

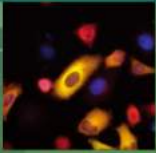
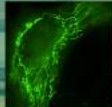


Imaging Software  
**NIS-Elements**  
Advanced Solutions for your Imaging World

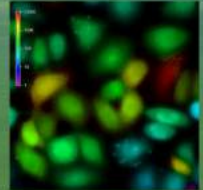
**N**  
Nikon

**I**  
Imaging

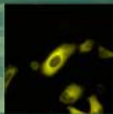
**S**  
Software



**Ar**



**Br**



**D**

# Nikon offers total software solution covering image capture,

NIS-Elements is an integrated software imaging platform developed by Nikon which delivers comprehensive microscope control, image capture, documentation, image analysis and data management. NIS-Elements handles multidimensional imaging tasks flawlessly with support for capture, display, peripheral device control, and analysis & data management of images of up to six dimensions. The system also contributes to experiment efficiency with an intuitive image analysis feature set and database building capabilities developed to handle archiving and management of large numbers of multidimensional image files. Unified control of the entire imaging system offers significant benefits to microscopists for cutting-edge research, such as live cell imaging



The NIS-Elements suite is available in three packages scaled to address specific application requirements.



**Ar**

NIS-Elements AR is optimized for advanced research applications, featuring fully automated acquisition and device control through full 6D (X, Y, Z, Lambda (Wavelength), Time, Multipoint) image acquisition and a wide range of image analysis.

**Br**

NIS-Elements BR is suited for standard research applications, photodocumentation of fluorescent samples and image analysis including intensity and counting measurements. It features acquisition and device control through 4D (up to four dimensions can be selected from X, Y, Z, Lambda (Wavelength), Time, Multipoint) acquisition.

**D**

NIS-Elements D supports color documentation requirements in bioresearch, clinical and industrial applications, with basic measuring and reporting capabilities.

# archiving, and analysis

## Why NIS-Elements?

As a leading microscope manufacturer, Nikon realizes the importance of providing its customers with system-based solutions to free them to focus on their projects and research and not on the complexities of the microscope. Never before has a software package offered such comprehensive control of microscope systems, image acquisition, image analysis and data management.

### Total Imaging Solution

In designing and bringing to market the most technologically advanced optical systems, Nikon has worked very hard to provide a "total imaging solution" that meets the ever-evolving demands of the microscope user.



### •Highest Quality Optical Performance

The world-renowned Nikon CFI60 infinity optical system effectively set a new standard for optical quality by providing longer working distances, higher numerical apertures, and the widest magnification range and documentation field sizes.

As a leader in digital imaging technology, Nikon recognized the importance of adapting its optics to optimize the digital image. Nikon's new objectives and accessories are specifically engineered for digital imaging.

Because what you see depends greatly on the quality of your microscope, we strive to power our microscope systems with optical technologies that are nothing but state-of-the-art.

### •Diverse Line of Powerful Digital Cameras

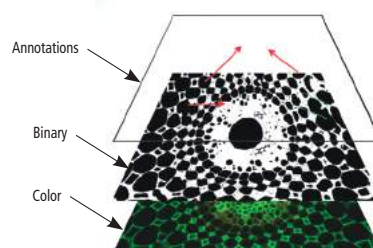
Image capture has become a high priority in microscopy and the demand for products that deliver high quality and versatile functionality has grown considerably in recent years. In accordance, Nikon offers a full line of digital cameras, addressing the varied needs of microscopists in multiple disciplines. Each Nikon digital camera is designed to work seamlessly with Nikon microscopes, peripherals, and software. With Nikon Digital Sight (DS) series cameras, even novice users can take beautiful and accurate microscopic images. For the advanced researcher, hiresolution image capture and versatile camera control is fast and simple. Together with Nikon's new software solutions, image processing and analysis have reached new levels of ease-of-use and sophistication.

### •Intelligent Software Solutions

Designed to serve the needs of advanced bioresearch, clinical, industrial and documentation professionals, NIS-Elements provides a totally integrated solution for users of Nikon and other manufacturers' accessories by delivering automated intelligence to microscopes, cameras, and peripheral components. The software optimizes the imaging process and workflow and provides the critical element of information management for system based microscopy.

### Multi-layer Document Structure

NIS-Elements uses a sophisticated image documentation structure making it possible to achieve non-destructive archiving of image data including annotation (arrows, lines, text notes), measurement data, binary data for storing results of threshold or classification processes, and meta-data information for recording acquisition and device conditions at the time of image acquisition.



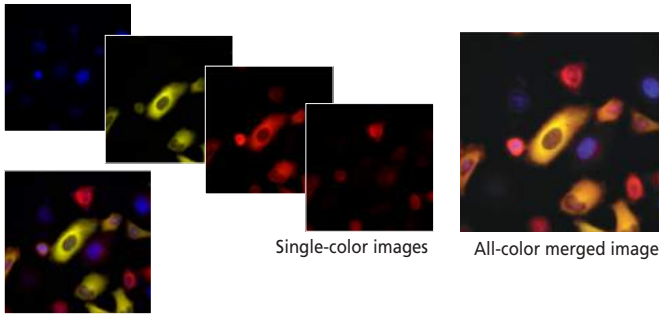
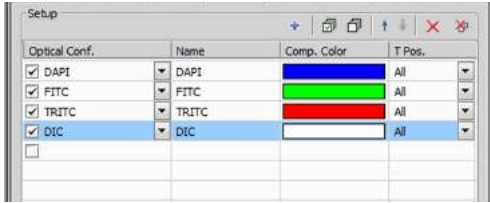
# Image Acquisition

NIS-Elements offers the most suitable image acquisition for various applications with the integrated control of the camera, motorized microscope and peripheral devices.

## Multichannel (multi color)



NIS-Elements can acquire full bit depth multi-color images, combining multiple fluorescence wavelengths and different illumination methods (DIC, phase contrast etc.), while offering independently scalable channels.



Single-color images

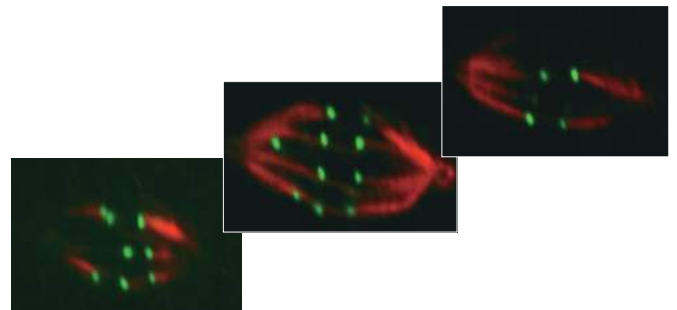
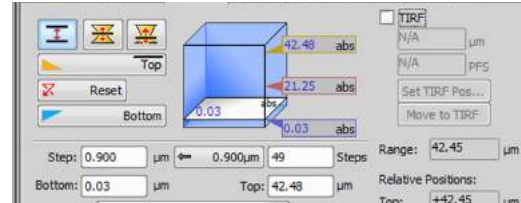
All-color merged image

Specified-color merged image

## Z-series



Through motorized focus control, NIS-Elements reconstructs and renders 3D images from multiple Z-axis planes.

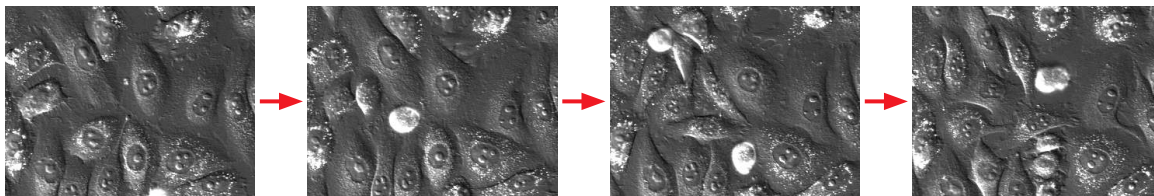
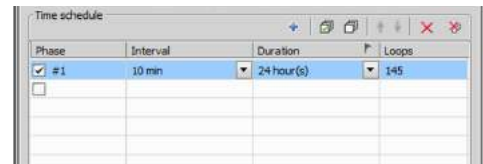


## Time Lapse



Time lapse imaging in NIS-Elements is easily configurable simply by setting the time interval and duration of capture.

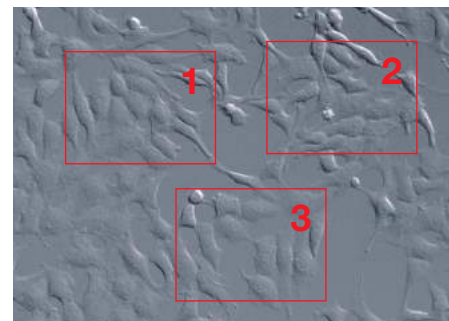
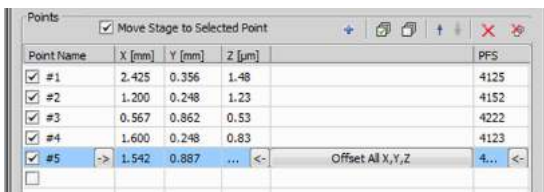
The Perfect Focus System of the motorized inverted microscope Ti-E enables high-accuracy image capture without focus drifting even during extended time time-lapse experiments.



## Multipoint Experiments



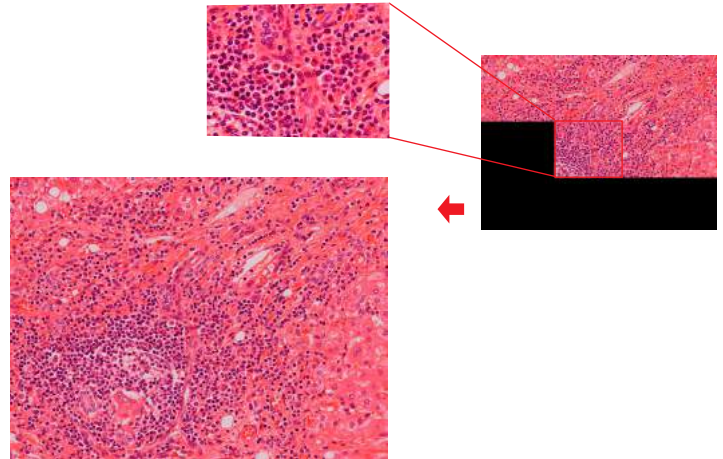
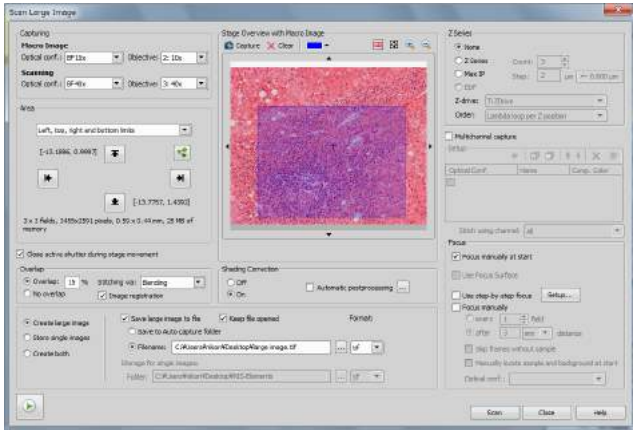
NIS-Elements' motorized stage control offers automated travel to multiple stage points of the sample of a multi-well plate or dish. Stage points are memorized and can be saved and loaded for future imaging sessions.



## Image Stitching (Large image)

Ar Br D

Large Image Acquisition generates a single high-magnification wide field-of-view image by automatically stitching multiple adjacent frames from a multipoint acquisition using a motorized stage or from multiple single images captured from a previous session. Users can easily select image acquisition ranges and areas from low magnification images.

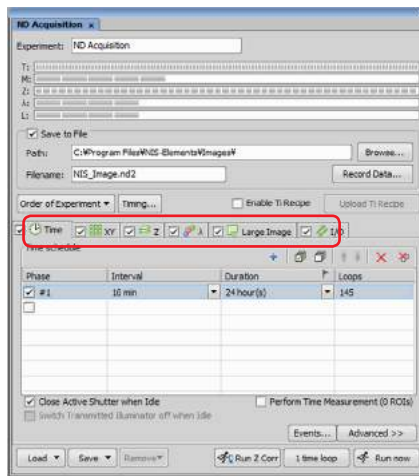


## Multidimensional imaging

### ND Acquisition

NIS-Elements captures images in a combination of multiple dimensions such as Time-Lapse, Multichannel, Z-series, and Multipoint. It is also possible to create and manage the acquisition of a multi-dimensional dataset with a thirty-minute time lapse of two wavelengths and a Z series across each well of a multi-well plate.

Option ( Ar Br )

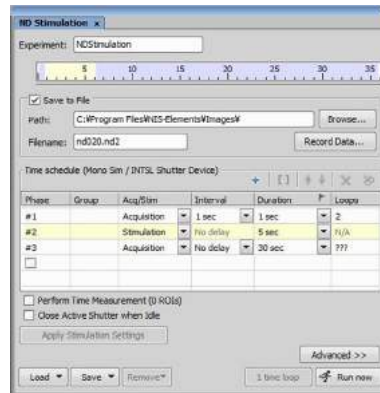


\* Available dimensions vary depending on the package.

### ND Stimulation

Option ( Ar )

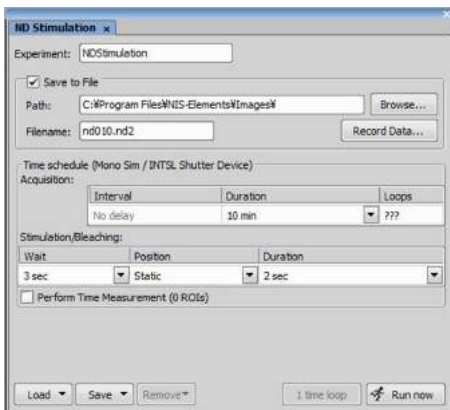
NIS-Elements controls photo stimulation and image acquisition.



### ND Simultaneous Stimulation

NIS-Elements enables image acquisition during photo stimulation.

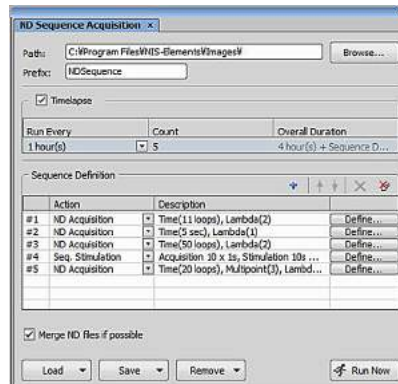
Option ( Ar )



### ND Sequential Acquisition

NIS-Elements allows various sequential imaging experiments to be combined with other functions, such as simultaneous photo stimulation and imaging, or multidimensional acquisition.

Option ( Ar )



## Optical Configuration

Presets or 'Optical Configurations' can be saved for each observation method such as FITC fluorescence and DIC imaging, memorizing the settings of the microscope, camera and peripheral devices. The optical configurations are created through a one click set up and are displayed as icons in the tool bar for easy access and use.

The image illustrates the process of saving optical configurations. On the left, a camera is shown next to its 'DS-Q12 Settings' dialog box, which includes options for Resolution (Fast (Focus) 14bit 536x536, Quality (Capture) 14bit 1608x1608), Exposure (Auto Exposure 30 ms, Analog Gain 1.0x, Live Acceleration: 1.0x), and Commands. In the center, a microscope is shown with a plus sign. To its right is the 'Microscope Setting' dialog box, which includes various parameters like Magnification (10x, 20x, 40x, 60x, 100x, 150x), Light Path (S30, S35, S40, S45, S50, S55), Drive (Camera, Accuracy, Offset, Defocus Plane), Stages (Coarse, Fine, Duty File), Shutters (EFTS, OSA), Filters (Turn11), Condenser (Zoom, L10210, L109), and Scope (Use Scope: OSC.Park1, OSC1). On the far right, a software toolbar is shown with icons for View, Devices, Window, Applications, and a set of optical configuration icons: BF, DIC, DAPI, FITC, TRITC. A red box highlights these icons with the text 'Memorize settings of the camera and microscope'.

## Movie Capture, Fast Image Capture

NIS-Elements has several options to observe and capture a sample's change and fast movement.

### Fast Time Lapse

Fast Time Lapse is designed for ultra high-speed cameras. The hard disk drive can be used together with PC memory to enable a longer acquisition time.

The 'Fast Time Lapse' dialog box shows options for saving to file (Path: C:\Program Files\NIS-Elements\Images\, Filename: experiment.n22), Number of Frames (500), Start Acquisition (Manually, TTL signal, Stimulate), and Sample Storing (Memory Only, Memory and Disk). It includes 'Apply', 'Close', and 'Run now' buttons.

### RAM Capture

RAM Capture allows for acquisition at the fastest possible rate of the camera. A RAM buffer is utilized to enable capture and retrieve a high speed time lapse, which aids in the capture of fleeting events such as calcium sparks, motility and translocation.

The 'Settings' dialog box shows 'Frames buffered before' and 'Frames buffered after' both set to 1000. A note states: 'Current settings will require buffer with 1,000 frames. The size of used memory will be 687 MB.' It includes a 'Settings...' button.

### AVI Live-Stream Capture

AVI Acquisition automatically captures live data into an easily exportable and viewable AVI format.

The 'AVI Acquisition' dialog box shows 'Record' and 'Stop' buttons, 'Live' mode set to 'Fast', 'Rate' set to 'Max Fps', and 'Duration' set to 'Infinite'. It indicates 'Ready 275.9 GB free on drive C:'.

## Time Saving Acquisition by Hardware Optimization

### Triggered Acquisition

Triggering external devices directly from the camera enables synchronized control of various devices such as the laser unit without passing through the PC. This allows for the fastest performance of the system components for multi-wavelength excitation in TIRF observation.

The 'Triggered Acquisition' dialog box shows Camera (DLU-897) and Multi-excitation Device (LU5A). It includes a table for line settings:

#	Line	Power	Exposure	Focus Offset
1	Line: 488 nm	10 %	33 ms	0 μm
2	Line: 561 nm	15 %	33 ms	0.8 μm
3	Line: 640 nm	10 %	33 ms	0.2 μm

It also has checkboxes for 'Use different exposures' and 'Enable Triggering'.

### Ti-recipe

This function enables the HUB-A controller of the motorized inverted microscope Ti-E to control both image acquisition and change of fluorescent filter, motorized stage and fluorescent shutter by directly connecting the camera and a HUB-A controller without passing through a PC. As a result of optimizing the communication times of all connected devices, acquisition times for multi-dimensional datasets are greatly reduced.

\*Some cameras are not compatible with this function. For more information, please contact Nikon or its authorized representative.

A checkbox labeled 'Camera exposure signal is connected to DSC port for faster experiments:' with a dropdown menu showing 'DSC1'.

## NI-DAQ Control

Option ( Ar Br D )

TTL and analog signal input/output with NI-DAQ Control enables experiments using various triggerable devices.

Device triggering minimizes imaging time lag when used to control a Piezo Z device, shutter and wavelength switching, as it performs at the faster rate of the device without software communication delay.

The calibration of analog signal output from devices to such as temperature enables data to be measured and observed during experiments.

The 'NI-DAQ Configuration' dialog box shows 'Available devices' (TTL Input, TTL Output, Analog Input, Analog Output, Calibrated Analog Input, Calibrated Analog Output, RealTime TTL Input, Shutter, Piezo Z, Piezo XY, Piezo XYZ, Illumination Device, HW Box, Switcher, Filter Wheel, Exposure Signal TTL In, Light) and 'Installed devices' (Piezo Z (name: Piezo Z), Analog Input (name: Dev1/ai0), RealTime TTL Input (name: Dev1/port0/line0), Shutter (name: Shutter), Illumination Device (AOTF), Illumination Device Shutter (AOTF), Light (Light1)). It includes 'Add -->' and '<-- Remove' buttons. At the bottom, it shows 'Free resources: Analog Input: 15, Analog Output: 37, TTL I/O: 34' and 'Triggering' options for PCI-6221, PCI-6713, and PCI-6723.

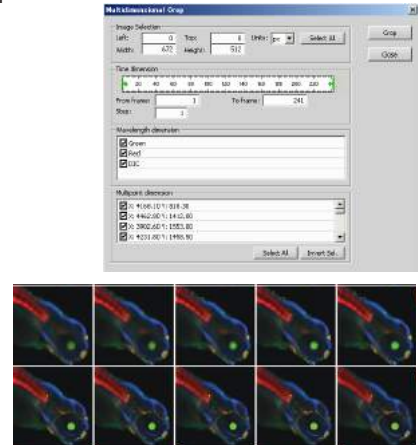
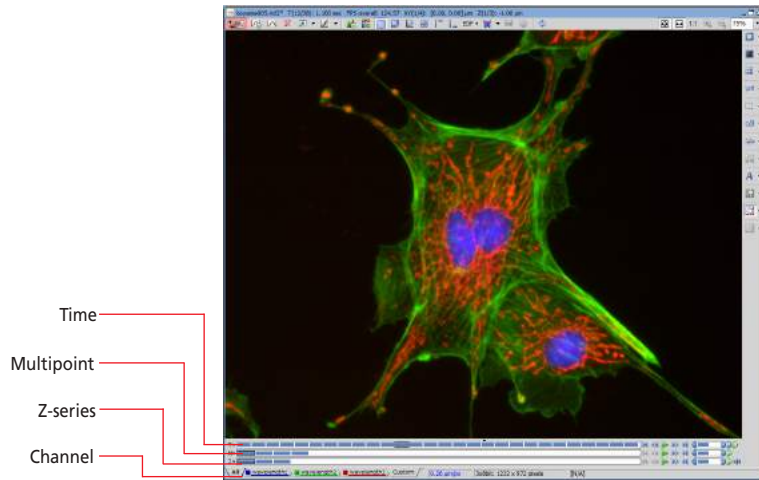
# Display and Data Processing

Various methods are available for displaying and processing captured images and datasets.

## Multi-dimensional Image Display

Ar Br

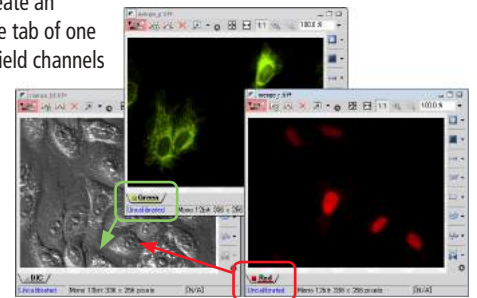
NIS-Elements displays time lapse, multi-channel, multiple X, Y, Z positions in an intuitive layout, which allows for automatic playback and the ability to select subsections of the data to be saved as a new file.



## Merge Channels

Ar Br D

Multiple single channel images (ex., two from three-channel acquisition images) can be merged together to create an overlay of full depth separately scalable images. With AR and BR, images can be merged by simply dragging the tab of one image onto another image. With D, images are merged by selecting each image for red, blue, green and brightfield channels



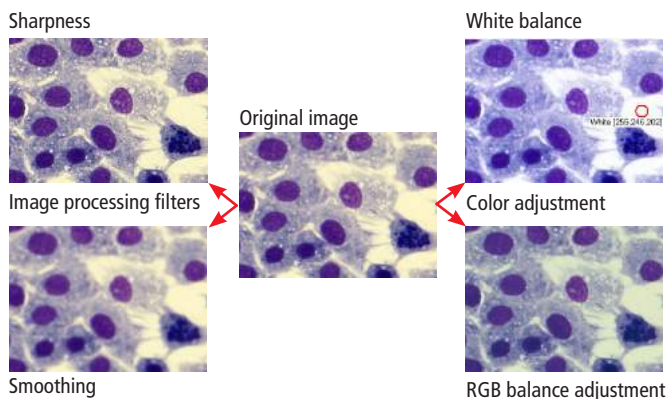
## Image Processing

### Image Filtering, Color Adjustment

Ar Br D

\*Usable functions vary depending on the package.

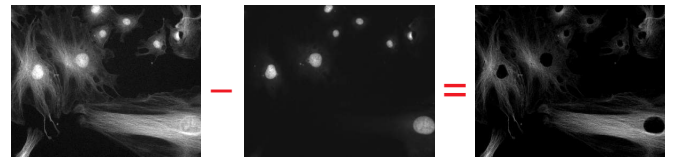
With NIS-Elements image processing tools, it is possible to modify image display and feature extraction using various filters for, for example, sharpness, smoothing and detection. White balance and RGB/HIS balance adjustment are additional available options.



### Arithmetic operation (Image arithmetic)

Ar Br Option ( D )

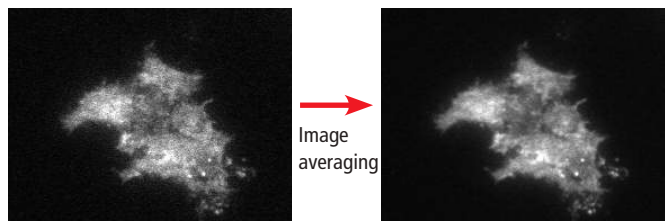
NIS-Elements enables arithmetic operations such as addition, subtraction, multiplication and division on an image or between multiple images. Arithmetic operation between multiple images is also possible.



### Arithmetic operation (Image averaging)

Ar

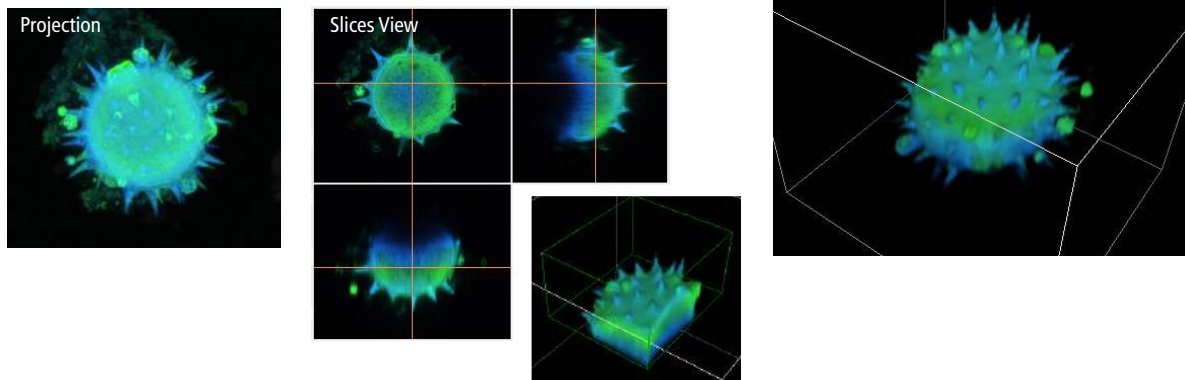
NIS-Elements reduces the noise of an image by averaging multiple sequential images such as time-lapse images. Rolling averaging that does not reduce frame rate is available as well.



## Z-Series Image Display

\* Volume view and slice view are only possible with AR and BR. Ar Br D

Z-series images can be displayed in various formats such as max. and min. projections, X-Z axis and Y-Z axis cross-sectional slice view and 3D volume view. Rotatable 3D volume rendered views from 3D datasets are easily converted to an AVI or MOV format for file sharing and export.

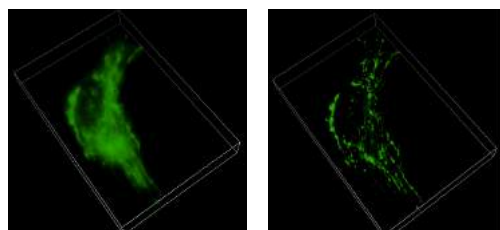


## Deconvolution

Option ( Ar )

### 3D Deconvolution

Haze and blur of the acquired fluorescence image can be eliminated. By reassigning out-of-focus intensities back to the spatial locations to where they originated, the intensity of the image is kept and allows for quantitative analysis. Algorithms for wide-field fluorescence, point-scanning confocal and spinning-disk confocal images are available.

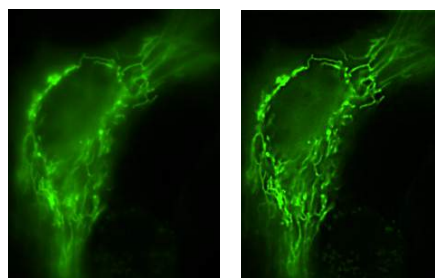


Before deconvolution

After deconvolution

### 2D Deconvolution

The 2D deconvolution module can be applied to a live image or an already acquired dataset. The module also allows the elimination of out-of-focus blur from live images and multidimensional images.



Before deconvolution

After deconvolution

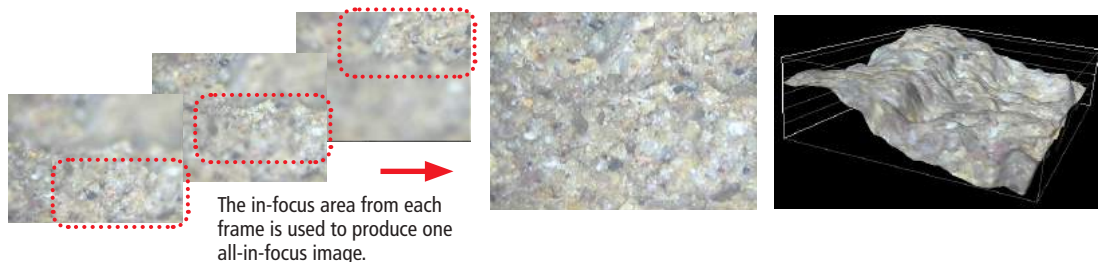
## Extended Depth of Focus (EDF)

Option ( Ar Br D )



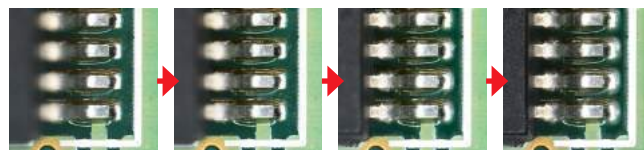
### With motorized focusing

The EDF function selects the in-focus area from multiple Z-stack images, and produces one all-in-focus image. The composite image can be viewed and rotated as a virtual 3D image, as it contains Z-axis information.



### With manual focusing

An all-in-focus image is created in real-time in synchronization with the rotation of the focus knob. While the focus is manually adjusted, the in-focus areas of the image at different depths are successively captured and combined for the EDF image.

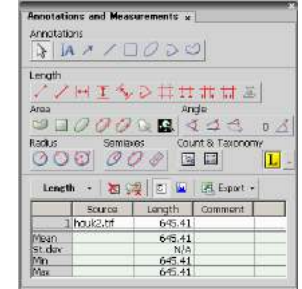
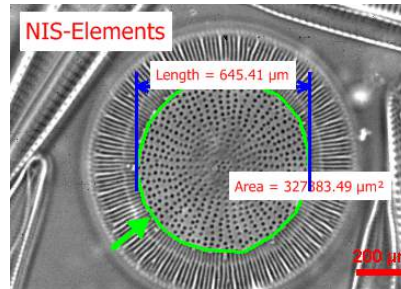




# Measurement and Analysis

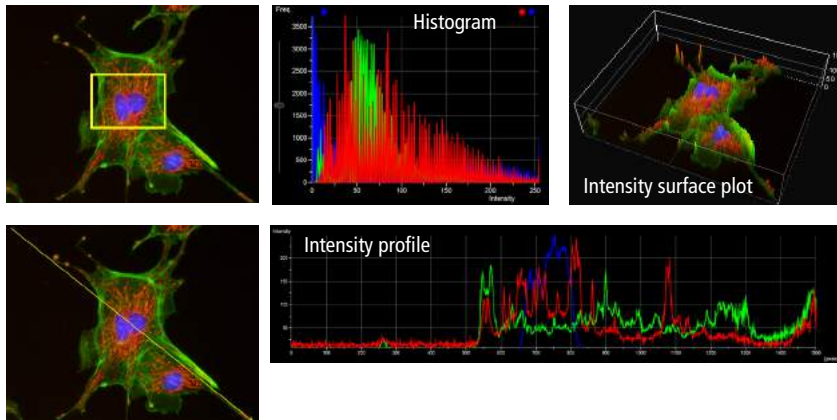
## Manual Measurement (Interactive Measurement) and Image Annotation

Interactive Measurement allows easy measurement of length and area by drawing lines or an object directly on the image. The results can be attached to the image, and also exported as text or to an Excel spreadsheet. Annotations such as arrows, circles, squares, text are also available display options.



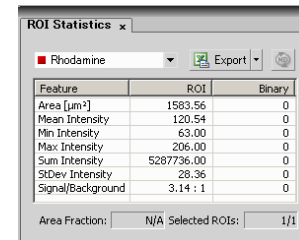
## Histogram/Intensity Line Profile/Intensity Surface Plot

Histogram measurement measures the intensity distribution of pixels across the whole image or a defined region. An intensity line profile measurement shows the intensity distribution on a defined line. The Intensity Surface plot shows the intensity distribution of an image with the height of the z-axis line.



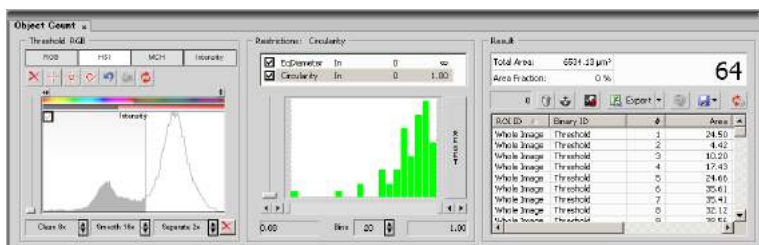
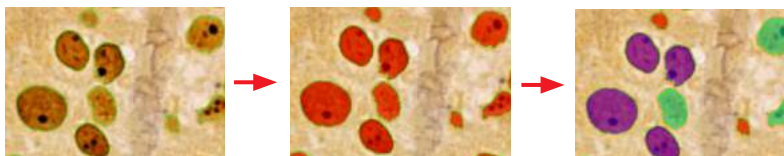
## ROI Statistics

\*Usable functions vary depending on the package. Common pixel measurements such as area, maximum or minimum intensity are possible with the user defined ROI (Region Of Interest). ROI or multiple ROIs statistic results for a single image or a multi-dimensional dataset are displayed and easily exported as text or an excel file.



## Auto Measurement (Object Counting) Option ( D )

Auto measurement measures the number or area of objects which are extracted from images by the creation of a binary layer through thresholding using RGB/HIS or intensity values. The results can be listed or exported as text or an excel file. It is possible to save and reuse thresholding parameters.



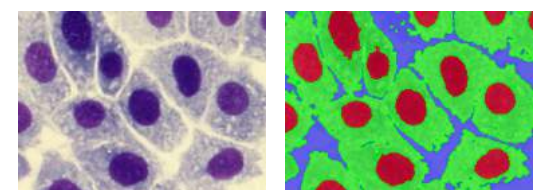
## Classifier

### Object Classifier

Object classifier uses objects identified by thresholding along with additional features such as shape factors, and other statistical methods including nearest neighbor and neural networks for classifying objects into multiple categories. It is also possible to teach the module based on interactive 'picking' of image pixels.

### Pixel Classifier

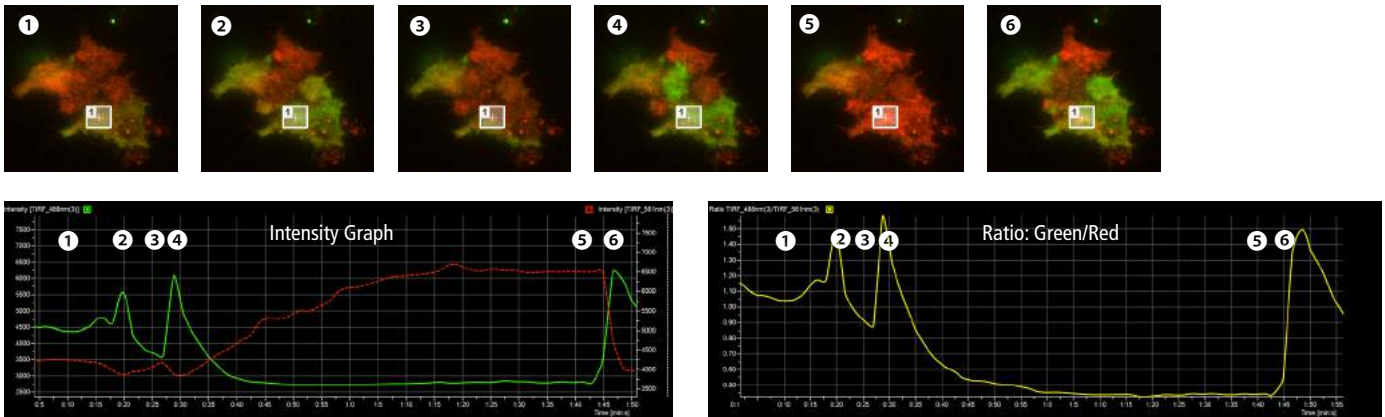
This function classifies each pixel in the image with RGB/HIS and intensity across the whole image. Results are reported in percentage and it possible to save and reuse parameters across a large sample of images. Multiple binary layers are also displayed with multiple colors on the image and are available with other analysis tools within the software package.



## Time (Intensity) Measurement

Option (Ar)

Time measurement creates a graph of sequential intensity changes while time-lapse imaging or from captured time-lapse images. Ratio view function\* allows the measurement of the ratio of two wavelengths across multiple ROIs and shows the ratio value by pixel. Numeric data and graph images are exportable and the measurements on the graph are available as well. (\* Only with AR)

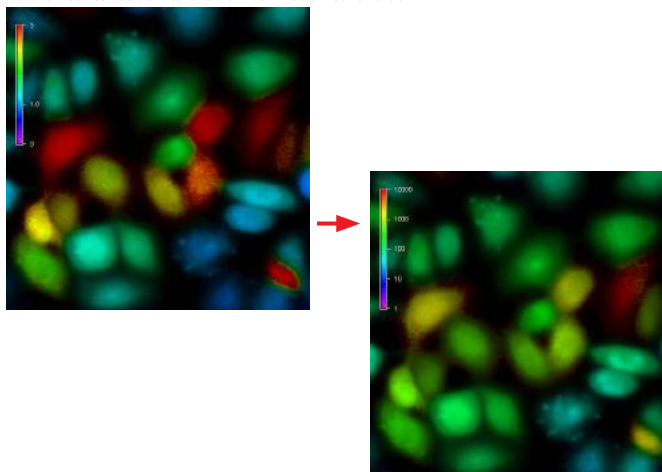


## Calcium & FRET

Option (Ar)

Ca<sup>2+</sup> ion concentration calibration of the ratiometric fluorochrome Fura2, for example, is available using an easily configurable wizard. Corrected FRET image and FRET efficiency, reported in percentage is also available using three filter sets (three types of excitation-fluorescent combination: "Donor - Donor," "Acceptor - Acceptor" and "Donor - Acceptor") and two bleed-through factors.

Ca<sup>2+</sup> ion concentration calibration from ratiometric value



FRET analysis

Create FRET Image

Donor - acceptor (Da): Da (FRET sample).tif    Donor - donor (Dd): Dd (FRET sample).tif    Acceptor - acceptor (Aa): Aa (FRET sample).tif    Acceptor - donor (Ad): <None>

Thumbnail    Thumbnail    Thumbnail    Thumbnail

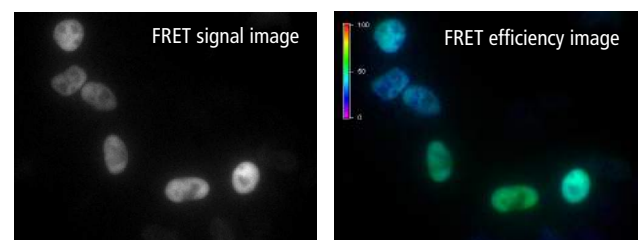
FRET Method

Method: Sensitized emission

Acceptor in FRET (CoA): 0.19    Donor in Acceptor: 1

Donor in FRET (CoB): 0.55    Acceptor in Donor: 1

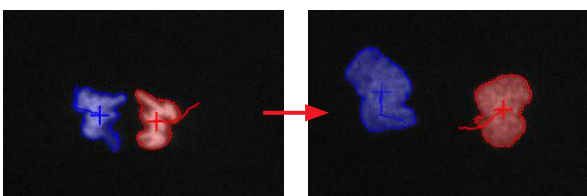
Buttons: Calibrate.., OK, Load, Cancel, Save, Help, Remove



## Object Tracking

Option (Ar)

2D tracking of an object utilizes the threshold of objects over time and produces measurements such as velocity, acceleration, and distance from a specified origin. The tracking module offers both automated tracking and manual tracking methods.



Index	Time [s]	Position X [µm]	Position Y [µm]	Position Z [µm]	Distance [µm]	Velocity [µm/s]	Acceleration [µm/s²]	Path Length [µm]
1	0.000000	32.980999	25.647466	0.000000	0.000000	N/A	N/A	0.000000
2	60.000000	33.054195	25.598298	0.000000	0.039640	0.001482	N/A	0.039640
3	120.000000	33.228145	25.628420	0.000000	0.176788	0.002946	0.000024	0.265728
4	180.000000	33.243192	25.698851	0.000000	0.124026	0.002151	-0.000013	0.394762
5	240.000000	33.678441	25.746813	0.000000	0.247395	0.004646	0.000032	0.637548
6	300.000000	33.593850	25.772921	0.000000	0.032265	0.000504	-0.000359	0.667813
7	360.000000	33.858804	25.827052	0.000000	0.109126	0.001819	0.000022	0.776559
8	420.000000	33.795225	25.769797	0.000000	0.116727	0.001945	0.000002	0.885666
9	480.000000	33.917277	25.762958	0.000000	0.127637	0.002127	0.000003	1.021303
10	540.000000	33.971434	25.745410	0.000000	0.062857	0.001359	-0.000017	1.085660
11	600.000000	33.865979	25.692417	0.000000	0.144598	0.002459	0.000022	1.221158
12	660.000000	34.190742	25.544193	0.000000	0.214804	0.003580	0.000020	1.444603
13	720.000000	34.403611	25.577152	0.000000	0.221536	0.003692	0.000002	1.667538
14	780.000000	34.521361	25.593659	0.000000	0.159235	0.002254	-0.000024	1.897713
15	840.000000	34.692465	25.624591	0.000000	0.211248	0.003521	0.000021	2.014021
16	900.000000	34.756459	25.641291	0.000000	0.065137	0.001102	-0.000040	2.090158

Graph \Data | Path Length: 28.262714 [µm]    Ave Velocity: 0.001818 [µm/s]    Ave Acceleration: 0.000000 [µm/s²]

## GA (General Analysis)

Option ( )

General Analysis streamlines the setup of image analysis combining functions, such as image preprocessing and thresholding, processing of binary images and features, and measurement settings.

- GA creates a new measurement area by combining multiple binary layers, and creates a new measurement parameter by applying these custom measurement settings.
- Each setting can be stored as a recipe that can be rerun for routine analysis under the same conditions across multiple datasets.

The screenshot shows the 'General Analysis' dialog box with several sections highlighted by red boxes and labels:

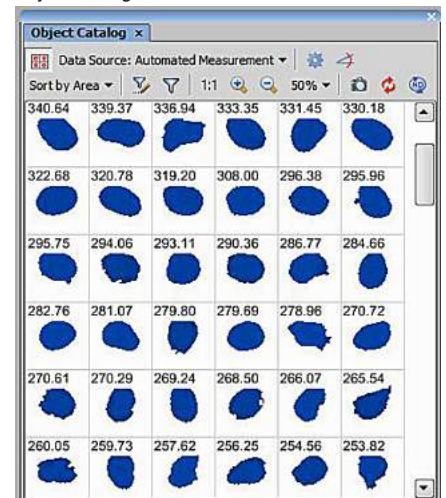
- Combine binary layers**: Points to the 'Analysis Palette' icons.
- Create new measurement lists**: Points to the 'Analysis Steps' dropdown.
- Setup of multiple analysis tasks**: Points to the overall configuration area.
- Image preprocessing**: Points to the 'Preprocessing' section, including 'Remove Dark Background' and 'Median'.
- Image thresholding**: Points to the 'Threshold' section, including the threshold value (120) and 'Smooth 2x'.
- Processing of binary layers (measurement areas)**: Points to the 'Binary Processing' section, including 'Smooth' and 'Morpho Separate Objects'.
- Measurement parameters**: Points to the 'Save Binary' section, which lists features like 'Count', 'Area [px²]', and 'Circularity' with their respective values.

## Output of analyzed results

Automated Measurement results

So...	F...	ObjID	ND.M	A...	MeanIntensity	Circularity	Length	Width	MaxFeret	MeanPh	MinFeret	E
1	New	1	12	1	186.01	1285.19	0.874	20.28	9.17	20.28	N/A	11.59
116	New	1	132	1	185.79	1259.96	0.853	19.97	9.30	19.97	N/A	12.68
176	New	1	200	1	185.06	1090.23	0.396	32.67	5.66	19.57	N/A	14.22
10	New	1	21	1	184.32	1266.93	0.818	20.37	9.05	20.37	N/A	11.41
37	New	1	48	1	182.73	1269.58	0.617	22.30	8.20	20.63	N/A	14.28
20	New	1	31	1	182.41	1255.16	0.829	17.90	10.19	17.90	N/A	13.88
26	New	1	37	1	182.20	1282.40	0.906	18.22	10.00	18.22	N/A	12.89
23	Mean	1	22	1	184.04	1283.66	0.820	21.82	8.38	21.82	N/A	10.66
Mean					214.14	1230.37	0.817	19.65	10.24	20.05	N/A	13.77
St.dev					100.77	60.90	0.125	5.33	3.15	5.24	N/A	3.84
Min					4.86	1024.16	0.336	3.10	1.51	3.10	N/A	2.15
Max					719.41	1314.45	0.989	37.24	20.49	36.41	N/A	26.63

Object catalog





The HC Template is a dedicated software module within NIS-Elements.

- It allows fully automated acquisition and analysis of a large number of high-content, multi-dimensional images with integrated control of the high-speed motorized focus and stage of Ti-E motorized inverted microscope, camera and peripheral devices.
- HC Template within NIS-Elements allows for quick experimental setups with several autofocusing options and an immediate view of measurement data well by well during acquisition and via a heat map for trend observation and further analysis.



### High Content Analysis System

- The microscope-based High Content Analysis System combined with the Ti-E offers a wide range of interchangeable options, including a full range of camera models, such as high-speed and high-definition cameras, as well as a choice of magnifications and fluorescence filters.
- High-speed peripheral devices are optionally available to boost throughput of integrated devices. These include the high-speed piezo Z-objective positioner for fast autofocusing and the automatic plate loader for auto analysis of multiple well plates.

#### High Content Analysis System

Microscope	Motorized inverted microscope ECLIPSE Ti-E
Compatible well plate types	6-, 12-, 24-, 48-, 96-well plates, 384-well plate (for fluorescence observation)
Multiple FOV experiments	Center, Covering, Random, Random+Center and Regular pattern in each well
Illumination methods	Fluorescence, Phase contrast, DIC
Image acquisition	Multichannel, Time lapse, Multidimensional imaging
Acquisition speed	2 min/96 well plate (1 point/well, 30 msec/shot, without Z stacking, PFS (Perfect Focus System))
Data visualization	Plate view, Sample labeling, Heat map, Graphing
Recommended objectives	S Fluor 4x, Plan Fluor 10x, S Plan Fluor ELWD 20xC/40xC (phase contrast objectives can also be used for image acquisition)
Recommended imaging devices	Camera : DS-Qi2 (Nikon), Zyla sCMOS (Andor technology), iXon3 DU-897 (Andor technology) Confocal microscope : A1+, A1R+, C2+ (Nikon)



#### High-speed Piezo Z-objective Positioner (Option)

For fast autofocusing

Z travel range	Up to 200 nm
Z travel speed	30 msec
Repeatability	±5 nm

(Physik Instrumente (PI) GmbH & Co. KG)



#### Automatic Plate Loader (Option)

For automatic imaging with multiple well plates

Number of well plates	Up to 20 plates
Loading speed	30 sec/plate
Barcode reader option	1D and 2D barcode

(Prior Scientific Instruments Ltd.)

## Sequential HC Template workflow from acquisition to analysis

### 1 Experimental setups

The wizard format simplifies settings for each experimental parameter. It also enables sequential operational settings, such as well plate definition, acquisition patterns in a well, wavelengths, autofocus and image analysis.

#### Define general job parameters

- Z-stack
- Sample labelling
- Autofocus
- Sending task completion by e-mail or SNS

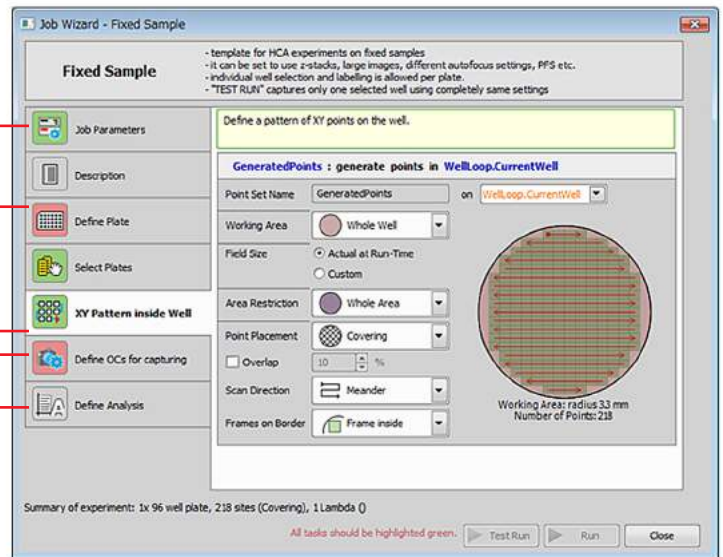
#### Well plate setting

- Define well plate to use
- Select well plate for image capture
- Define XY image capture pattern inside a well

#### Define optical configurations for image capture

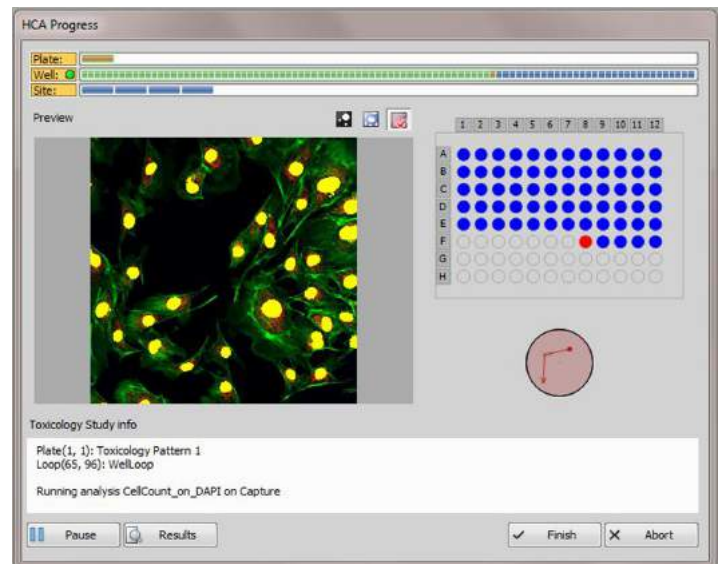
#### Define analysis

(Set the custom image analysis using General Analysis on page 11.)



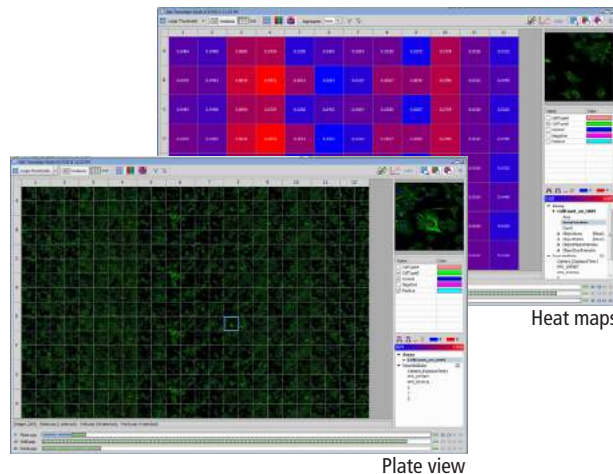
### 2 Progress display

Current well-plate acquisition point is displayed in real time along with a live image. Image analysis is conducted during image acquisition, allowing for immediate observation of data collection and experimental status.

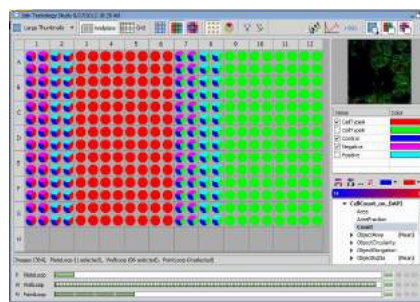


### 3 Results display

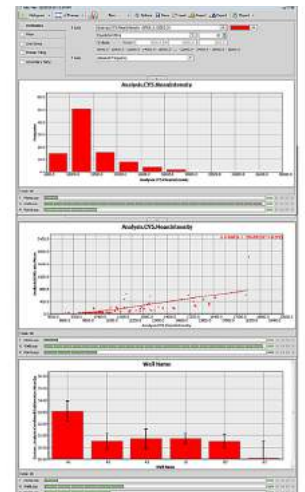
Various formats are available for displaying results. Plate view is a centralized view that shows acquired images and all associated data. Sample labeling manages image data by linking cells by name, type and quantity of reagent. Heat maps visualize trends in measurement results and analytical values. Graphical displays of analytical values such as histograms, scatter plots and bar graphs are also available.



Heat maps



Sample labeling



Visual programming tool that enables creation of highly flexible experimental templates

## JOBS Editor

Option (Ar)

JOBS Editor is a visual programming tool that can create experimental templates (JOBS) simply by dragging and dropping the settings for "tasks" in the sequence of experimental procedures, such as sample definition, image acquisition settings and analysis settings, into the JOBS Editor window.

The HC Template offers streamlined operation of high-throughput imaging/analysis. However, JOBS Editor enables easy creation of more complex and custom experimental templates, from image acquisition to analysis, by providing a number of available tasks, without the need of advanced data programming knowledge and the need for creating a macro.

**Well Plates**

- Define Plate
- Select Wells
- Loop over Wells
- Manual Plate List
- Loop over Plates
- Move to Well Center

**Autofocus + Focus Surface**

- Autofocus Settings
- Autofocus
- Move to Focus Surface

**PFS**

- PFS On and Focus
- PFS Off

**Time Series**

- Time Lapse
- Time Sequence

**Stage XY Points**

- Generate Points
- Predefined Points
- Loop over Points
- Export Point Set to ND
- Import Point Set from ND

**Z-Stack**

- Define Z-Stack
- Z-Stack Loop
- Assign Z-Stack to Point/Well

**Acquisition**

- Capture Definition
- Capture

**Analysis**

- Cell Count Analysis
- Intensity Analysis

Main tasks

Task definition

For offline analysis of high-content data

## Remote Database/JOBS Viewer\* (for HC Template and JOBS Editor)

Option (Ar)

Enables image analysis and management of a large volume of high-content imaging data on an offline PC.

Operating image acquisition and data analysis on separate computers boost total throughput.

General Analysis module is required for offline image analysis.

\*JOBS Viewer is not available in some regions.

- Remote Database allows offline exchange of data between computers and network servers.
- Using JOBS Viewer and/or General Analysis, images acquired through HC Template or JOBS can be analyzed on a dedicated offline computer.

Present in Local	Present in Remote	Action to perform
✗ Absent	✓ Present	Keep Remote
✓ Present	✗ Absent	Upload to Remote
! Jobs may not match each other		Keep Remote

Selected 43 runs and 318 images. Images takes 876 MB disk space.

Present in Local	Present in Remote	Action to perform
✗ Absent	✓ Present	Download to Local
✓ Present	✗ Absent	Keep Local
! Jobs may not match each other		Keep Local

Selected 43 runs and 318 images. Images takes 876 MB disk space.

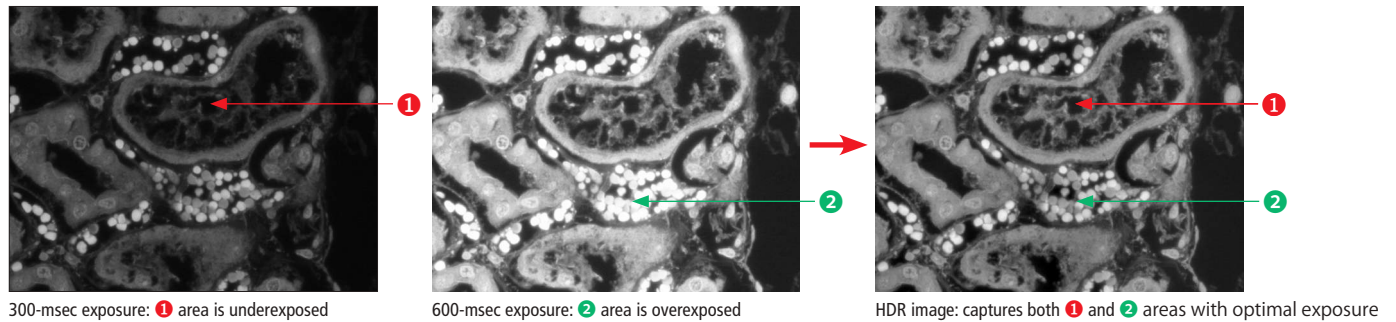
Data upload/download using Remote Database

# Others

## HDR (High Dynamic Range) Image Acquisition

**Ar Option ( Br D )**

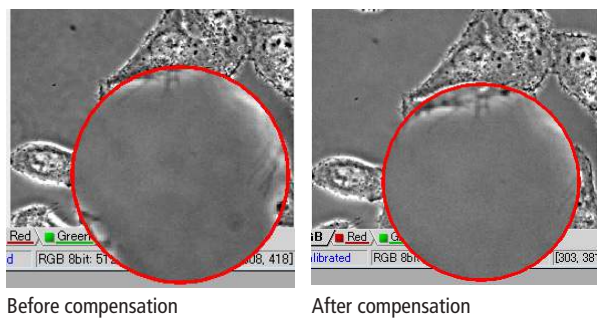
HDR creates an image with appropriate brightness in both the dark and bright regions in a sample by combining multiple images acquired with different exposure settings. It is also possible to create HDR image using multiple captured images.



## Background Compensation

**Ar Br D**

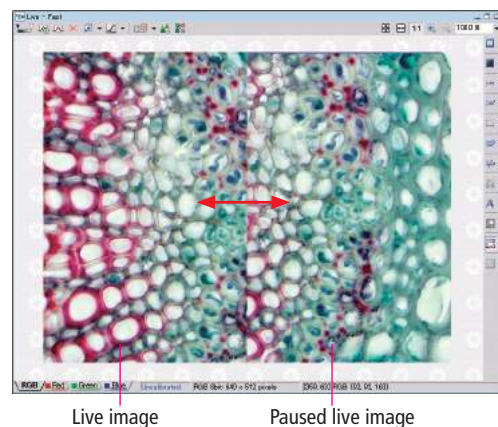
Background correction uses previously captured images to correct uneven background brightness while imaging or of captured images.



## Live Image Comparison

**Ar Option ( Br D )**

Live Compare enables easy image comparison between a sample image and a live image. Live observation side by side with a paused live image is also available in split screen mode.



## Database

**Option ( Ar Br D )**

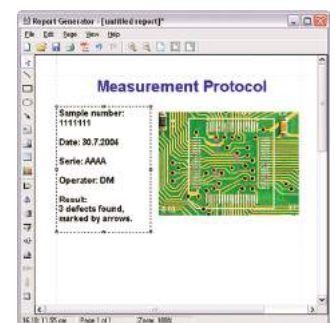
Using the organizer function, captured images are displayed in thumbnails for easy retrieval of the desired image. By simply clicking on the thumbnail image in this view, the image is easily opened. Sorting and filtering this database of images and datasets using acquisition details such as objective settings, date and author is an easy method for data management as well.



## Report Generation

**Ar Br D**

Images captured with NIS-Elements have information such as acquisition details and analysis results, allowing export and PDF conversion of the image and the associated image header and data information.



## User Rights/Control

**Ar Br D**

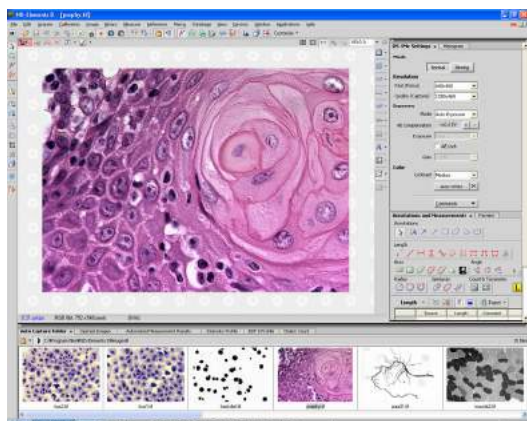
For safe system management, it is possible to individually limit each user authorization using the user account of Windows® (such as the Administrator or Guest) or the user account of NIS-Elements. It limits the authorization and modification of the device settings (microscopes, cameras or others), optical configuration and layout editing.

# GUI Option

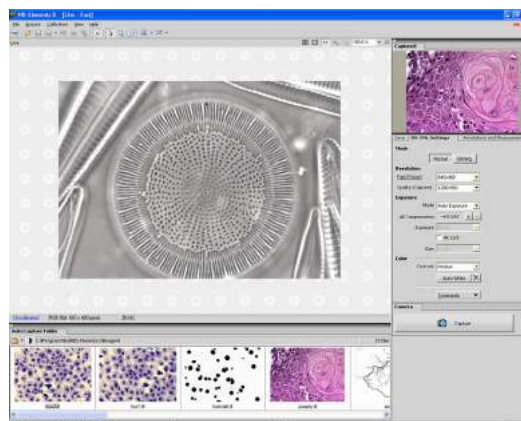
## Industrial Simple GUI

D

With D package, the simple GUI mode provides controls for the most common operations such as image capture and simple measurement.



Standard GUI mode: Displays all functions of D package

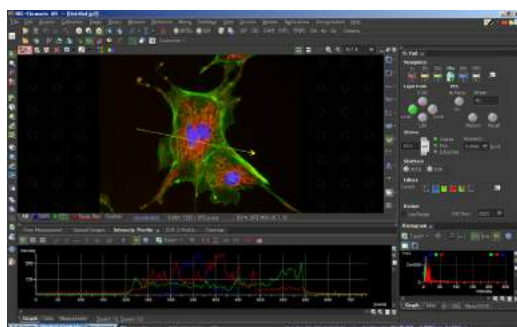


Simple GUI mode: Display only image capturing and measurement

## Dark Color Scheme

Ar Br

This popular display option mode has a brightness level interface color palette suitable for use in a dark microscopy room.



## Layout Manager

Ar Br D

Layout manager enables customizing layouts of controls, toolbars and menus and application (image acquisition or measurement). Saving custom layouts is possible and accessible through one-click tab access.



## Compatibility with Third-party Products

NIS-Elements is compatible not only with Nikon products but also with third-party products such as high-sensitivity CCD cameras and peripheral devices. Third party devices and cameras are easy to integrate through the NIS-Elements intuitive install and device manager.

## Off-line Package for Analysis

The NIS-Elements off-line software package offers analysis tools such as intensity measurements and object counting of tiff and multi-dimensional format images captured with Nikon's microscopes and third-party software.

## Viewer Software

This is free software for image display of single images and datasets captured using NIS-Elements. Possible views include Tile View, Max/Min Projections and 3D Volume View. Saving multi-dimensional files into TIFF format is available as well. The viewer is downloadable from the Nikon website.



## Software Upgrade Agreement (SUA) License

NIS-Elements can be upgraded for one year from the date of purchase. The Software Upgrade Agreement (SUA) License, which is purchasable in one-year license segments, extends the access to the latest version of NIS-Elements.





## Supporting Broad Microscope Imaging

NIS-Elements is a common software platform for Nikon microscope systems, which allows the comprehensive control of wide range of functions for cameras, confocal imaging systems and super resolution microscopes.

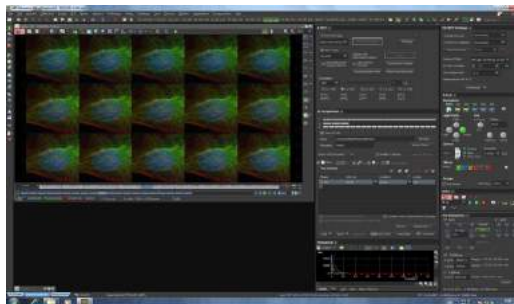
### NIS-Elements C

NIS-Elements C is an optimized software package for confocal imaging. It is compatible with high-speed/high-resolution confocal microscope A1+/A1R+, multiphoton confocal microscope A1 MP+/A1R MP+, confocal microscope C2+ and spectral confocal imaging systems.



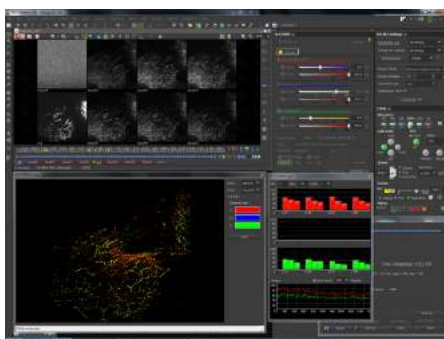
### N-SIM Analysis option

N-SIM Analysis option allows control of Nikon Super-Resolution Microscope N-SIM, which can achieve an image resolution of 85nm and temporal resolution of up to 0.6 sec/frame using high frequency Structured Illumination.



### N-STORM Analysis option

N-STORM Analysis option enables control of Nikon Super-Resolution Microscope N-STORM, which realizes an incredible image resolution of approx. 20nm by utilizing Stochastic Optical Reconstruction Microscopy (STORM).



## Features

	AR	BR	D
Window style	MDI (Multiple Document Interface)	MDI (Multiple Document Interface)	SDI (Single Document Interface)
Dark color scheme	○	○	—
Industrial simple GUI	—	—	○
Camera control	○	○	○
Microscope control	○	○	○
Nikon made peripheral control	○	○	○
Non-Nikon peripheral control	○	○	○
Live image capture	○	○	○
Time-lapse image capturing (T)	○	○	△
Z-series image capturing (Z)	○	○	○
Multichannel image capturing ( $\lambda$ )	○	○	—
Multipoint image capturing (MP)	○	○	○
Multidimensional image capturing	● Up to 6D	● Up to 4D	—
Stimulation experiment	●	—	—
RAM capture	○	—	—
HDR image capture	○	●	●
AVI live-stream capture	○	○	○
Objective calibration	○	○	○
Capturing data savings (Meta-data)	○	○	○
Image filtering	○	△	△
Binary	○	△	▲
LUT (look up table)	○	○	○
Histogram	○	○	○
Manual measurement	○	○	○
Auto measurement	○	○	●
Intensity line profile	○	○	○
Intensity surface plot	○	○	○
Time (intensity) measurement	○	●	—
3D measurement	●	▲	▲
Volume measurement	○	—	—
Database	●	●	●
Macro	○	△	△
Advanced interpreter	○	●	●
Report generator	○	○	○
Live compare	○	●	●
Volume view	○	△	△
EDF (Extended depth of focus)	●	●	●
3D surface view	●	●	●
Ratio view	○	—	—
SD deconvolution	●	—	—
AQ blind deconvolution	●	—	—
2D real time deconvolution	●	—	—
2D deconvolution	●	—	—
3D deconvolution	●	—	—
3D blind deconvolution	●	—	—
Wavelength switcher	●	●	●
TTL/analog IO	●	●	●
Object classifier	●	—	—
Object tracking	●	—	—
Calcium & FRET	●	—	—
General analysis	●	—	—
HC Template	●	—	—
JOBS Editor	●	—	—
N-SIM analysis	●	—	—
N-SIM offline analysis	●	—	—
N-STORM analysis	●	—	—
N-STORM offline analysis*	●	—	—
Metalogical analysis	—	—	●

○ : Full function   △ : Limited function   — : Not available   ● / ▲ : Option

\* N-STORM analysis is required.

## NIS-Elements Supported Devices (ver. 4.30 or later)

### Nikon Cameras

DS-Ri2  
 DS-Qi2  
 CCU: DS-U2/L2 (for camera head DS-2Mv/Vi1/2MBW/2MBWc/5M/5Mc/Fi1/  
 Fi1c/Qi1/Ri1)  
 CCU: DS-U3/L3 (for camera head DS-Vi1/Fi1/Fi1c/Qi1/Ri1/Fi2)  
 DQC-FS<sup>2</sup>

### Third-party Cameras

#### Photometrics

Evolve  
 QuantEM  
 CoolSNAP HQ2  
 CoolSNAP ES<sup>2</sup>  
 Cascade 128+<sup>2</sup>  
 Cascade II 512<sup>2</sup>  
 Cascade 1K<sup>2</sup>

#### Andor Technology

Luca S, Luca R  
 iXon+ 897, 888, 885  
 iXon X3  
 iXon Ultra  
 iKon-M  
 Clara  
 Neo sCMOS  
 Neo 5.5 sCMOS  
 Zyla 5.5 sCMOS (3-Tap, 10-Tap)  
 Zyla 4.2 sCMOS

#### QImaging

Retiga EXi Aqua/Blue  
 Retiga 2000R - Mono/Color  
 Retiga SRV + RGB-HM-S Slide  
 Rolera EMC2  
 QIClick  
 QICAM

#### Hamamatsu

ImagEM  
 ImagEM 1K  
 ImagEM X2  
 ORCA-R2  
 ORCA-Flash2.8  
 ORCA-Flash4.0  
 ORCA-Flash4.0 LT  
 ORCA-D2  
 ORCA  
 C9100-02, C9100-12

#### Imaging Source

DFK/DMK 31,41,51,72 series  
 DFK/DMK 23U274  
 DFK/DMK 23UP031  
 DMK 23UM021  
 DFK 23U445

#### Others

TWAIN Device<sup>2</sup>

### Nikon Microscope Devices

Biological Microscope Ti (HUBC/A, HUBC/A-U, Ti-LAPP System<sup>4</sup>)  
 Biological Microscope TE2000 (Perfect Focus System)  
 Biological Microscope 90i (ND filter, Stage, DIH-E/M)  
 Biological Microscope 80i (DIH-E/M, D-FL-E, C-Box/C-Box2)  
 Biological Microscopes Ni-E, Ni-U, Ci-E  
 Fixed-stage Microscope FN1 + D-DH-E-A1<sup>1</sup>  
 Stereo Microscope SMZ25/18/1270i/1270/800N  
 Multizoom Microscope AZ100M  
 Industrial Microscope LV-N Series<sup>3</sup>  
 Measuring Microscope MM-400/800<sup>2</sup>  
 Metallographic Microscope MA200  
 Inspection Microscopes L200N/L300N  
 C-HGFIE HG Fiber Illuminator Intensilight  
 C-LEDFI Epi-FI LED Illuminator  
 Laser Module LU-N Series  
 LV-NCNT-N Nosepiece controller  
 NI-SH-CON

### Third-party Devices

Prior Scientific  
 ProScan III  
 ProScan II  
 Prior PCI II  
 OptiScan II ES10  
 NZ100,200,400 NanoStageZ  
 ES10ZE  
 Ludl Electronic Products  
 MAC5000, MAC6000  
 Märzhäuser Wetzlar  
 TANGO Desktop, Tango PCI  
 LSTEP  
 Vincent Associates (Uniblitz)  
 VCM-D1  
 Sutter Instrument  
 Lambda 10-2, 10-3, SC, 10-B, XL  
 Physical Instrument  
 PI E-662, 665 (RS232)  
 Photometrics  
 Dual View  
 ASI (Applied Scientific Instrumentation)  
 MS-2000  
 FW-1000  
 SC-2000  
 National Instruments  
 TTL Input/Output (NI Card)  
 Tokai Hit  
 WSKM  
 Mad City Labs  
 Nano-Drive  
 Yokogawa  
 CSU-W1<sup>4</sup>  
 CSU-X1<sup>4</sup>

<sup>1</sup> Only compatible with 64-bit version OS

<sup>2</sup> Not compatible with 64 bit version OS

<sup>3</sup> Not compatible with LV-DAF Auto Focus Unit

<sup>4</sup> Only compatible with NIS-Elements AR

## Supported Operation System

Windows 7 Professional SP1 (32/64-bit version)

\* NIS-Elements Ar and C are only compatible with the 64-bit version.

NIS-Elements is compatible with all common file formats, such as JP2, JPG, TIFF, BMP, GIF, PNG, ND2, JFF, JTF, AVI, ICS/IDS. ND2 is a special format for NIS-Elements. ND2 allows storing sequences of images acquired during nD experiments. It contains information about the hardware settings and the experiment conditions and settings.

Specifications and equipment are subject to change without any notice or obligation on the part of the manufacturer. September 2014 ©2006-14 NIKON CORPORATION



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 Industrial Metrology Business Unit



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**NIKON CORPORATION**

Shin-Yurakucho Bldg., 12-1, Yurakucho 1-chome, Chiyoda-ku, Tokyo 100-8331, Japan  
 Microscope Solutions Business Unit, phone: +81-3-3216-2375 fax: +81-3-3216-2385  
 Industrial Metrology Business Unit, phone: +81-3-3216-2384 fax: +81-3-3216-2388  
 (Scheduled to relocate to the following address on Nov. 25, 2014:  
 Shinagawa Intercity Tower C, 2-15-3, Konan, Minato-ku, Tokyo 108-6290 Japan)  
[www.nikon.com](http://www.nikon.com)

**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 METROLOGY, INC.**

12701 Grand River Avenue, Brighton, MI 48116 U.S.A.  
 phone: +1-810-220-4360 fax: +1-810-220-4300  
 E-mail: [sales\\_us@nikonmetrology.com](mailto:sales_us@nikonmetrology.com)  
<http://us.nikonmetrology.com/>  
<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 METROLOGY EUROPE NV**

Geldenaaksebaan 329, 3001 Leuven, Belgium  
 phone: +32-16-74-01-00 fax: +32-16-74-01-03  
 E-mail: [sales\\_europe@nikonmetrology.com](mailto:sales_europe@nikonmetrology.com)  
<http://www.nikonmetrology.com/>

**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 INDIA PRIVATE LIMITED**

INDIA phone: +91-124-4688500 fax: +91-124-4688527

**NIKON CANADA INC.**

CANADA phone: +1-905-602-9676 fax: +1-905-602-9953

**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 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

**NIKON UK LTD.**

UNITED KINGDOM phone: +44-208-247-1717 fax: +44-208-541-4584

**NIKON METROLOGY UK LTD.**

UNITED KINGDOM phone: +44-1332-811-349 fax: +44-1332-639-881

E-mail: [sales\\_uk@nikonmetrology.com](mailto:sales_uk@nikonmetrology.com)

**NIKON FRANCE S.A.S.**

FRANCE phone: +33-1-4516-45-16 fax: +33-1-4516-45-55

**NIKON METROLOGY SARL**

FRANCE phone: +33-1-60-86-09-76 fax: +33-1-60-86-57-35

E-mail: [sales\\_france@nikonmetrology.com](mailto:sales_france@nikonmetrology.com)

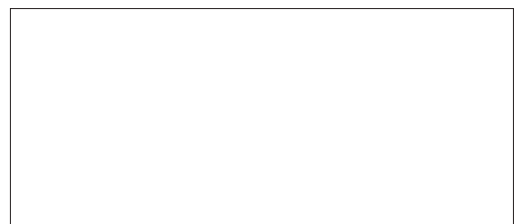
**NIKON GMBH**

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

**NIKON METROLOGY GMBH**

GERMANY phone: +49-6023-91733-0 fax: +49-6023-91733-229

E-mail: [sales\\_germany@nikonmetrology.com](mailto:sales_germany@nikonmetrology.com)



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