Nikon

Modular Confocal Microscope System

C1

DIGITAL ECLIPSE

CFI60

Three-dimensional confocal fluorescent images with unsurpassed resolution and contrast—now obtainable at a minimum cost for use in broad bio-research applications

Nikon proudly introduces a universal confocal microscope system that is ultra-compact and lightweight, yet provides confocal images of the highest quality in its class. All main components are modular, including the world's smallest and lightest scanning head, making expansion and maintenance easy. Furthermore, 3-channel detection is possible with minimum upgrade, and operation is facilitated by the intuitive software. With the C1, confocal microscopy is now a mainstream technique affordable by all.

Highest quality optical performance

A successful fusion of Nikon's optical and electronics technologies, the C1's resolution, contrast, and fluorescent image brightness are all top-class and "State of the Art." Image sizes of up to 2K by 2K at 12 bit image depth can be easily scanned. See page 4-5.

Interchangeable filters

Changing the filter to match the fluorescent dyes you want to use is simple and quick, enabling the use of the latest probes or dyes available today. See page 5, 6.

3-channel simultaneous detection

The C1 supports almost any imaging technique required today, including simultaneous 3-channel fluorescence, 3-channel plus DIC, time-lapse recording, and spatial analysis. See page 5, 6.

Modular design saves space and facilitates upgrading

All main components are modular including the laser box, scanner head, and detector module, saving desk space and allowing easy upgrades and maintenance. See page 12.

Intuitive software promotes multifaceted microscopic analysis

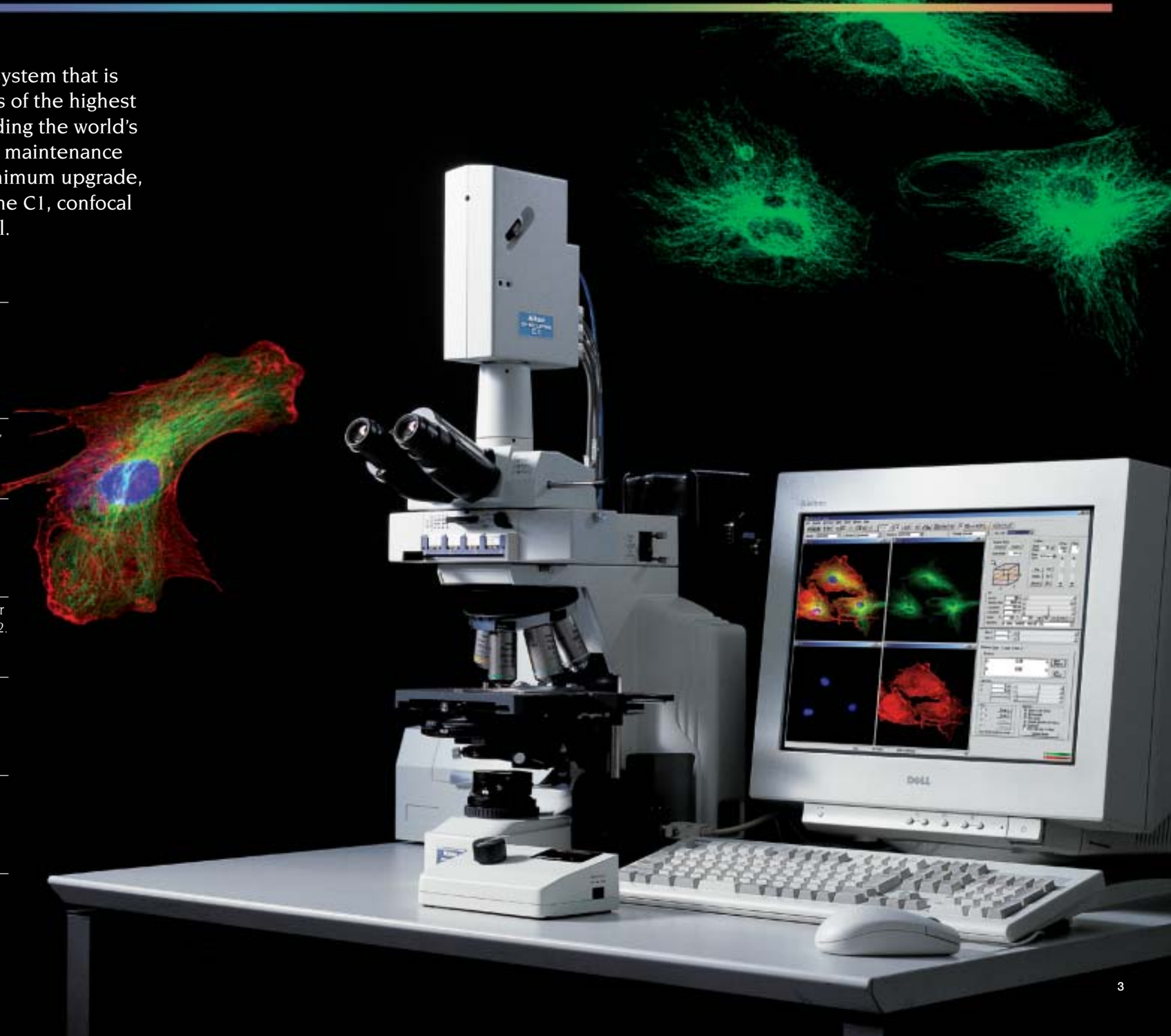
The C1's Graphical User Interface (GUI) is extremely simple and intuitive. From the time of initial use, you may never need to refer to the manual for typical operation. See page 7-11.

Easy to configure, easy to operate

Each of the modules is pre-calibrated, eliminating the need for calibration during setup. Just connect the modules you need, and you are ready for optimal image capture. See page 12.

Compact design

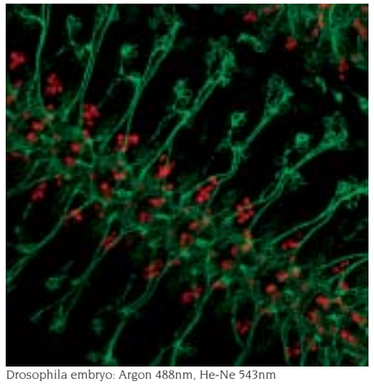
With a small footprint, the C1 does not get in the way of other lab equipment. Consequently, there is plenty of work space left around the microscope. See page 12.



Unprecedented image quality

Why are the C1's images so good?

Every aspect of the C1 that affects image quality has been thoroughly examined—from optical, to mechanical, to electronic—to create a confocal microscope of the highest level ever in this class. In addition, to get the utmost performance out of Nikon's CFI60 series objectives, we developed new complimentary scanning optics expressly for this microscope. The following are the major results of our improvements:



Drosophila embryo: Argon 488nm, He-Ne 543nm

- Stray light, which is usually generated in the scanner head and common in some other designs, is thoroughly eliminated, while reflection loss on the I/O ends of the optical fiber is minimized. This makes it possible to obtain images with extremely high contrast and photon efficiency.
- Fluorescence transmission efficiency has been dramatically improved to obtain fluorescent images 3 times brighter than previous Nikon models. This, coupled with the use of high quantum-efficiency photomultiplier tubes, gives clear, sharp images even with fluorescent specimens that in the past were too dim to observe.
- Signal-to-noise ratio has been increased 7-fold (compared to previous Nikon models), resulting in a significantly improved image quality.
- The new frame grabber A/D image board improves signal quality when converting between analog and digital image signals.
- High-precision scanning facilitated by highly accurate galvanometer scanner control and superb control of sampling signals optimizes high-resolution images of up to 4 million pixels. Even higher zoom magnifications do not impact optical resolution and are usually not required for maximum resolution.
- 12-bit digitization pixel depth ensures the quality, dynamic range and sensitivity that is required of a 4-megapixel image.

CFI60 optical system

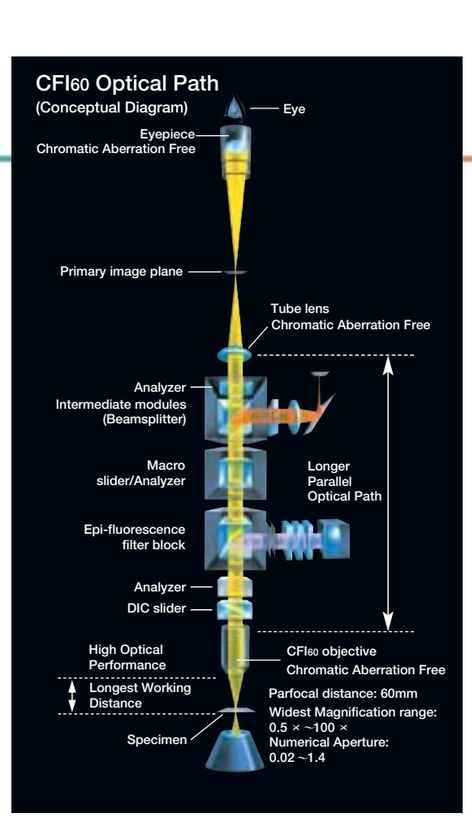
To achieve images of the utmost quality, the C1 adopts the CFI60 infinity optics, the industry-acclaimed optics developed by Nikon utilizing its unique technologies.

Higher N.A.'s, longer working distances, and aberration-free

CFI60 optics achieve both higher N.A.'s and longer working distances than ever before possible, while succeeding in separately correcting both axial and lateral chromatic aberrations in the objective and the tube lens.

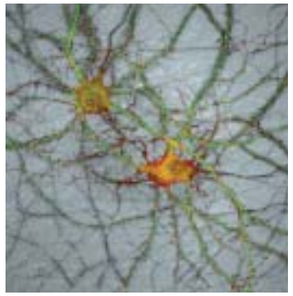
Reduced blur, increased contrast

The CFI60 design dramatically reduces blur during microscopy. To curtail fluorescence light emitted from the objectives themselves, Nikon chose the appropriately formulated glass and optical coatings for the lenses and designed them in the optimal configuration, improving contrast during epi-fluorescence observations. Nikon's CFI60 optical system delivers top-notch performance, enabling its use in increasingly sophisticated biological research.

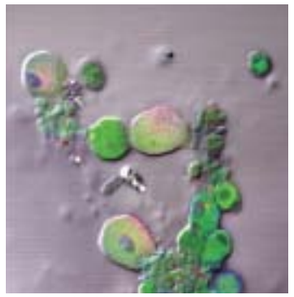


Superbly accommodates various imaging techniques

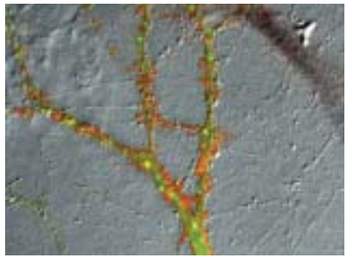
The C1 supports almost any imaging technique required today, including simultaneous 3-channel fluorescence, 3-channel plus diascopic DIC, time-lapse recording, and spatial analysis.



Merged confocal fluorescence and IRM (Interference Reflection Microscopy) image of cultured neurons; Alexa 488 (Ar 488), Alexa 568 (He-Ne 543); Plan Apo 60x oil. ❶



Merged confocal fluorescence and DIC image of excised mouse dorsal root ganglion: Cy3 (He-Ne 543), Cy5 (He-Ne 633), Autofluorescence (Ar 488); Plan Fluor 40x oil. ❷



Merged confocal fluorescence and DIC image of cultured neurons: Alexa 488 (Ar 488), Alexa 568 (He-Ne 543); Plan Apo 60x oil. ❸

Interchangeable filters: the filters for scanning and detection are easily interchangeable, so changing the filter to match the fluorescent dyes to be used is simple and quick. This design facilitates the use of the latest probes or dyes used in laboratories today.

Triple-band filter: a triple-band RGB Dichroic Mirror is supplied as standard, providing instant support for simultaneous 3-channel fluorescence imaging.

Easy attachment of a diascopic DIC module: superimposing confocal fluorescent images with the DIC morphology image provides greater depth information to the image.

Comprehensive time-lapse recording: supports all time-lapse observation needs, from second to hourly intervals.

Spatial analysis and ROI placement: measurements of intensity or size of the desired area is possible; Regions of Interest or ROIs can be placed at multiple points.

Universal scanner head: The scanner head can be easily mounted on either upright or inverted microscopes to support a wide range of applications.

C1 system configured with E600 microscope

C1 system configured with TE2000 microscope

Freedom in the use of lasers and dyes

Various laser types

The wider range of excitation light wavelengths facilitating the use of an increased number of fluorochromes in research has increased the demand for the number of lasers available on the market. The C1 comes with a series of lasers supporting almost every fluorescent dye used in research today. In addition, the use of these available laser lines can be mixed and matched to suit your excitation requirements. The unit employs the tried-and-true, stable 3-laser method for RGB excitation—the main wavelengths in use.

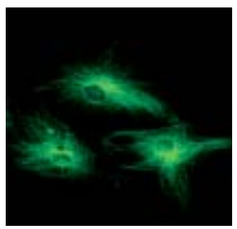
- Choose the 2- or 3-laser unit according to your needs.
- The excitation light from the laser is transmitted to the scan head via single-mode fiber, freeing up desk space, as the laser module can be installed in an adjacent location.

Wide choices of fluorochromes

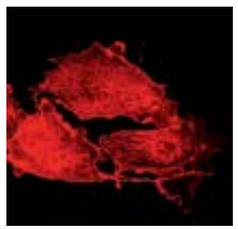
Besides the excitation lasers, a full selection of emission filters is available to support a wide range of fluorescence observation needs. The detection module is supplied with appropriate dichroic mirrors, allowing simultaneous 3-color observation of fluorescence images of specimens prepared for 3-color imaging .

Compatible Lasers, Wavelengths, and Dyes

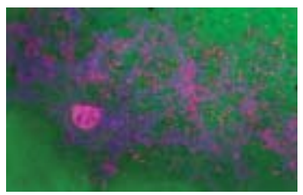
Argon laser (488nm)	FITC, Fluo-3, GFP, Cy-2, Alexa Fluor 488, BODIPY, Calcium Green, Acridine Orange, BCECF, Oregon Green
Helium neon laser (Green, 543nm)	TRITC (Rhodamine), Cy-3, PI, DsRed, Alexa 546, Alexa 568, BOBO-3, Calcium Orange, DiI, Mitotracker Orange, DS Red
Helium neon laser (Yellow, 594nm): Optional	Texas Red, DsRed, Alexa 568, Alexa 594, Calcium Crimson, Mitotracker Red
Helium neon laser (Red, 633nm)	Cy-5, Alexa 633, Alexa 647, Allophycocyanin, TOPRO-3



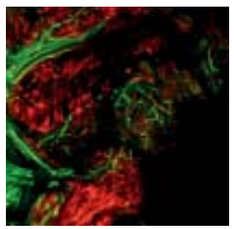
Tubulin of pulmonary artery endothelial cells: BODIPY-FL



Actin of pulmonary artery endothelial cells: Texas-Red labelled phalloidin

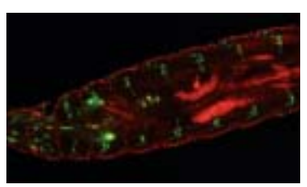


Biofilm: Cy3 (He-Ne 543), Cy5 (He-Ne 633), Reflection (Ar 488)

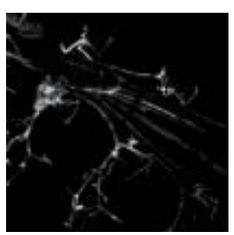


DiO (Ar 488), DiI (He-Ne 543)

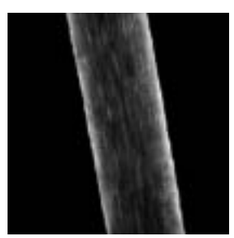
Nuclei of pulmonary artery endothelial cells: DAPI



Drosophila maggot: GFP (Ar 488), DsRed (He-Ne 543); Plan Apo 10x



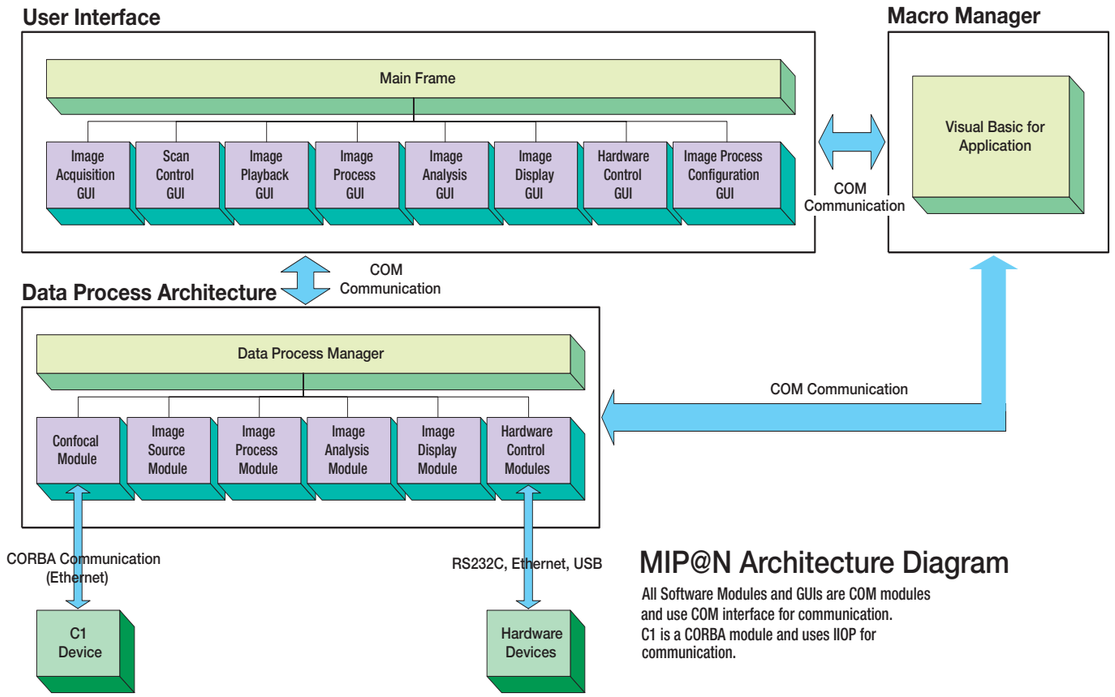
Drosophila embryo: GFP (Ar 488); Plan Apo 10x



Human hair: Autofluorescence (Ar 488); Plan Apo 60x

Operation is a breeze—and customizing just as simple

The C1 software is designed to run on Nikon's brand-new application platform, the MIP@N (Microscope Imaging Platform), and most of its program modules utilize COM (Component Object Model), a component software architecture from Microsoft®. Each module is an independent software component that can be freely combined or added to meet the needs of individual research tasks. The modular design also supports functional upgrades that you require when new techniques are developed. From this point on, Nikon will use MIP@N as the application platform and interface on which optical microscopes, confocal microscopes, digital cameras and other equipment will function.



MIP@N Architecture Diagram

All Software Modules and GUIs are COM modules and use COM interface for communication.
C1 is a CORBA module and uses IIOP for communication.

Intuitive GUI is easy to use

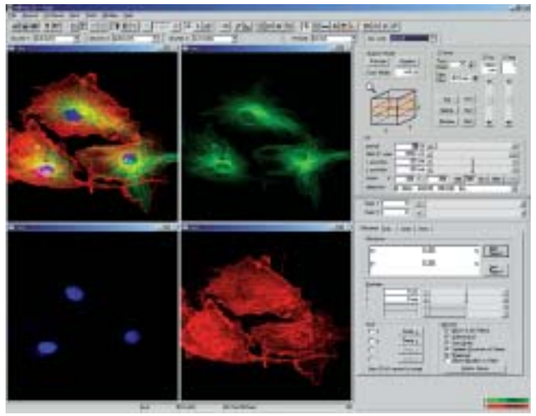
A thorough analysis of actual operations resulted in the development of a simple and intuitive GUI (Graphical User Interface). All necessary buttons are arranged in order of use so that even beginners will quickly grasp the operating process. From the initial handling, users may never even need to refer to the manual for operation.

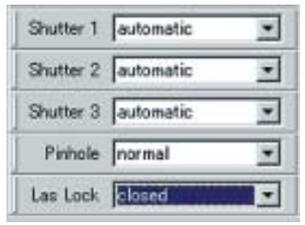
Image processing routines are easily selected

The various conditions under which an image was captured are recorded and displayed, information such as processing filter selection, ROI area setting, ratioing details etc., are graphically displayed in a directory tree in the Process View window for easy selection. Through this window, you can easily view and select these settings, or if necessary, you can add new selections by a simple cut-and-paste process.

Live images can be captured with ease

All settings and procedures required for live image capture—fundamentals in confocal microscopy—can be viewed in a single window, eliminating the need for the operator to switch between many windows. The operation panel gives you an at-a-glance picture of all important settings including scan mode, scan speed, pixel size, zoom/pan, PMT settings, pinhole, shutter, and color image look-up table. With the C1, scanning modes are expanded from 2D (XY, YZ, XZ), to 3D (XYZ, Xyt), and even further to 4-dimensional (XYZt) scans.





At-a-glance setting panel

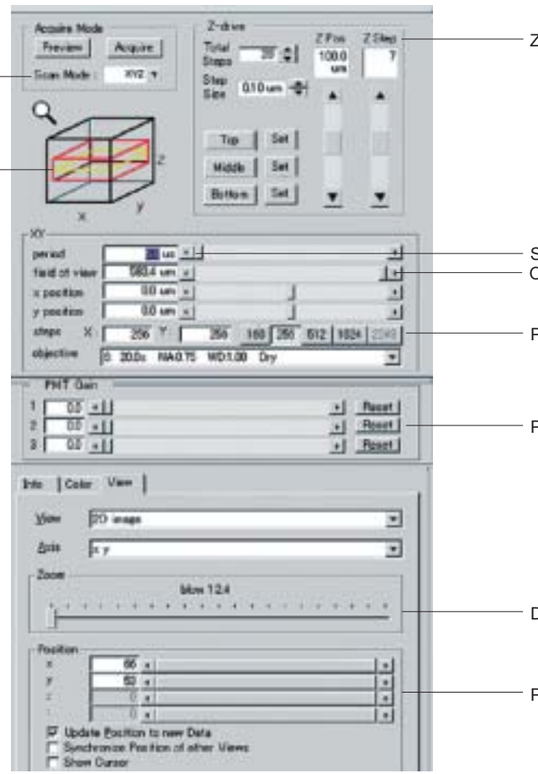
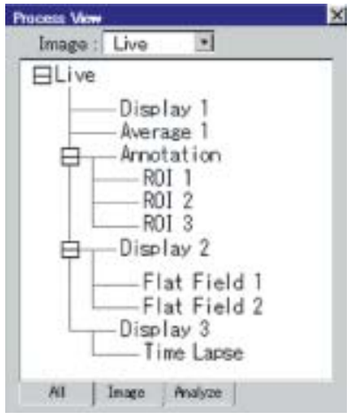


Image processing and display is a snap

Various steps you need to incorporate when processing an image, such as filter process setting, ROI area setting, and ratioing properties, are graphically indicated in a directory tree within the Process View window for simple operation. To process the image, select the desired process functions by pointing and clicking from the Process Module window to add them to the Process View window. You no longer need to search in other screens for the image processing routine you want to use.



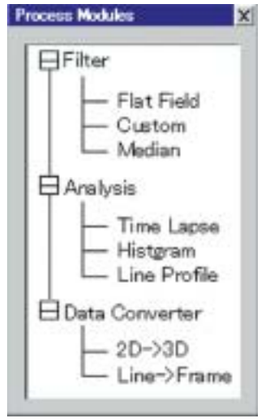
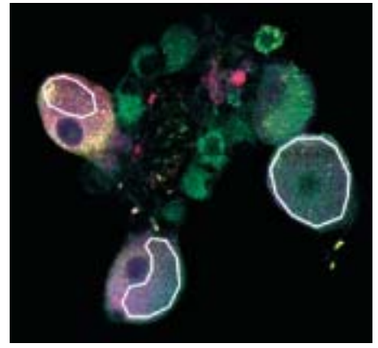


Image processing and analyses functions

A wide variety of processing filters are available including Median, Low-Pass, Kirch, Laplacian, Custom-Kernel, Square-Kernel, Round-Kernel. Averaging is possible in various ways such as averaging by specifying the number of frames, frame by frame, continuous, by specifying the image divisor or rolling average number. Image enhancing features enable correction of contrast, brightness, gamma, color balance, white balance, background, shading and other factors to optimize the scanned or captured image.

Annotation tool bar

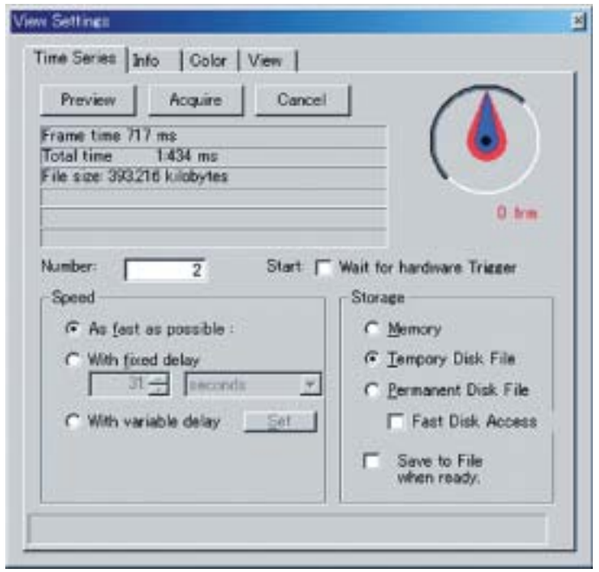
Setting of multiple regions of interest (ROI's) within the specimen is possible by selecting desired tools, so you can easily obtain detailed data on specific regions, such as size or the intensity versus the time course of the experiment.



Time-lapse recording is also a simple matter

All required procedures for time-lapse recording, including the setting of inter-cycle times, frame intervals, and the number of images to be captured can be provided in a single window for quick, easy operation. Image capture via sync-control from automated accessories or connected electrophysiology equipment is also possible.
In addition, a time stamp feature allows the operator to put accurate temporal ID stamps on the desired frames during observation, and to use these data marks to easily recognize the temporal basis of the marked images during playback.

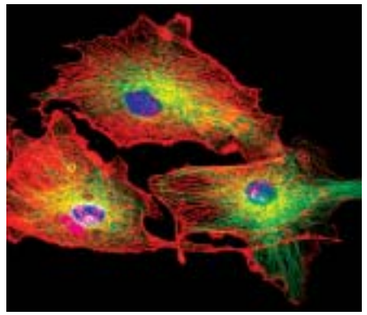
Last but not least, thanks to the C1's exceptional signal-to-noise ratio, the intensity of the excitation light can now be lowered much more than previously possible. This is a big advantage for living specimens. Photobleaching is dramatically reduced making the C1 confocal microscope system extremely useful for a broader range of applications.



Powerful results from advanced confocal imaging

Triple staining

Triple staining of pulmonary artery endothelial cells. Tubulin stained with BODIPY-FL (green), actin stained with Texas-Red labelled phalloidin (red), and nuclei stained with DAPI. Acquired with Plan Apo 60x oil.

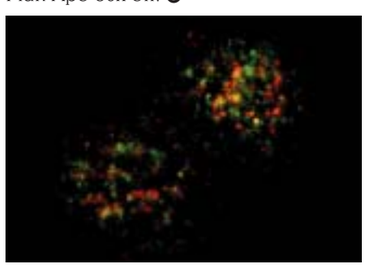


Fluorescence and DIC

Merged maximum projection of a 20µm stack through a developing drosophila maggot with a single plane scanned DIC image: TRITC (He-Ne 543); Plan Apo 10x

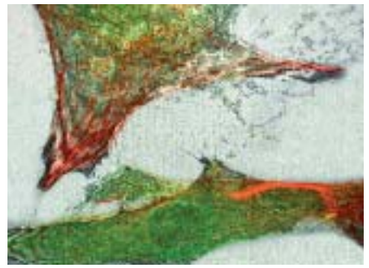
FRET (Fluorescence Resonance Energy Transfer)

FRET between FITC (donor) and Cy3 (acceptor); (Ar 488); Plan Apo 60x oil. ❺



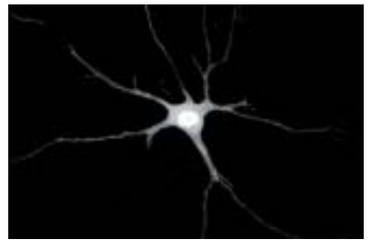
Cell motility

Merged interference reflection and confocal fluorescence image of fibroblasts growing out over a glass coverslip substrate: GFP (Ar 488), TRITC (He-Ne 543); Plan Apo 60x oil. ❻



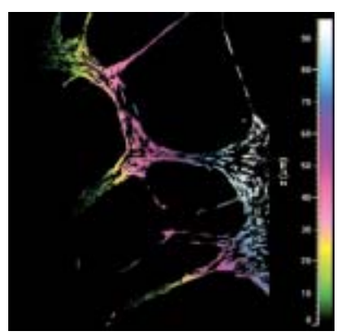
Deconvolution

3D blind deconvolution of a 40µm section illustrating YFP expressing motor neurons in mouse spinal cord; (He-Ne 543); Plan Apo 20x. ❼



Layer-by-layer quasi-coloring

Neuroglial cells stained with FITC. The image is presented in quasi-colors specified by layer to layer from top to bottom in the z-axis direction. Acquired with Plan Fluor 40x oil.

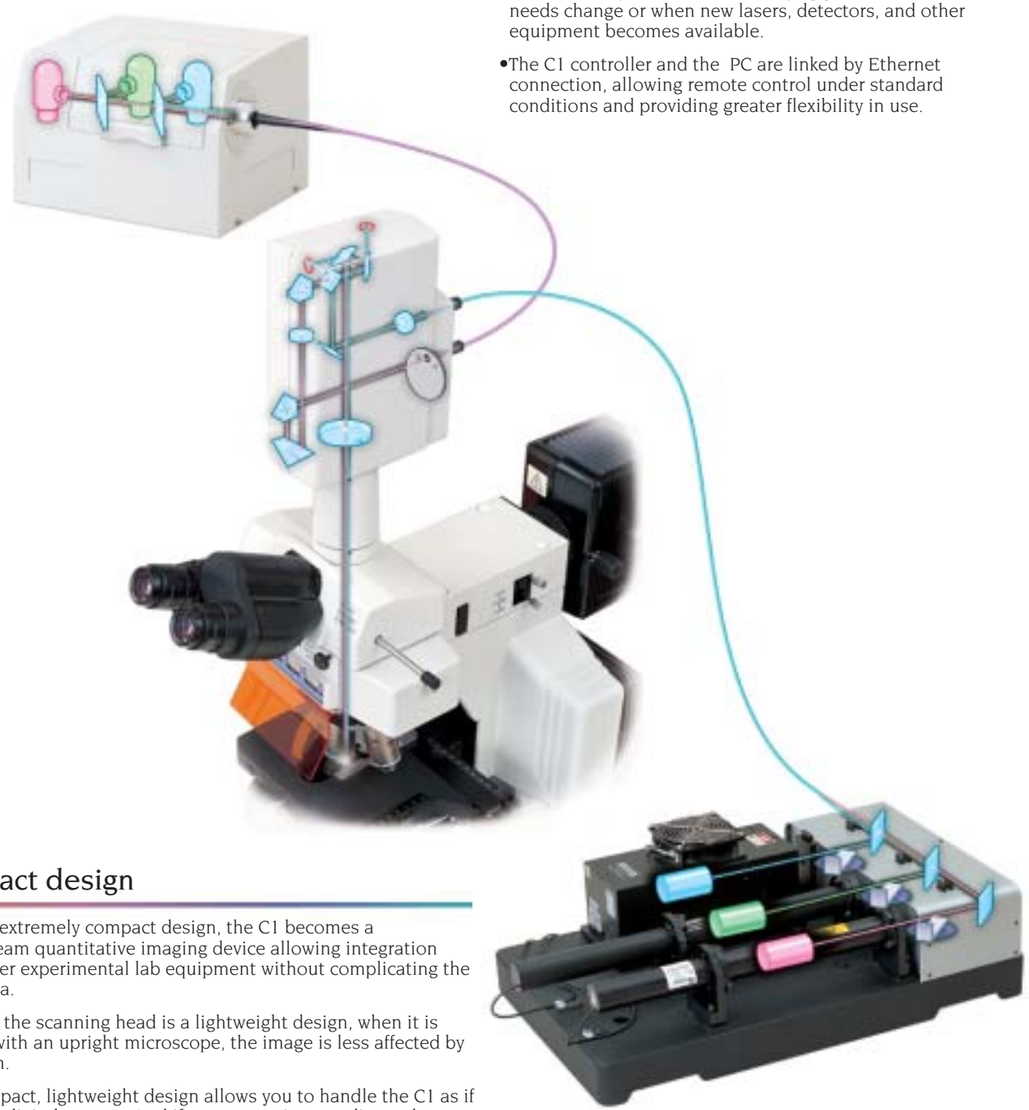


Compact and flexible

Flexible, modular design

All main components are modular including the laser box, scanner head, and detection module, facilitating simple expansion to meet diverse user needs and facilitate maintenance. The scanner head can be mounted on either upright or inverted microscopes to support a wide range of applications.

- Each of the C1's modular hardware units is pre-calibrated, greatly simplifying the setup. Just connect the modules you need, and you are ready for optimal image capture.
- Modules other than the scanner head do not need to be placed on the desk, providing the freedom to place other equipment where it is needed for your experiments.
- The flexible system allows for easy upgrades when research needs change or when new lasers, detectors, and other equipment becomes available.
- The C1 controller and the PC are linked by Ethernet connection, allowing remote control under standard conditions and providing greater flexibility in use.



Compact design

- With an extremely compact design, the C1 becomes a mainstream quantitative imaging device allowing integration with other experimental lab equipment without complicating the work area.
- Because the scanning head is a lightweight design, when it is loaded with an upright microscope, the image is less affected by vibration.
- The compact, lightweight design allows you to handle the C1 as if it were a digital camera. And if you use an intermediate tube, you can attach multiple cameras of various types.

A wealth of accessories

Z-focus module

This module, which can be retrofitted to the C1 system, features a minimum focusing increment of 100nm. You can freely set image capture environments such as XZ, YZ, XYZ, or XYZt in conjunction with the CI's original spatial (X, Y) and time (T) axes.

Diascopic DIC module

Consisting of a retrofitable separate modular, yet compact, transmitted light detector, this configuration leaves ample space around the microscope. The images obtained will have greater depth and co-localization image information by superimposing a fluorescence image over the high-resolution DIC image captured through laser scanning.

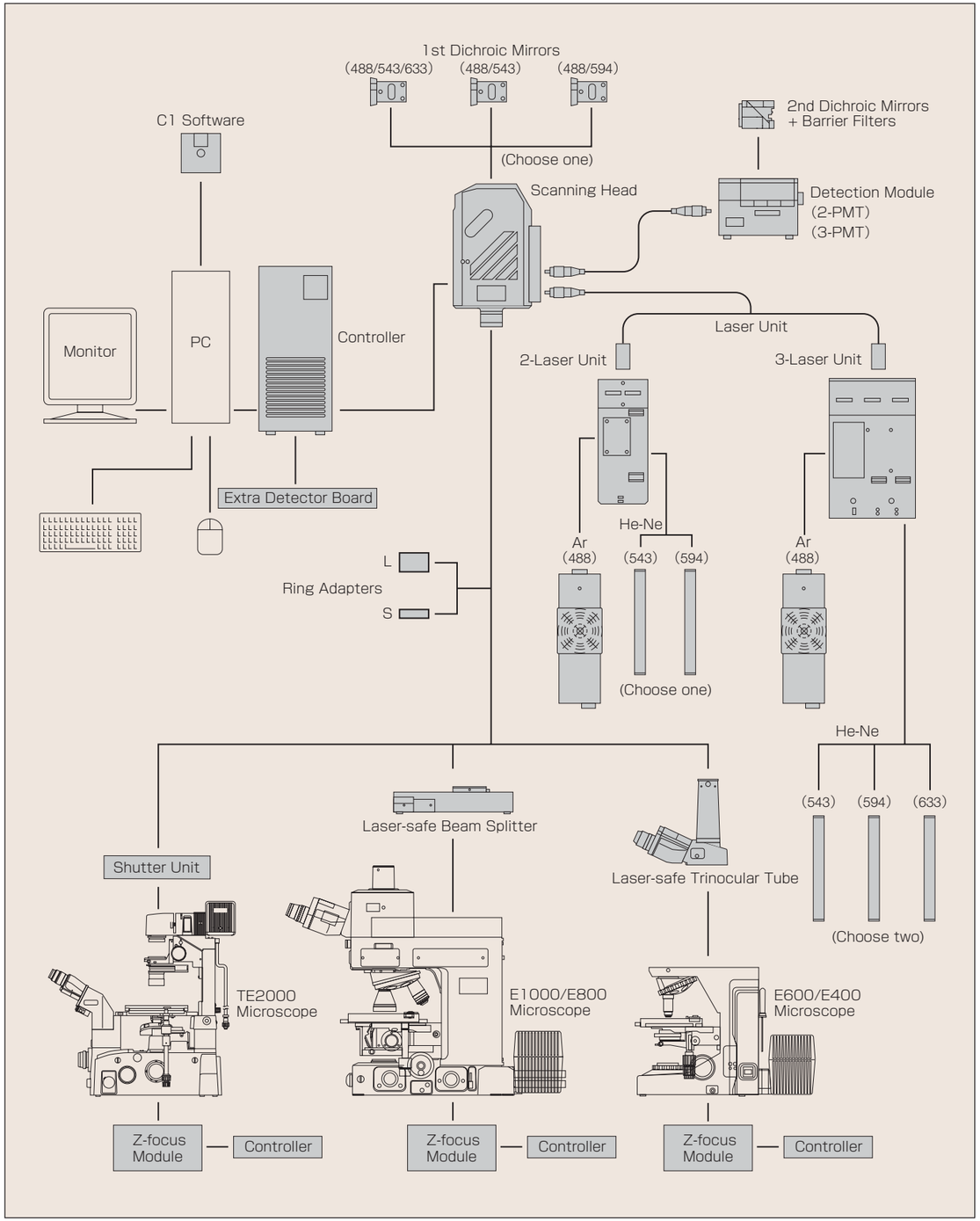
3-laser unit

The pictured 3-laser unit module is used for observing triple-stained specimens. It provides extremely stable and consistent, long laser life performance for this application. Because it has been designed for durability and longevity, this module will provide excellent results for many years.

3-PMT detection module

Like the standard 2-PMT unit, connection of the optional 3-PMT detector unit with the scanner head is established via a fiber cable, eliminating restrictions on where to locate it while being able to environmentally isolate it to prevent the deterioration of signals. Changing the filter sets to match the fluorescence dyes is quick and easy, while the use of interchangeable filters allows the ability to provide perfect wavelength tuning of new probes or dyes when they become available.

System diagram



Specifications

Laser unit	Laser type	Ar (488), G-HeNe (543), Y-HeNe (594), R-HeNe (633)
	Number of lasers mountable	Up to 3 lasers mountable
	Laser intensity control	Manual for each laser (4 steps)
	Laser shutter	Motorized shutter for each laser
Detection module	Channel	Standard: 2 fluorescence channels Maximum: 3 fluorescence channels + 1 transmission diascopic DIC channel
Pinhole	Variable	3 pinhole size steps + OPEN (motorized)
Dichroic mirror	1st DM 2nd DM 3rd DM	Interchangeable. For standard combinations, see the table below.
Scanning head	Scanning resolution	256 x 256—2048 x 2048 pixels
	Scanning speed	Standard: 1 sec. for 512 x 512 Line: 500L/sec.
	Scanning mode	2D: X-Y, X-Z, Y-Z 3D: X-Y-Z, X-Y-t 4D: X-Y-Z-t Special modes: Point, Band-scan, Area-scan
	Zoom	Continuously variable from 1X to 10X
	F.O.V.	Square inscribed in a ø18mm circle
Image bit depth		12 bits
Diascopic DIC detector (option)		Attach to the microscope main-body and lamphouse
Compatible microscopes	Upright type	Via dedicated laser-safe trinocular tube: E600, E400 Via dedicated laser-safe beam splitter: E1000, E800
	Inverted type	By adding dedicated laser-safe shutter: TE2000
	Fixed stage type	Via dedicated laser-safe trinocular tube: E600FN
Z-axis motor (option)	Control	Stepping motor
	Minimum readout	100nm
Compatible PC	PC	PC/AT compatible
	OS	Windows®2000
	Interface	Ethernet
Compatible objectives		CFI Plan Fluor series, CFI Plan Apo series
Dimensions (W x D x H), weight	Scanning head	Approx. 70 x 145 x 226 mm, 2.1kg
	Detector unit	Approx. 205 x 170 x 146 mm, 3kg
	Laser unit (2-laser)	Approx. 635 x 260 x 139 mm, 20kg (excluding laser)
	Controller	Approx. 200 x 430 x 380 mm, 10kg

Combination Examples of Lasers and Filters According to Dye

Dual Stain

B excitation	G excitation	Laser 1	Laser 2	Filter configuration
FITC or **Alexa 488**	**TMR** or **Cy-3**	Ar (488)	G-HeNe (543)	1st DM: 488/543 2nd DM: 530 Em filter: 515/30, 570LP
FITC or **Alexa 488**	**Texas Red** or **Alexa 594**	Ar (488)	Y-HeNe (594)	1st DM: 488/594 2nd DM: 565 Em filter: 530/50, 610LP

Triple Stain

B excitation	G excitation	R excitation	Laser 1	Laser 2	Laser 3	Filter configuration
FITC or **Alexa 488**	**TMR** or **Cy-3** or **Alexa 546**	**Cy-5**	Ar (488)	G-HeNe (543)	R-HeNe (633)	1st DM: 488/543/633 2nd DM: 530 3rd DM: 625 Em filter: 515/30, 585/40, 665LP

The following specimens used in this brochure are courtesy of:
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Specifications and equipment are subject to change without any notice or obligation on the part of the manufacturer. January 2002.

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

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