlapping emission spectra.





External trigger function

A1 MP+/A1R MP+ and NIS Elements C support triggering applications. This is effective for synchronizing frame and scanning times with electrophysiology recordings, or to externally trigger the confocal to scan.

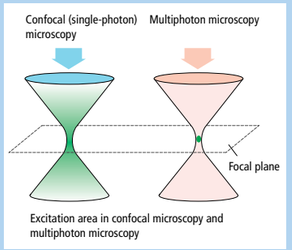


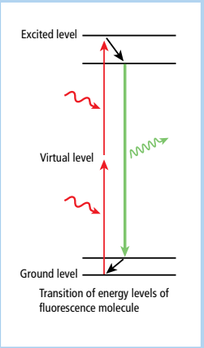
Microscope control

NIS-Elements C allows microscope functions such as focusing, as well as switching objective lenses and light paths, to be controlled from a PC. In addition, by memorizing optical configurations, NIS-Elements C enables observation methods to be changed with a single click of the mouse.

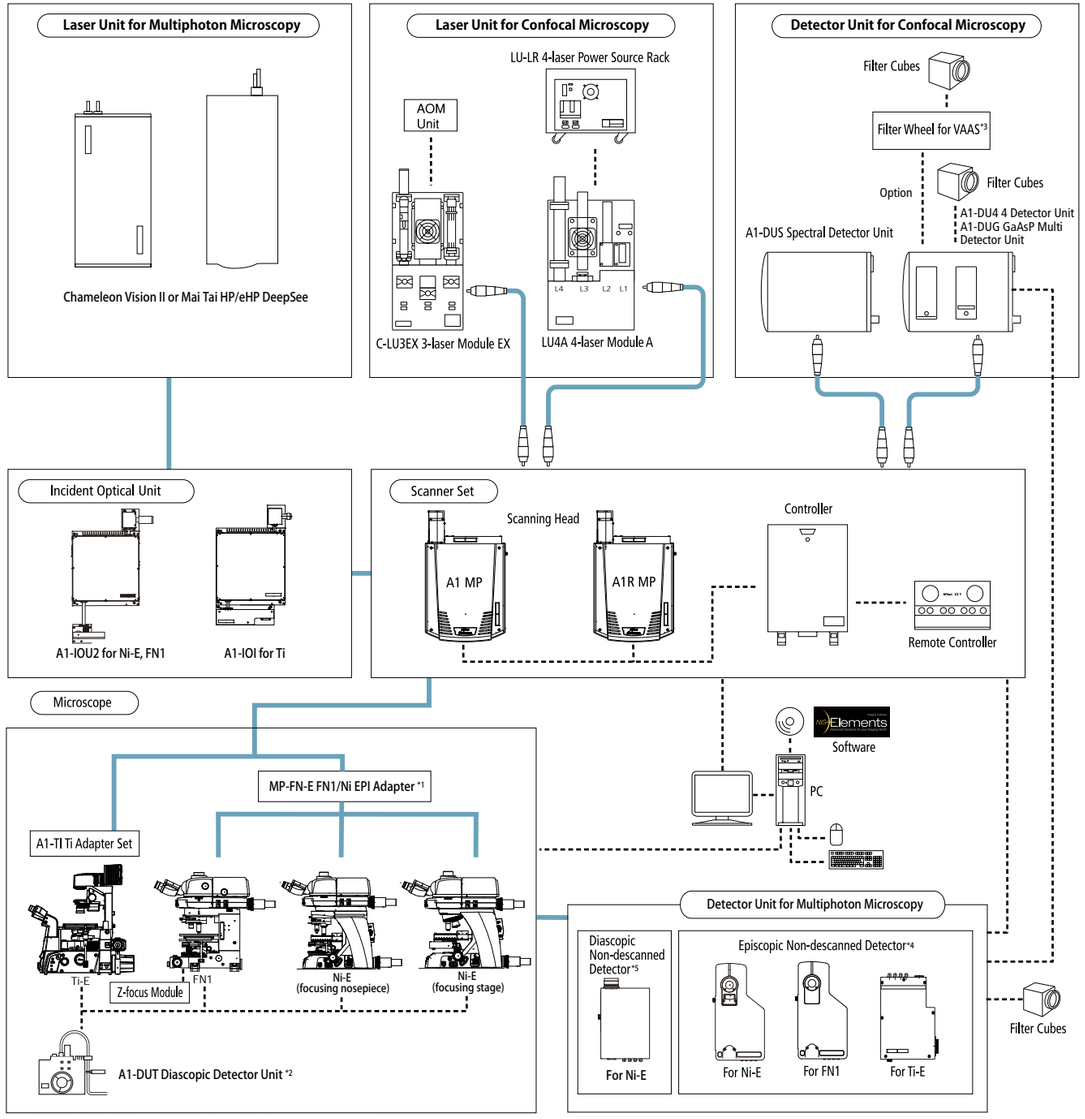
Principle of multiphoton excitation

When two photons are absorbed simultaneously by a single fluorescent molecule (two-photon excitation), the excitation efficiency is proportional to the square of the excitation light intensity.
In order to achieve multiphoton excitation, a pulsed beam with high photon density or flux is used. Because the laser beam is delivered in very short (femtosecond) pulses and is converged on a focal point through an objective lens, the probability of simultaneous absorption of two photons becomes high enough to be useful for imaging.
In two-photon excitation, the excitation efficiency decreases inversely with the fourth power of the distance from the center of the focal volume. As a result, only fluorescence molecules located within the diffraction-limited focal volume of the objective lens are excited and can emit fluorescence. This principle allows the use of non-descanned detectors (NDD's), where an emission pinhole is not necessary to achieve confocal results.
There is less absorption and scattering of near infrared light than visible wavelengths through a specimen so the excitation beam can easily penetrate deep into thick tissue. Because two photon excitation is highly confined to only the diffraction-limited focal volume of the objective lens, the need for a confocal pinhole aperture to block the emitted fluorescence from out of focus plane from reaching the detector is eliminated. Photo damage to a specimen can be minimized, and maximum fluorescence detection is made possible, creating conditions suitable for *in vivo* imaging of living tissue. The combination of the group velocity dispersion pre-compensation "pre-chirping" system incorporated in the multiphoton laser and the use of the non-descanned detector (NDD) allows fluorescence imaging deeper into a specimen than is possible with standard confocal technique.





System diagram



*1 When attaching a diascopic detector to the Ni-E, use the MP-FN1/NI DIA/EPI Adapter.
*2 Dedicated adapter is required depending on microscope model.
*3 Only for A1-DU4 4 Detector Unit
*4 Normal PMT type and GaAsP type both available
*5 Normal PMT type only

Femtosecond pulsed lasers

When pulsed light of very short duration, typically about 100 femtoseconds, passes through microscope optics (e.g. objective), the pulse is spread out in time on its way to the specimen because of group velocity dispersion, (the variation by wavelength in velocity of the speed of light through glass substrates),causing a reduction of peak power.
To prevent the reduction of peak pulse power, Nikon has equipped the femtosecond pulsed lasers for multiphoton microscopy with built-in group velocity dispersion precompensation that restores the original pulse width at the specimen. The parameters of the precompensation have been optimized for Nikon's optical system. This enables bright fluorescence imaging of areas deep within a specimen with minimum laser power.

Mai Tai HP/eHP DeepSee, Newport Corp., Spectra-Physics Lasers Division (Nikon specifications)

Chameleon Vision II, Coherent Inc. (Nikon specifications)

Specifications

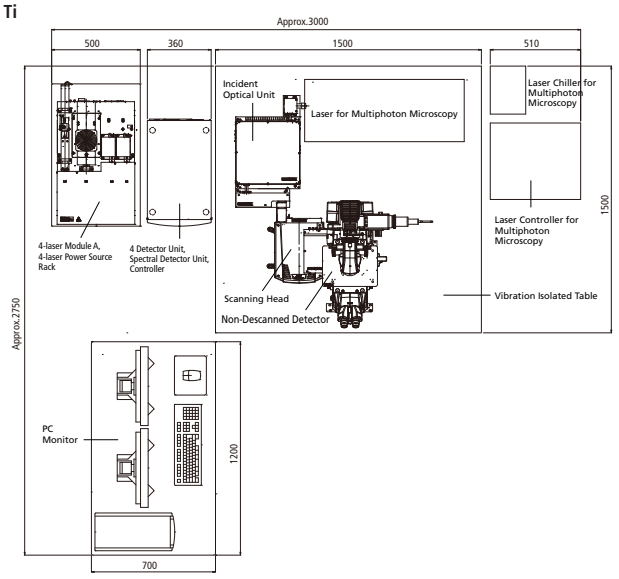
		A1 MP+	A1R MP+
Scanning head input/output port		3 laser input ports 3 signal output ports for standard, spectral and third-party detectors (FCS/FCCS/FLIM) Output port for VAAS can be added*2	
Laser for multiphoton microscopy	Compatible laser	Mai Tai HP/eHP DeepSee (Newport Corp.) Chameleon Vision II (Coherent Inc.)	
	Modulation	Method: AOM (Acousto-Optic Modulator) device Control: power control, return mask, ROI exposure control	
	Incident optics	700-1080 nm, auto alignment	
Laser for confocal microscopy (option)	Compatible laser	405 nm, 440/445 nm, 488 nm, 561/594 nm, 638/640nm, Ar laser (457 nm, 488 nm, 514 nm), HeNe laser (543 nm)	
	Modulation	Method: AOTF (Acousto-Optic Tunable Filter) or AOM (Acousto-Optic Modulator) device Control: power control for each wavelength, return mask, ROI exposure control	
	Laser unit	Standard: LU4A 4-laser module A or C-LU3EX 3-laser module EX Optional: C-LU3EX 3-laser module EX (when 4-laser module is chosen as standard laser unit)	
NDD for multiphoton microscopy	Wavelength	400-650 nm	
	Detector	4 PMTs	
	Filter cube	Filter cubes commonly used for a microscope Recommended filter sets for multiphoton: 492SP, 525/50, 575/25, 629/53, DM458, DM495, DM511, DM560, DM593	
	Detector type	Episcopic NDD (for Ni-E/FN1/Ti-E), Episcopic GaAsP NDD (for Ni-E/FN1/Ti-E) Diascopic NDD (for Ni-E only)	
Standard fluorescence detector (option)	Wavelength	A1-DU4 4 Detector Unit: 400-750 nm (400-650 nm for multiphoton observation) A1-DUG GaAsP Multi Detector Unit: 400-720 nm (400-650 nm for multiphoton observation)	
	Detector	A1-DU4 4 Detector Unit: 4 normal PMTs A1-DUG GaAsP Multi Detector Unit: 2 GaAsP PMTs + 2 normal PMTs	
	Filter cube	6 filter cubes commonly used for a microscope mountable on each of three filter wheels Recommended wavelengths for multiphoton/confocal observation: 450/50, 482/35, 515/30, 525/50, 540/30, 550/49, 585/65, 595/50, 700/75	
Diascopic detector (option)	Wavelength	440-645 nm	
	Detector	PMT	
FOV		Square inscribed in a ø18 mm circle	
Image bit depth		4096 gray intensity levels (12 bit)	
Scanning head	Standard image acquisition	Scanner: galvano scanner x2 Pixel size: max. 4096 x 4096 pixels Scanning speed: Standard mode: 2 fps (512 x 512 pixels, bi-direction), 24 fps (512 x 32 pixels, bi-direction) Fast mode: 10fps (512 x 512 pixels, bi-direction), 130 fps (512 x 32 pixels bi-direction)*1 Zoom: 1-1000x continuously variable Scanning mode: X-Y, X-T, X-Z, XY rotation, Free line	
	High-speed image acquisition	—	Scanner: resonant scanner (X-axis, resonance frequency 7.8 kHz), galvano 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, X-T, X-Z Acquisition method: Standard image acquisition, High-speed image acquisition, Simultaneous photoactivation and image acquisition
	IR laser wavelength range	700 to 1000 nm	700 to 1080 nm
	Dichroic mirror	Low-angle incidence method Position: 8 Standard filter: 405/488, 405/488/561, 405/488/561/638, 400-457/514/IR, 405/488/543/638, BS20/80, IR, 405/488/561/IR	
	Pinhole	12-256 µm variable (1st image plane)	
Spectral detector*3 (option)	Wavelength detection range	400-750nm (400-650nm for multiphoton observation)	
	Number of channels	32 channels	
	Spectral image acquisition speed	4 fps (256 x 256 pixels), 1000 lps	
	Wavelength resolution	80 nm (2.5 nm), 192 nm (6 nm), 320 nm (10 nm) Wavelength range variable in 0.25 nm steps	
	Unmixing	High-speed unmixing, Precision unmixing	
Compatible microscopes		ECLIPSE Ti-E inverted microscope, ECLIPSE FN1 fixed stage microscope, ECLIPSE Ni-E upright microscope (focusing nosepiece type and focusing stage type)	
Z step		Ti-E: 0.025 µm, FN1 stepping motor: 0.05 µm Ni-E: 0.025 µm	
Option		Motorized XY stage (for Ti-E/Ni-E), High-speed Z stage (for Ti-E), High-speed piezo objective-positioning system (for FN1/Ni-E), VAAS*2	
Software	Display/image generation	2D analysis, 3D volume rendering/orthogonal, 4D analysis, spectral unmixing	
	Image format	JP2, JPG, TIFF, BMP, GIF, PNG, ND2, JFF, JTF, AVI, ICS/IDS	
	Application	FRAP, FLIP, FRET (option), photoactivation, three-dimensional time-lapse imaging, multipoint time-lapse imaging, colocalization	
Control computer	OS	Microsoft Windows®7 Professional 64 bits SP1 (Japanese version/English version)	
	CPU	Intel Xeon E5-2667 (2.90 GHz/15 MB/1666 MHz) or higher	
	Memory	16 GB (4 GB x 4 + additional 2 GB x3)	
	Hard disk	300 GB SAS (15,000 rpm) x2, RAID 0 configuration	
	Data transfer	Dedicated data transfer I/F	
	Network interface	10/100/1000 Gigabit Ethernet	
	Monitor	1600 x 1200 or higher resolution, dual monitor configuration recommended	
Vibration isolated table		1800 (W) x 1500 (D) mm required (for FN1/Ni-E), or 1500 (W) x 1500 (D) mm required (for Ti-E)	

*1 Fast mode is compatible with zoom 8-1000x and scanning modes X-Y and X-T. It is not compatible with Rotation, Free line, CROP, ROI, Spectral imaging, Stimulation and FLIM.
*2 Only for A1-DU4 4 Detector unit
*3 Compatible with galvano scanner only

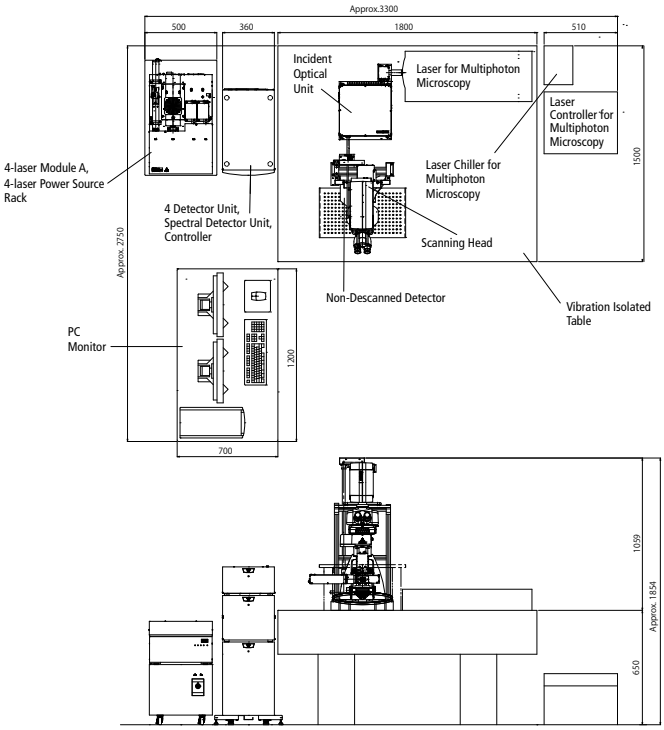
Layout

Unit: mm

With Ti



With FN1



Operation conditions

- Temperature: 20 °C to 25 °C (± 1 °C), with 24-hour air conditioning
- Humidity: 75 % (RH) or less, with no condensation
- Completely dark room or light shield for microscope

Power source

Multiphoton system	Multiphoton system (scanner set, laser unit)	120 VAC	6.7 A
		220 VAC	3.6 A
	Computer unit	120 VAC	12.2 A
		220 VAC	6.6 A
Lazer	Ar laser (457 nm, 488 nm, 514 nm)	120 VAC	12.5 A
		220 VAC	6.8 A
	Except Ar laser (457 nm, 488 nm, 514 nm)	120 VAC	2.5 A
		220 VAC	1.4 A
	Laser for multiphoton microscopy (laser, water chiller, others)	120 VAC	19.2 A
		220 VAC	10.5 A
Microscope	Inverted microscope Ti-E with HUB-A and epi-fluorescence illuminator	120 VAC	4.4 A
		220 VAC	2.4 A

Dimensions and weight

Scanning head	276 (W) x 163 (H) x 364 (D) mm	Approx. 10 kg
Incident optical unit (A1-IOU2)	363 (W) x 186 (H) x 404 (D) mm	Approx. 11 kg
Controller	360 (W) x 580 (H) x 600 (D) mm	Approx. 40 kg
4 Detector Unit	360 (W) x 199 (H) x 593.5 (D) mm	Approx. 16 kg (approx. 22 kg with VAAS)
Spectral detector unit	360 (W) x 323 (H) x 593.5 (D) mm	Approx. 26 kg
Episcopic NDD (for Ti-E)	206 (W) x 60 (H) x 262 (D) mm	Approx. 5 kg
Episcopic NDD(for FN1, Ni-E)	216 (W) x 112 (H) x 425 (D) mm	Approx. 10 kg
Diascopic NDD (for Ni-E)	301 (W) x 66 (H) x 185 (D) mm	Approx. 10 kg
4-laser module	438 (W) x 301 (H) x 690 (D) mm	Approx. 43 kg (without laser)
4-laser power source rack	438 (W) x 400 (H) x 800 (D) mm	Approx. 20 kg (without laser power source)
3-laser module EX	365 (W) x 133 (H) x 702 (D) mm	Approx. 22 kg (without laser)

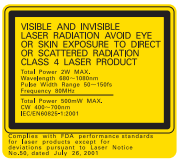
Dimensions exclude projections.

Specifications and equipment are subject to change without any notice or obligation on the part of the manufacturer. December 2012

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

Monitor images are simulated.

N.B. Export of the products* in this brochure is controlled under the Japanese Foreign Exchange and Foreign Trade Law. Appropriate export procedure shall be required in case of export from Japan.
*Products: Hardware and its technical information (including software)



NIKON CORPORATION
Shin-Yurakucho Bldg., 12-1, Yurakucho 1-chome, Chiyoda-ku, Tokyo 100-8331, Japan
phone: +81-3-3216-2375 fax: +81-3-3216-2385
http://www.nikon.com/instruments/

ISO 9001 BUREAU VERITAS Certification — ISO 9001 Certified NIKON CORPORATION Instruments Company

ISO 14001 BUREAU VERITAS Certification — ISO 14001 Certified NIKON CORPORATION

NIKON INSTRUMENTS INC.
1300 Walt Whitman Road, Melville, N.Y. 11747-3064, U.S.A.
phone: +1-631-547-8500; +1-800-52-NIKON (within the U.S.A. only)
fax: +1-631-547-0306
http://www.nikoninstruments.com/

NIKON INSTRUMENTS EUROPE B.V.
Tripolis 100, Burgerweeshuispad 101, 1076 ER Amsterdam, The Netherlands
phone: +31-20-7099-000 fax: +31-20-7099-298
http://www.nikoninstruments.eu/

NIKON INSTRUMENTS (SHANGHAI) CO., LTD.
CHINA phone: +86-21-6841-2050 fax: +86-21-6841-2060
(Beijing branch) phone: +86-10-5831-2028 fax: +86-10-5831-2026
(Guangzhou branch) phone: +86-20-3882-0552 fax: +86-20-3882-0580

NIKON SINGAPORE PTE LTD
SINGAPORE phone: +65-6559-3618 fax: +65-6559-3668

NIKON MALAYSIA SDN. BHD.
MALAYSIA phone: +60-3-7809-3688 fax: +60-3-7809-3633

NIKON INSTRUMENTS KOREA CO., LTD.
KOREA phone: +82-2-2186-8400 fax: +82-2-555-4415

NIKON CANADA INC.
CANADA phone: +1-905-602-9676 fax: +1-905-602-9953

NIKON FRANCE S.A.S.
FRANCE phone: +33-1-4516-45-16 fax: +33-1-4516-45-55

NIKON GMBH
GERMANY phone: +49-211-941-42-20 fax: +49-211-941-43-22

NIKON INSTRUMENTS S.p.A.
ITALY phone: +39-055-300-96-01 fax: +39-055-30-09-93

NIKON AG
SWITZERLAND phone: +41-43-277-28-67 fax: +41-43-277-28-61

NIKON UK LTD.
UNITED KINGDOM phone: +44-208-247-1717 fax: +44-208-541-4584

NIKON GMBH AUSTRIA
AUSTRIA phone: +43-1-972-6111-00 fax: +43-1-972-6111-40

NIKON BELUX
BELGIUM phone: +32-2-705-56-65 fax: +32-2-726-66-45

Printed in Japan (1212-06)T
Code No.2CE-SCAH-4
This brochure is printed on recycled paper made from 40% used material.

En