

# Shaping high-end imaging for your research



## **ZEISS LSM 980 with Airyscan 2**

Your Unique Confocal Experience for Fast and Gentle Multiplex Imaging



# Your Unique Confocal Experience for Fast and Gentle Multiplex Imaging

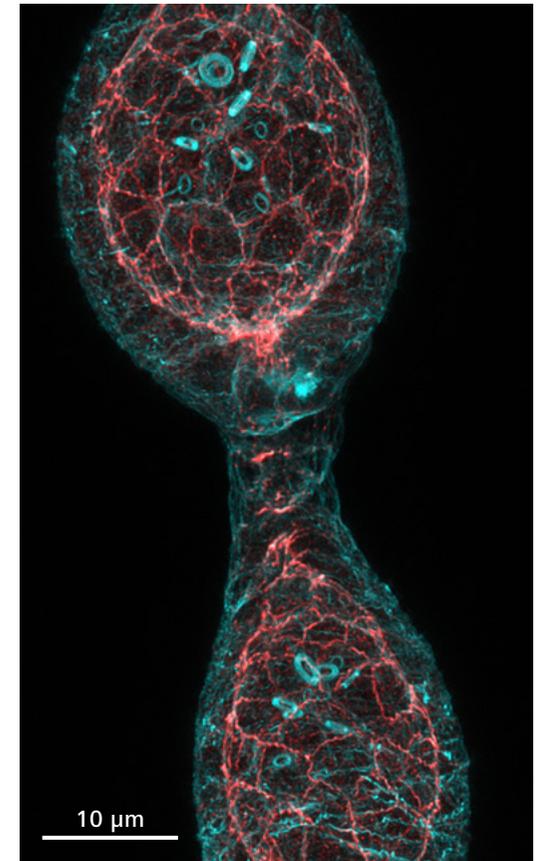
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To analyze life with as little disturbance as possible, you need excellent imaging performance combined with low phototoxicity and high speed.

LSM 980, your platform for confocal 4D imaging, is optimized for spectral multiplexing by simultaneous spectral detection of multiple labels, from 380 to 900 nm emission. LSM Plus puts you at the forefront of a new confocal experience across all modes: whether it be live imaging experiments, spectral imaging with up to 36 channels, or near-infrared and multiphoton experiments. With LSM Plus, you can reliably gather more information while increasing productivity.

Add Airyscan 2 for gentle super-resolution imaging. Capture larger fields of view or dynamic processes with the flexible Multiplex modes using shorter acquisition times than previously possible. Or push resolution further to identify new structures with Airyscan Joint Deconvolution (jDCV). Discover the underlying molecular dynamics of your living samples by fully utilizing the additional information only Airyscan with its 32-channel area detector can offer.

A wealth of software helpers will optimize your workflows, from image acquisition to data management. With ZEN Connect you can document and share all details of your experiments. You'll always maintain context as you combine overview images, ROIs and additional data, even across imaging modalities.



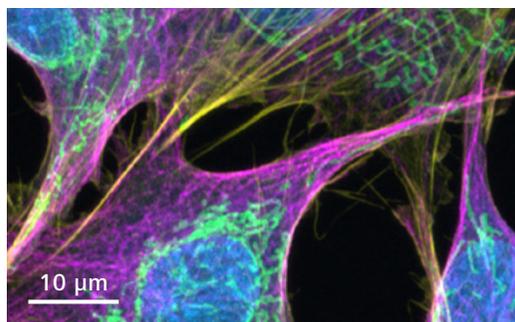
Staining of F-actin (Phalloidin, cyan) and DE-Cadherin (red) in the *Drosophila* germarium. Imaged with ZEISS Airyscan 2 followed by Joint Deconvolution.  
Courtesy of T. Jacobs, AG Luschnig, WWU Münster; with T. Zobel, Münster Imaging Network, Germany

# Simpler. More Intelligent. More Integrated.

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## A Unique Confocal Experience

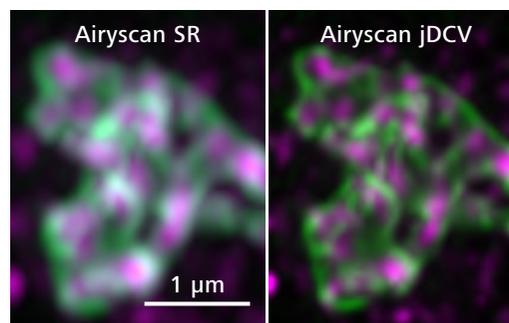
LSM 980 combines everything needed to image your most challenging samples. A light-efficient beam path with up to 36 simultaneous channels and full spectral flexibility into the near infrared (NIR) range give you the perfect basis for multi-color experiments with living samples. On top of this, LSM Plus effortlessly improves all your experiments. The unique combination of spectral imaging with improved signal-to-noise ratio and resolution enables lower laser power for your live cell experiments and avoids time loss due to averaging. If you need to resolve selected sample structures even further, seamlessly integrate Airyscan 2 in your confocal workflow to get sensitive super-resolution imaging.



GATTA-Cells 4C NIR. Labelled Nucleus (blue) DAPI, mitochondria (green) Tom20 with Alexa Fluor 488, actin (yellow) with Alexa Fluor 647, and microtubules (magenta)  $\alpha$ -Tubulin with Alexa Fluor 750. Imaged with LSM Plus.

## Image with More Sensitivity

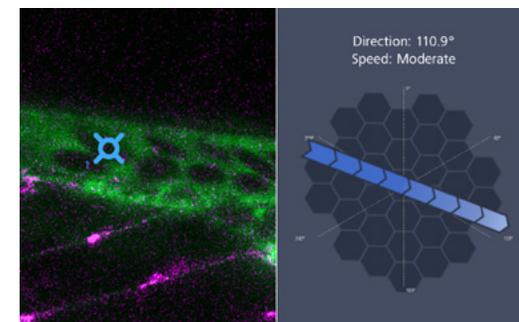
Revolutionary Airyscan 2 allows you to do more than any conventional LSM detector. Each of its 32 detector elements collects additional information, while all of them together gather even more light, yielding super-resolution quantitative results. By adding structural information with Joint Deconvolution (jDCV), you can push resolution even further. Or use the Multiplex modes to get super-resolution information up to 10 times faster. Adapted illumination and detection schemes let you image the most challenging three-dimensional samples with high framerates and beyond the diffraction limit, while still being gentle to your sensitive samples.



HeLa cells expressing TOM20-pHluorin (green), anti-Cox8a (purple). TOM20 is a mitochondrial importer subunit in the outer mitochondrial membrane, while Cox8a resides in the inner mitochondrial membrane as part of the electron transport chain. Courtesy of K. Busch, S. Morris, Westfälische Wilhelms-Universität Münster; with T. Zobel, Münster Imaging Network, Germany

## Increase Your Productivity

It's never been easier to set up complex confocal live cell imaging experiments. ZEN microscopy software puts a wealth of helpers at your command to achieve reproducible results in the shortest possible time. AI Sample Finder helps you quickly find regions of interest, leaving more time for experiments. Smart Setup supports you in applying best imaging settings for your fluorescent labels. Direct Processing enables parallel acquisition and data processing. ZEN Connect keeps you on top of everything, both during imaging and later when sharing the whole story of your experiment. It's easy to overlay and organize images from any source.



Dynamics Profiler enables flow measurements to determine speed and direction of blood flow in zebrafish larvae. Sample courtesy of V. Hopfenmüller, Leibniz Institute on Aging – Fritz Lipmann Institute (FLI), Germany.

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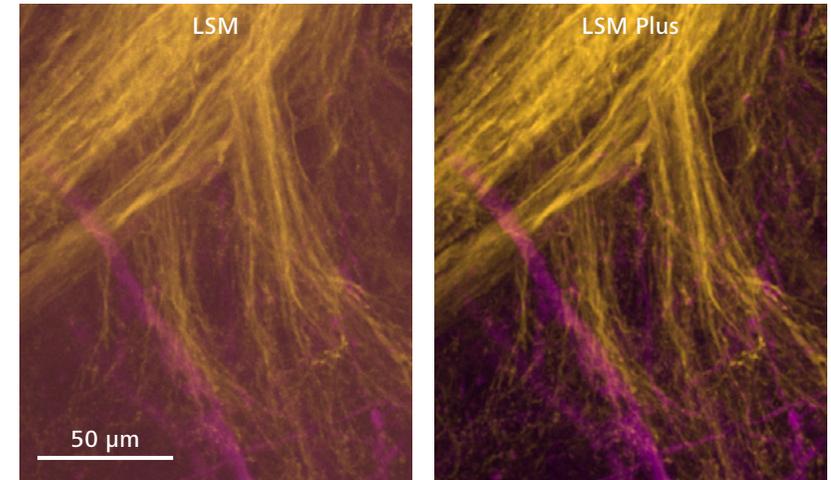
## LSM Plus: Improving the Whole Confocal Experience

Laser scanning microscopy is valued for its instant, high-quality imaging of optical sections and has set the imaging standard for a wide variety of samples and experiments. It's hard to imagine how the data quality of this technology can be improved further while fully preserving its appreciated ease of use and application flexibility.

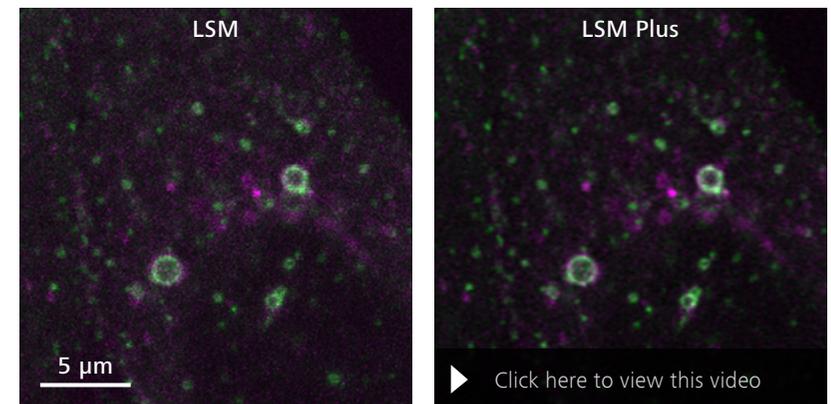
LSM Plus is doing just that: improving literally any confocal experiment with ease, independent of detection mode or emission range. Its linear Wiener filter deconvolution needs next to no interaction while still ensuring a reliable quantitative result. Just as in our time-tested Airyscan super-resolution processing, the underlying optical property information is adapted automatically based on objective lens, refractive index, and emission range.

Apply LSM Plus with no extra effort and benefit from:

- **Enhanced signal to noise (SNR)** at high acquisition speed and low laser power—particularly useful for live cell imaging with low expression levels
- **Improved resolution** of spectral data with up to 36 channels in a single scan
- **More spatial information** and even greater resolution enhancement for bright samples that allow to close the pinhole of the LSM
- **Integrated workflows** to combine the advantages of LSM Plus with Airyscan super-resolution imaging



Cockroach brain neurons (Alexa 488: yellow, Alexa 647: magenta) without (left) and with LSM Plus (right). Sample courtesy of M. Paoli, Galizia Lab, University of Konstanz, Germany



Live cell imaging experiment of U20S cells with Rab4a:mCherry and Rab5:mEmerald without (left) and with LSM Plus (right).

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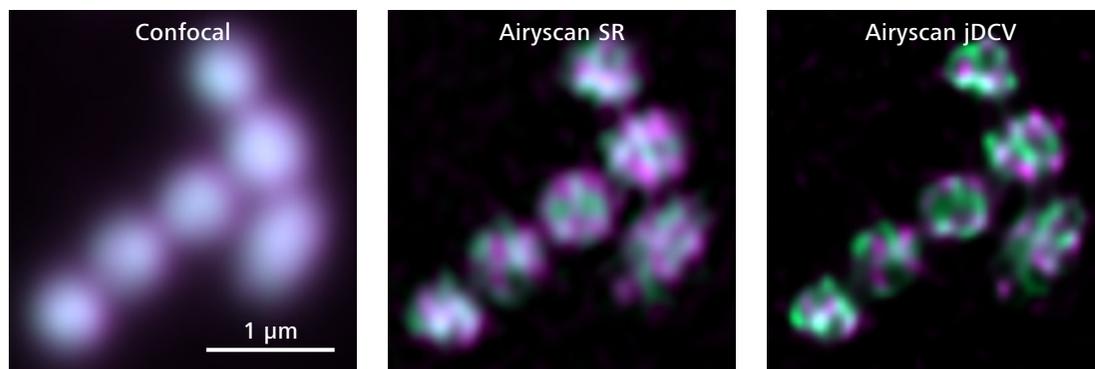
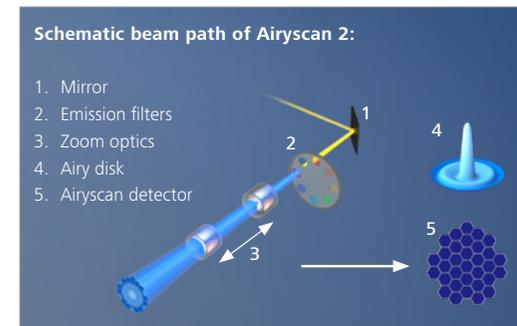
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## The Airyscan Principle: A Unique Combination of Super-resolution Imaging and High Sensitivity

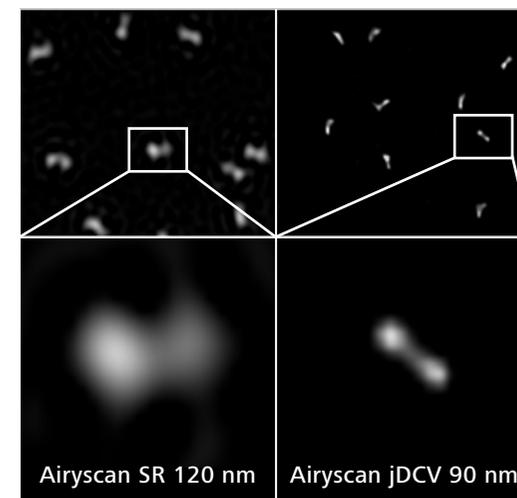
Classic confocal laser scanning microscopes use point illumination to scan the sample sequentially. The microscope optics transform each point to an extended Airy disk. A pinhole spatially limits this Airy disk to block out-of-focus light from the detector. Closing the pinhole gives higher resolution, but at the price of detecting fewer photons—which cannot be brought back. Airyscan 2 is an area detector with 32 circularly arranged detection elements. Each of these acts as a small pinhole, contributing to super-resolution information, while the complete detector area collects more light than the standard confocal setting. This produces much greater light efficiency while capturing enhanced structural information.

## 32 Views Mean More Information: Powerful Deconvolution with Airyscan jDCV

Each of the 32 detector elements has a slightly different view on the sample, providing additional spatial information that makes Joint Deconvolution possible. This reduces the distance that can be resolved between two points even further—down to 90 nm. Your super-resolution experiments will benefit from an improved separation of single or multiple labels.



Comparing the confocal image (left) with Airyscan SR (middle) and Airyscan Joint Deconvolution (right). Mitochondria in an *Arabidopsis thaliana* cell. mCherry (green) is targeted to the matrix and GFP (magenta) to the intermembrane space. Courtesy of J.-O. Niemeier, AG Schwarzländer, WWU Münster, Germany



GATTA SIM nanoruler imaged with Airyscan SR (GATTA-SIM 120B, left) and Airyscan jDCV (GATTA-SIM 90B, right).

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## The Multiplex Modes for Airyscan 2:

### Large Fields of View and Whole Sample Volumes in the Shortest Time

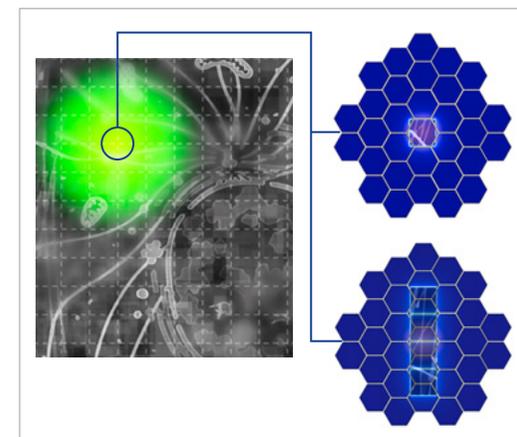


HeLa cells stained for DNA (blue, Hoechst 44432), microtubules (yellow, anti-tubulin Alexa 488) and F-actin (magenta, phalloidin Abberior STAR Red). Imaged with ZEISS Airyscan 2 in Multiplex mode for efficient super-resolution imaging of a large field of view. Courtesy of A. Politi, J. Jakobi and P. Lenart, MPI for Biophysical Chemistry, Göttingen, Germany

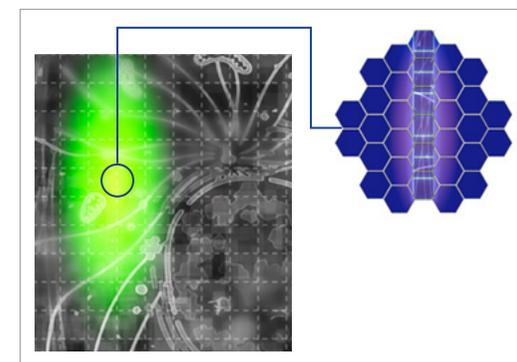
In Multiplex modes, Airyscan detector advantages are combined with adapted illumination and readout schemes, giving you a choice of different parallelization options. Multiplex modes use knowledge of the shape of the excitation laser spot and the location of single area detector elements to extract more spatial information, even during parallel pixel readout. This allows larger steps when sweeping the excitation laser over the field of view, improving acquisition speed.

Capturing more spatial information in the pinhole plane allows final image reconstruction with better resolution than the acquisition sampling.

| LSM 980 with Airyscan 2            |                |                 |                 |                 |
|------------------------------------|----------------|-----------------|-----------------|-----------------|
|                                    | Airyscan SR    | Multiplex SR-4Y | Multiplex SR-8Y | Multiplex CO-8Y |
| Parallelization                    | 1              | 4               | 8               | 8               |
| Resolution                         | 120/120        | 140/140         | 120/160         | 180/220         |
| FPS at max FOV                     | 0.2 (Zoom 1.7) | 1.0 (Zoom 1)    | 2.0 (Zoom 1)    | 9.6 (Zoom 1)    |
| FPS at 512 × 512 pixels            | 4.7            | 25              | 47.5            | 34.4            |
| Antibody labeling, fine structures | +++++          | ++++            | +++             | ++              |
| Antibody labeling, tiling          | ++             | ++++            | +++++           | +++             |
| Live cell imaging                  | ++             | +++             | ++++            | +++++           |



Airyscan 2 in Multiplex SR-4Y mode allows you to acquire up to four super-resolution image lines with high SNR in a single sweep. For each illumination position, Airyscan SR mode generates one super-resolution image pixel.



For Airyscan Multiplex SR-8Y and CO-8Y, the illumination laser spot is vertically elongated to capture 8 image pixels for each illumination position. Sampling can be done in super-resolution (SR) or confocal (CO) resolution. Use this speed advantage for ultrafast time series of single slices, rapid tiling of large areas, or fast volumetric time-lapse imaging.

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## ZEISS Dynamics Profiler: Add a New Dimension to Live Imaging

Molecular data offers new, and often overlooked, insights about living samples. Fluorescence Correlation Spectroscopy (FCS) is an established method to investigate molecular characteristics. While a precise and very sensitive method, traditionally it is limited to extremely low expression levels or molecule concentrations that can be well below the experimental expression levels in live research samples.

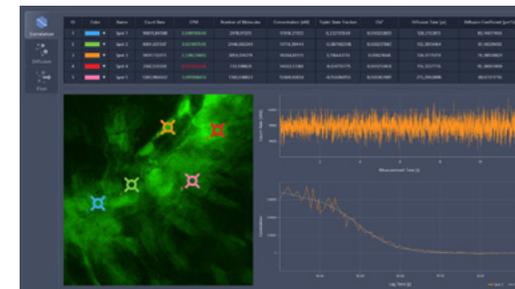
Airyscan uniquely employs all its detector elements to collect 32 individual FCS intensity traces per measurement. The mean value of the inner 19 elements provides robust and reliable measurements on molecular concentration and dynamics, even for bright samples.

Moreover, the area detector allows a variety of spatial cross-correlation analyses by using combinations of single detector elements. Asymmetric diffusion analysis is calculated by cross correlating the center element of the detector with the elements of the outer rings, uncovering heterogeneous characteristics within one excitation volume, perfect to investigate samples such as cellular condensates. Cross-correlation of detector pairs that are grouped and aligned in multiple directions along the excitation volume can measure speed and direction of actively moved molecules, such as fluorophores in microfluidic systems or within the bloodstream.

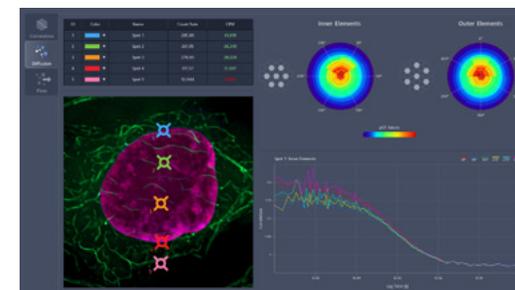
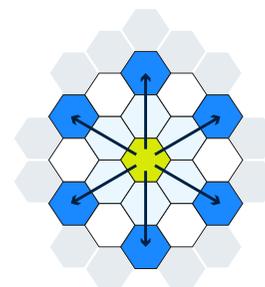
Furthermore, raw data of all 32 detector elements is saved with every single measurement, enabling you to perform your customized analysis as needed, either immediately or when the scientific question arises later.

**Learn more about ZEISS Dynamics Profiler:**

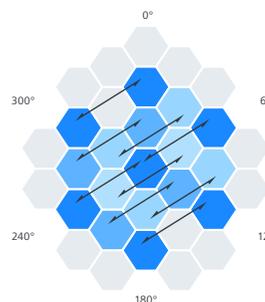
[www.zeiss.com/dynamics-profiler](http://www.zeiss.com/dynamics-profiler)



Molecular concentration and diffusion data are collected with the innermost 19 elements of the Airyscan detector. The read-out of separate detectors permits measurements at much higher total intensities (brightness) than conventional FCS would allow.



To measure asymmetric diffusion, single Airyscan detector elements of the third ring are cross-correlated with the center element. Polar heatmaps visualize asymmetric diffusion behavior within a measurement spot.



To determine the flow direction and speed within a liquid, a total of 27 detector element pairs are cross-correlated along 3 different axes of the Airyscan detector.

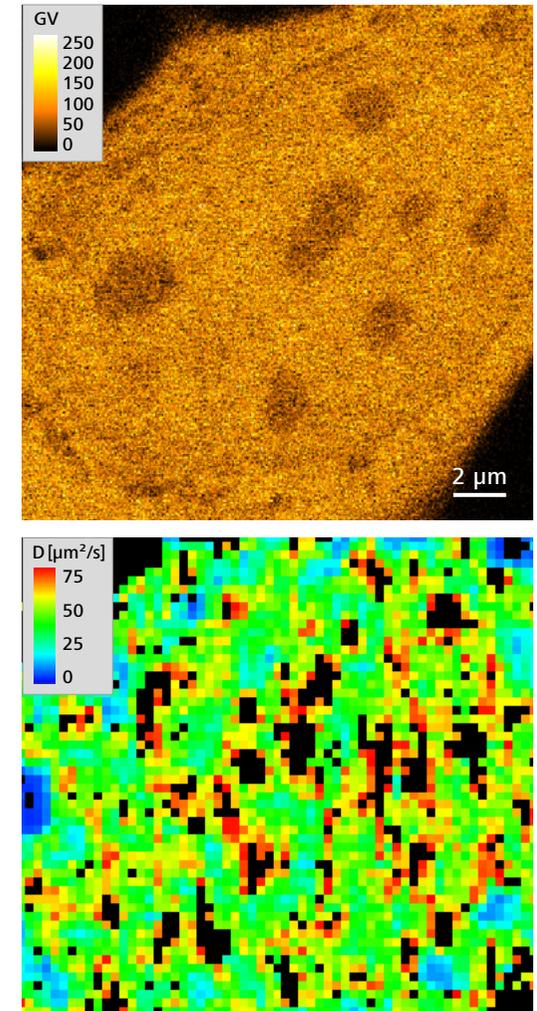
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## Data Beyond Imaging

Combining laser point illumination, linear scanning, and detectors that can capture the signal in photon counting mode make the LSM 980 more than an imaging device:

- **Raster Image Correlation Spectroscopy (RICS)** can generate a display map of molecule concentration and diffusion coefficients of a complete image frame of a cell, or other structures.
- **Fluorescence Correlation Spectroscopy (FCS)** allows a non-invasive insight into molecular concentration and diffusion processes, leading to a deeper understanding of cell functions. To measure on a single molecule basis, you can use single- or multiphoton laser lines and use the full emission range up to 900 nm.
- **Fluorescent Cross Correlation Spectroscopy (FCCS)** allows you to observe molecular interaction between two or more differentially labelled molecules by utilizing the 32 channels of the Quasar and performing FCCS with up to 7 individual channels.
- **Förster Resonance Energy Transfer (FRET)** is another method for investigating molecular distances or interactions, using sensitized emission or acceptor photobleaching approaches.
- **Fluorescence Recovery after Photobleaching (FRAP)** utilizes any of the laser lines to perform flexible photobleaching experiments. The same principle adheres to photomanipulation experiments in general, for example to investigate intracellular movement. Or follow cellular movement within whole organisms by photoconversion of fluorescent protein labels.
- **Fluorescence Lifetime Imaging Microscopy (FLIM)** uses differences in the fluorescence decay to separate components. It is used for functional imaging and takes into account how fluorescence lifetime can be influenced by multiple factors such as ion or oxygen concentration, pH, and temperature. FLIM is beneficial for FRET measurements, analyzing proximity and interaction of molecules (available upon request).



*RICS measurement, using U2OS cells expressing monomeric eGFP. The diffusion of the target can be displayed as a map (right) based on the intensity image (left).  
Sample courtesy of P. Hemmerich, Leibniz Institute on Aging – Fritz Lipmann Institute, Jena Germany*

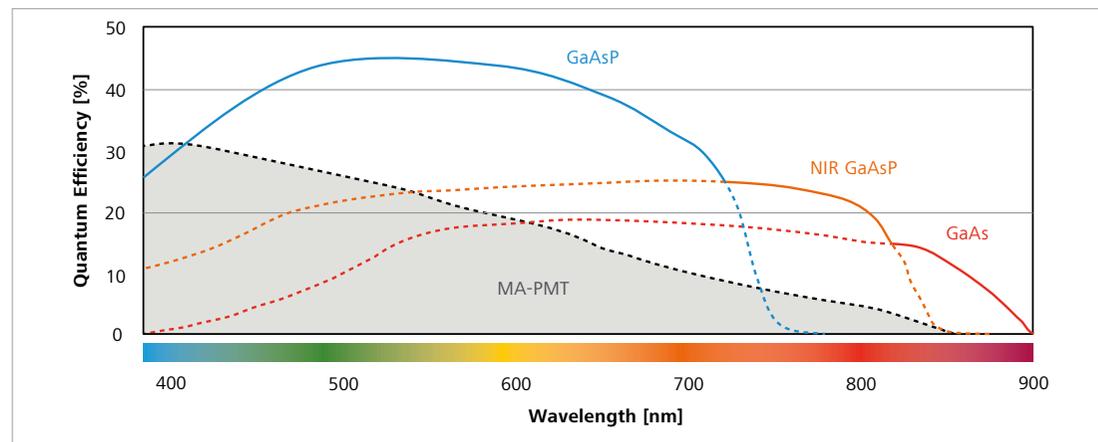
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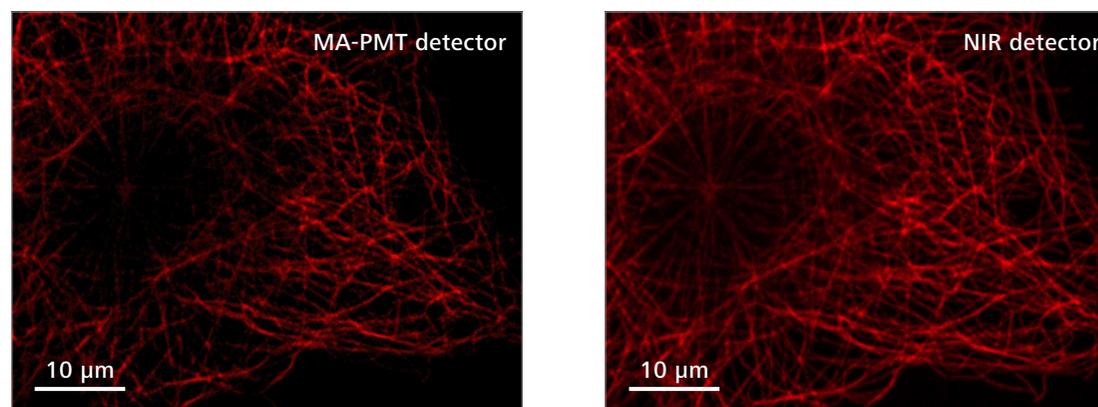
LSM 980 brings a great deal of freedom to your experimental setup. Every single component is optimized to deliver the highest sensitivity and spectral flexibility for your experiments—the perfect starting point for improving all your confocal images with LSM Plus, increasing SNR without adding time or laser light to your experiment.

## Sensitivity

The LSM 980 beam path design ensures that you don't need to worry about sensitivity, which is key to visualizing the lowest signal and resolving all structures. The linear galvo scanners ensure that each image pixel gets identical time contribution. More than 80 % of each frame time is used to collect a precious signal. The low angle Twin Gate beam splitter directs the excitation laser light towards your sample and keeps it efficiently separated from the emission signal. You can even cover the detection range over laser lines to collect all emission light, without worrying about stray light or laser light reflection. For your multi-label experiments, each emission color is captured with the best suited detector technology along the complete wavelength range from 380 nm up to 900 nm. Homogeneous spectral separation is achieved with the holographic grating. The unique recycling loop directs all photons towards the 3, 6, or 34 channel Quasar detection unit that allows you to define the emission detection bands, in nanometer precision, with ease. Use the lowest laser powers even at extremely low expression levels and capture information by switching GaAsP or GaAs detectors to photon counting mode.



Typical spectral quantum efficiency (QE) of ZEISS LSM 980 detectors



Microtubules of Cos-7 cell (Anti-Tubulin AF700). Comparison of the ZEISS LSM 980 MA-PMT and the ZEISS NIR GaAsP detector; excitation with 639 nm laser at same laser power. Emission range for the MA-PMT is set to 660 – 757 nm, and for the NIR detector is 660 – 900 nm, Sample courtesy of U. Ziegler and J. Doehner, University of Zurich, ZMB, Switzerland

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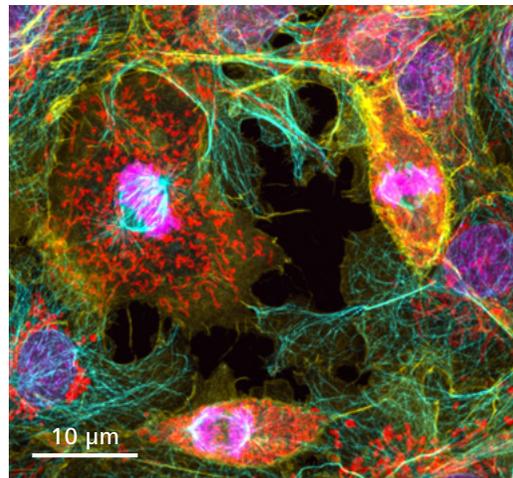
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## Spectral Flexibility

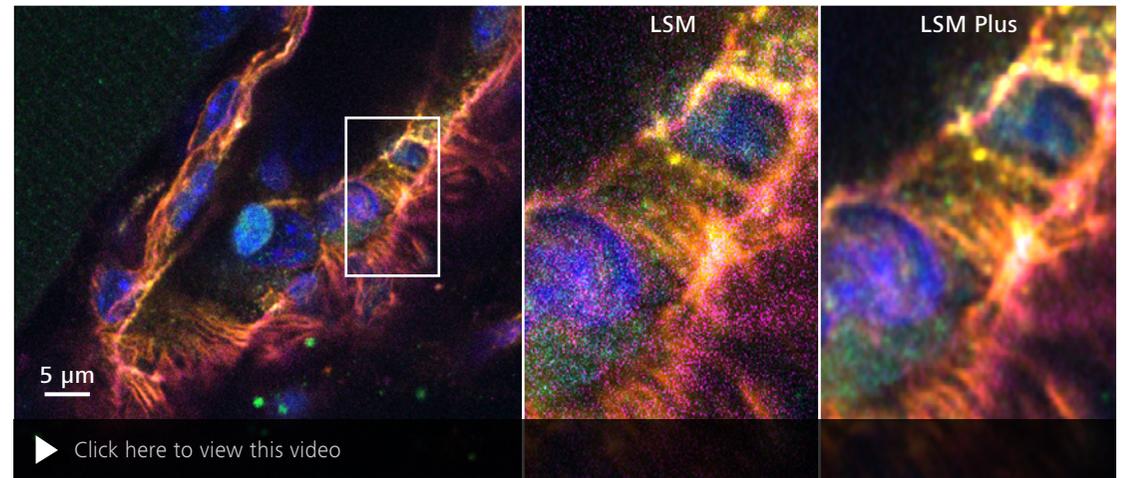
Freely select the fluorescent labels from 380 nm to the near infrared (NIR) range that are best for your experiment and combine them as needed. Smart Setup supports you with the best multi-fluorescent image setup by determining the most suitable solid-state laser lines for excitation and the detectors with the highest quantum efficiency for each wavelength. Expanding your spectral range into the NIR allows you to use more labels in parallel. Visualize additional structures with more dyes in

multi-color experiments, with the Quasar and NIR detectors efficiently supporting spectral multiplexing experiments. NIR fluorescent labels are less phototoxic for living samples due to the longer wavelength. This allows you to investigate living samples for longer periods of time while limiting the influence of light. Additionally, light of longer wavelength ranges is less scattered by the sample tissue, increasing penetration depth. For any of the advantages you pursue with NIR labels, the

dual-channel NIR detector combines two different detector technologies (extended red GaAsP and GaAs) for optimal sensitivity up to 900 nm. To separate even highly overlapping signals or to remove autofluorescence, you can take a Lambda Scan using the complete detection range with up to 36 detectors, keeping both illumination and time required to a minimum. Improve spectral imaging along the complete wavelength range, including Online Fingerprinting, with LSM Plus.



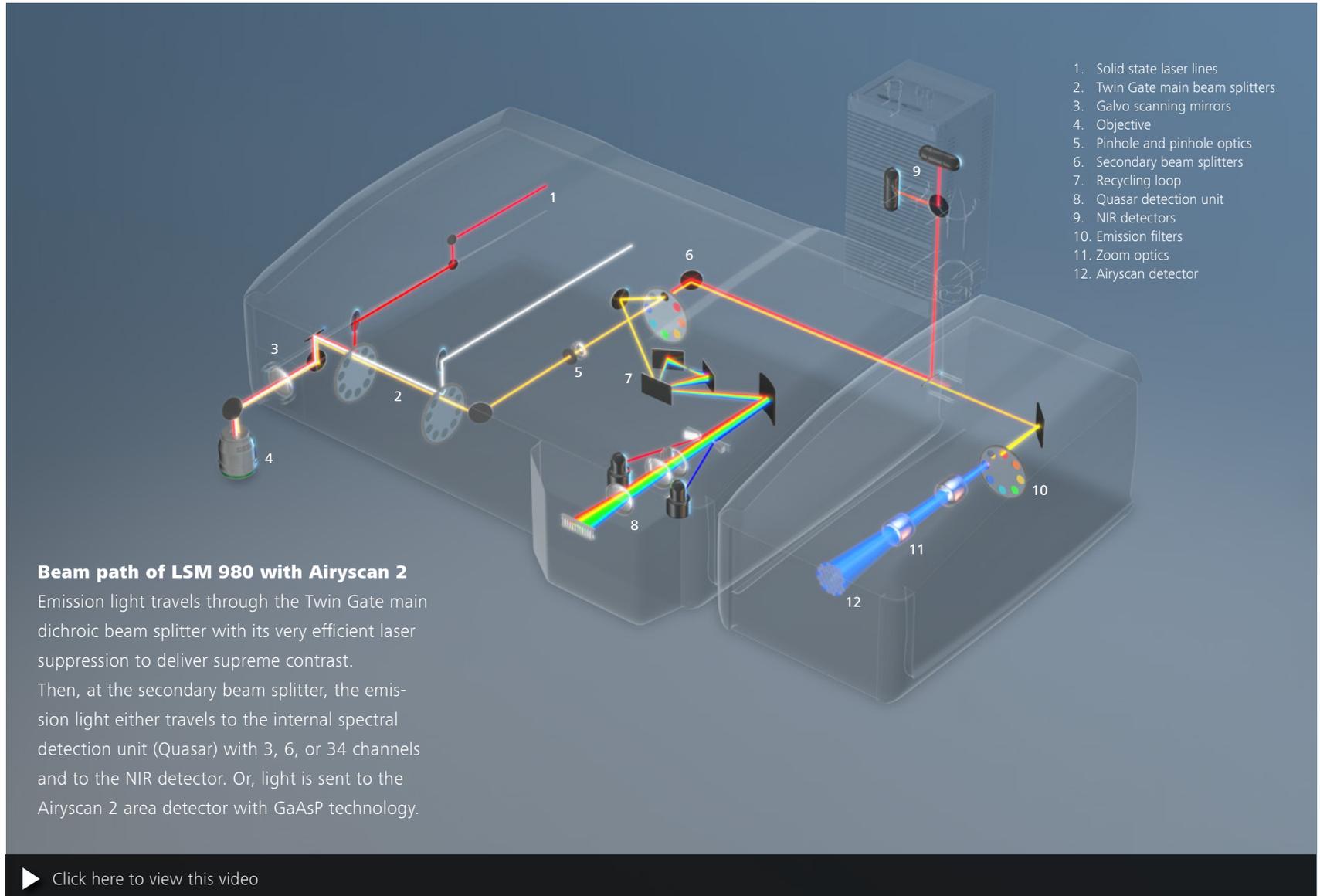
*Cos-7 cells, DAPI (magenta), Anti-tubulin Alexa 568 (blue), Actin Phalloidin-OG488 (yellow) and Tom20-Alexa 750 (red). Imaged in Lambda mode across the visible and NIR spectrum. Individual signals separated by Linear Unmixing. Maximum intensity projection of a z-stack. Sample courtesy of U. Ziegler and J. Doehner, University of Zurich, ZMB, Switzerland*



*Comparison of the improved SNR before and after LSM Plus processing. Murine cremaster muscle, multi-color label with Hoechst (blue), Prox-1 Alexa488 (green), neutrophil cells Ly-GFP, PECAM1 Dylight549 (yellow), SMA Alexa568 (orange), VEGEF-R3 Alexa594 (red), platelets Dylight 649 (magenta). Acquired with 32-channel GaAsP detector using Online Fingerprinting. Courtesy of Dr. S. Volkery, MPI for Molecular Biomedicine, Münster, Germany*

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## AI Sample Finder: Automated Sample identification for Efficient Imaging

Microscopes are becoming increasingly automated. For sample placement, however, microscope parts such as the condenser arm often have to be moved manually. Focus adjustment and identification of the relevant areas on the sample carrier require additional manual steps.

The AI Sample Finder automates this sequence, eliminating time-consuming manual adjustments and reducing the time to image from minutes to just seconds.

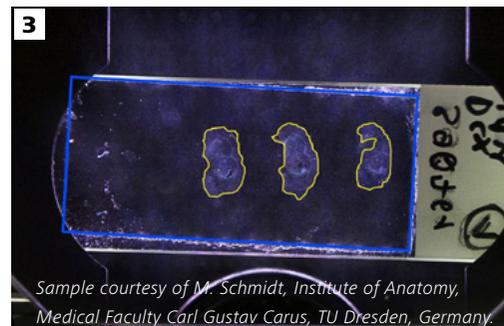
You can access all sample areas directly which allows you starting your experiment faster than ever. The AI Sample Finder greatly improves productivity as you can easily image only those regions containing sample not overlooking potentially important areas.



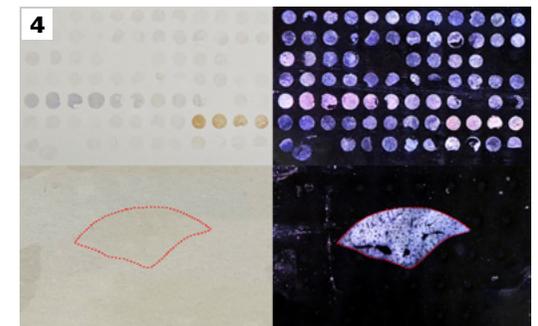
- After you placed the sample on the loading position, the AI Sample Finder automatically moves it to the objective.



- Without the need of manual sample positioning or focusing, an overview image for fast and convenient navigation is taken within seconds. Composite darkfield illumination creates a high-contrast image even for very low-contrast samples.



- Intelligent routines automatically identify your sample carrier, regardless if you use a petri dish, a chamber slide, or a multiwell plate. Carrier properties are automatically transferred to the software, eliminating manual settings.



- Your samples are reliably identified. Deep Learning algorithms precisely detect even unusual sample regions. You can navigate and access all sample areas directly which allows you starting your experiment faster than ever.

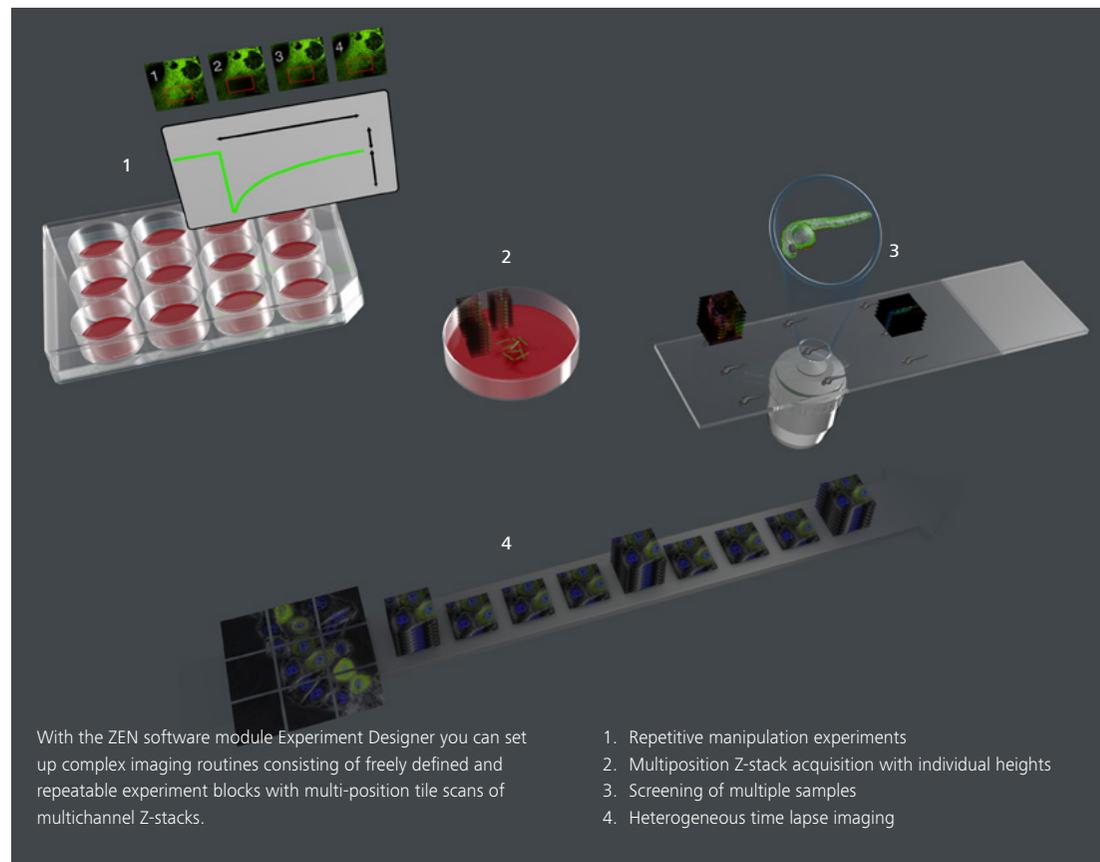
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### Acquire Reproducible Data with Ease

With all its various aspects and workflows, your research leaves you with no time to waste. That's why ZEN microscopy software was created—to make your confocal imaging both efficient and enjoyable.

Use Smart Setup to select your dyes and ZEN will automatically apply all necessary settings for all LSM imaging modalities. The integrated database with spectral data for more than 500 dyes helps you make an informed decision about your imaging options. You can always save imaging configurations or even whole experiments to reproduce settings quickly. The Reuse function allows you to extract and load imaging settings from the existing images. You will be amazed how easy imaging becomes when the AI Sample Finder automatically detects the sample carrier, adjusts the focus, and finds the sample regions relevant for your experiment. It takes less time to illuminate your sample and leaves you more of the precious time you've booked on the system for imaging. You can use the overview image to document all steps of your experiment and combine with other multimodal data or aspects of your sample. Sometimes your scientific questions will require complex acquisition strategies. Statistical analysis might call for repetitive imaging of a large number of samples with the same or even differing imaging conditions. Experiment Designer is



a powerful yet easy-to-use module that images multiple regions with all imaging modalities of your LSM 980.

It gives you access to a number of hardware and software options which will always keep your sample in focus, even during the most demanding long-term time-lapse experiments.

You can even view and save your valuable data during acquisition sessions to assess, analyze and react immediately.

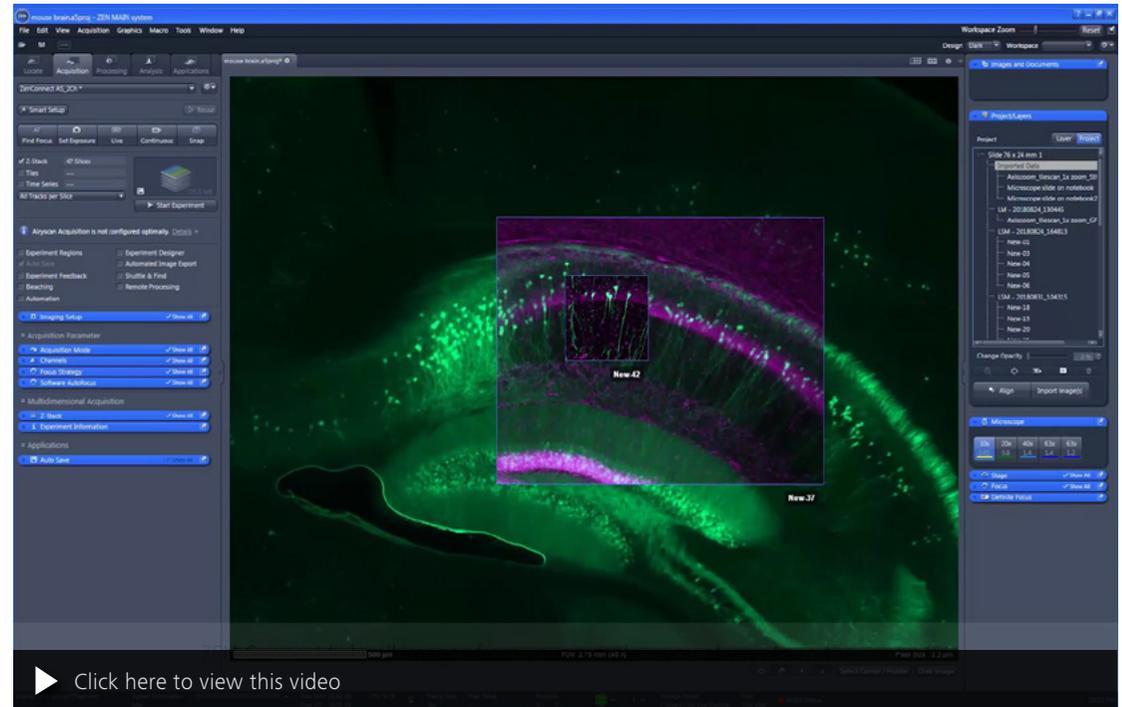
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## See More Details

Sometimes you need to see and assess your multimodal images during acquisition in order to plan your next steps. ZEN microscopy software gives you multiple options. You can sit at your connected computer to start the Direct Processing function for processing your Airyscan images during acquisition.

However, confocal imaging is only one part of the big picture, and you may need data from additional imaging modalities to complement the view on your sample. The Connect Toolkit can bring information from all your experiments together. Keep the context of your data by collecting all images of one experiment session in a single project in which you can combine overview and detailed high-resolution images, all perfectly aligned. Once you have created a project, you can always add and align content from any other imaging source, be it ZEISS, non-ZEISS or even cartoons and analysis graphs. You will stay on top of things at all times – both during your experiments and months or years later. Your ZEN Connect projects keep all associated datasets together. It's never been easier to share results and collaborate with others as a team.



*Connect all your imagery: With the Connect Toolkit you bring images and data from any system or modality together. You always keep the context and the overview about all data from your sample.*

The powerful integrated 3Dx1 Viewer, powered by ZEISS aravis, is optimized to render the large 3D and 4D image data you have acquired with the fast LSM 980. You can create impressive render-

ings and movies for meetings and conferences. After all, a good picture can say more than a thousand words.

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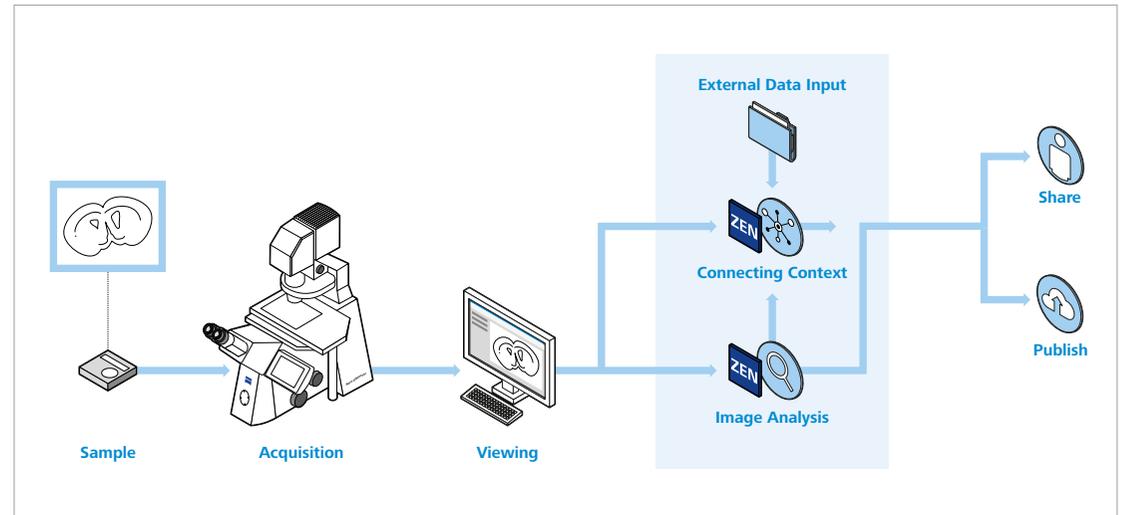
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## Get More Data from Your Sample

The real value of microscopy images is in the data they provide. The CZI file format of ZEN microscopy software makes sure that all important metadata of your experiments are safely stored and can be accessed openly for cross-platform data exchange. ZEN provides numerous analysis tools to extract all kinds of information from your images.

Building analysis workflows that adapt to specific applications is not an easy task. It requires knowledge of image processing and the ability to assemble a series of image operations. ZEN addresses this challenge with the BioApps modules for efficient image analysis. Each module is optimized for one type of application, e.g., cell counting or confluency measurement, with tailored segmentation settings and streamlined data presentation. If your applications require customized workflows, the wizard-based ZEN Image Analysis module will guide you step by step to create your unique measurements.

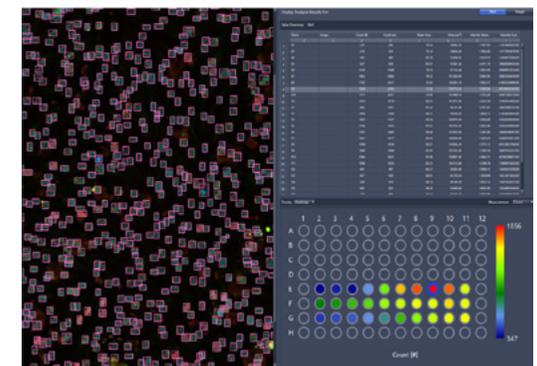
Within an image analysis workflow, segmentation and object classification are two of the most challenging steps. The AI Toolkit uses the latest machine learning algorithms to make these steps easier and more accurate, also allowing you to execute training on your own data sets. You can integrate the individual models seamlessly into your ZEN image analysis workflow.



ZEN microscopy software integrates all steps from your sample to reproducible data for publication.



AI Toolkit: Use the power of machine learning to easily segment your images.



Bio Apps Toolkit: From beautiful images to valuable data – analyze your images efficiently.

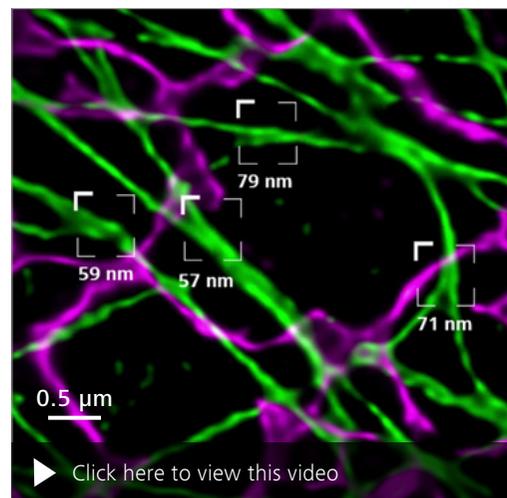
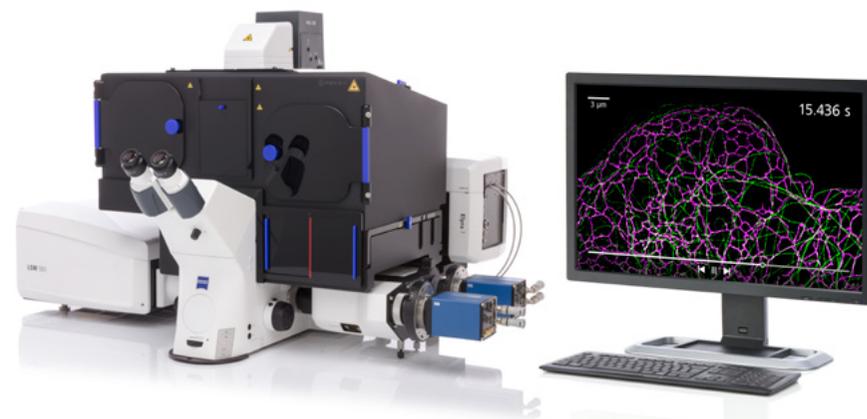
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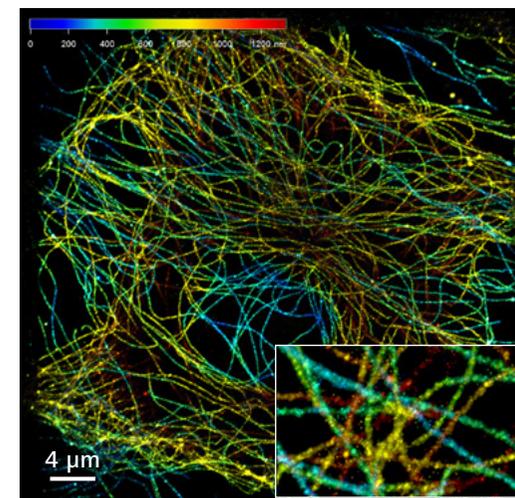
## Combine Multiple Super-resolution Techniques

Combine your LSM 980 with Elyra 7 and Lattice SIM<sup>2</sup> and you can always choose the best super-resolution technique for your experiment at hand. Lattice SIM<sup>2</sup> technology brings structured illumination microscopy (SIM) to a new level. Groundbreaking light efficiency gives you gentle live cell super-resolution imaging down to 60 nm resolution with incredibly high speed. With 6 different imaging modes to choose from, even lossless super-resolution imaging becomes possible, meaning you can now get one super-resolution image per acquired raw image for even gentler imaging of your most delicate samples. Add single molecule localization microscopy (SMLM) for techniques such as PALM, dSTORM and PAINT. You can now choose freely among your labels when imaging with resolutions down to 20 nm laterally and 50 nm axially. High power laser lines allow you to image your sample with ease, from green to far red.

Whether in an imaging facility or a single lab, your microscope users will appreciate the wealth of techniques for gentle 3D live cell imaging with super-resolution in one single system.



*Lattice SIM<sup>2</sup>: Simultaneous imaging of the endoplasmic reticulum (Calreticulin-tdTomato, magenta) and microtubules (EMTB-3xGFP, green) in a Cos-7 cell reveals highly dynamic interaction of these organelles at resolutions well below 100 nm. Objective: Plan-Apochromat 63x / 1.4 Oil*



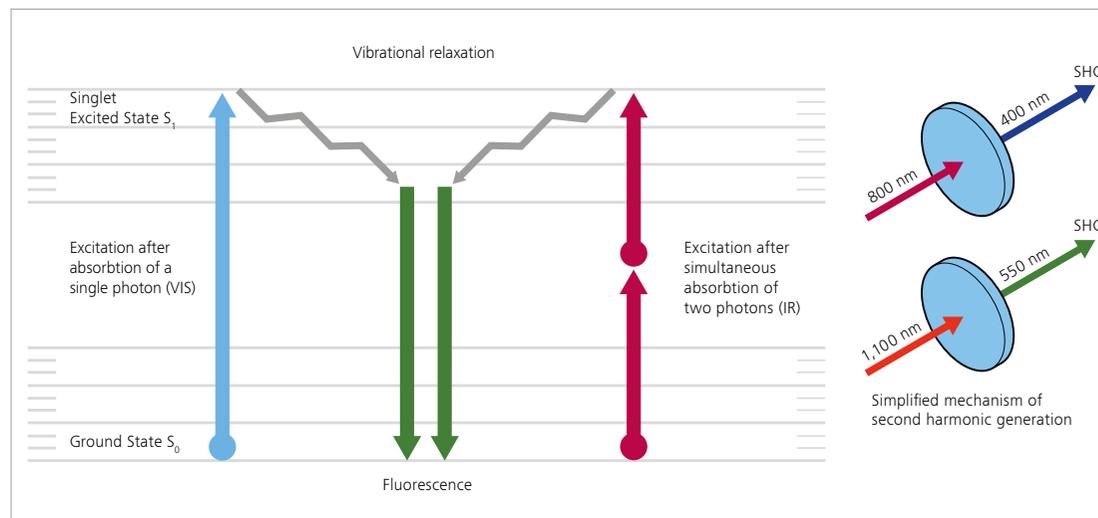
*SMLM: With ZEISS Elyra 7 you can image a z-depth of 1.4 μm in a single acquisition. 3D SMLM image of Alexa 647 α-tubulin color coded for depth. Sample courtesy of M. W. Davidson, Florida State University, USA*

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Multiphoton microscopy, also referred to as two-photon or non-linear optical (NLO) microscopy, is the preferred method for non-invasive, deep tissue imaging of living or fixed samples. It exploits one of the fundamental characteristics of light: longer wavelengths (600 – 1300 nm) are less absorbed and less scattered by tissues, traveling deeper into the sample while still forming a focal point. The energy required to excite a fluorescent dye is provided by not one photon but two photons, each with half the energy. Thus, the probability of two photons reaching the fluorophore at the same time is high only at the focal point. All emission light originates from the focal plane and can be directed to a non-descanned detector (NDD), generating an optical section while omitting a pinhole. For best signal transfer, sensitive GaAsP detectors can be placed in the transmission and reflection beam path, or even directly behind the objective lens. To further improve signal to noise, you can combine NDD imaging with LSM Plus.

An LSM that shares confocal and multiphoton capabilities allows you to access both technologies. Use multiphoton excitation with a fully open pinhole while using the confocal detectors.



Energy diagram of two-photon microscopy

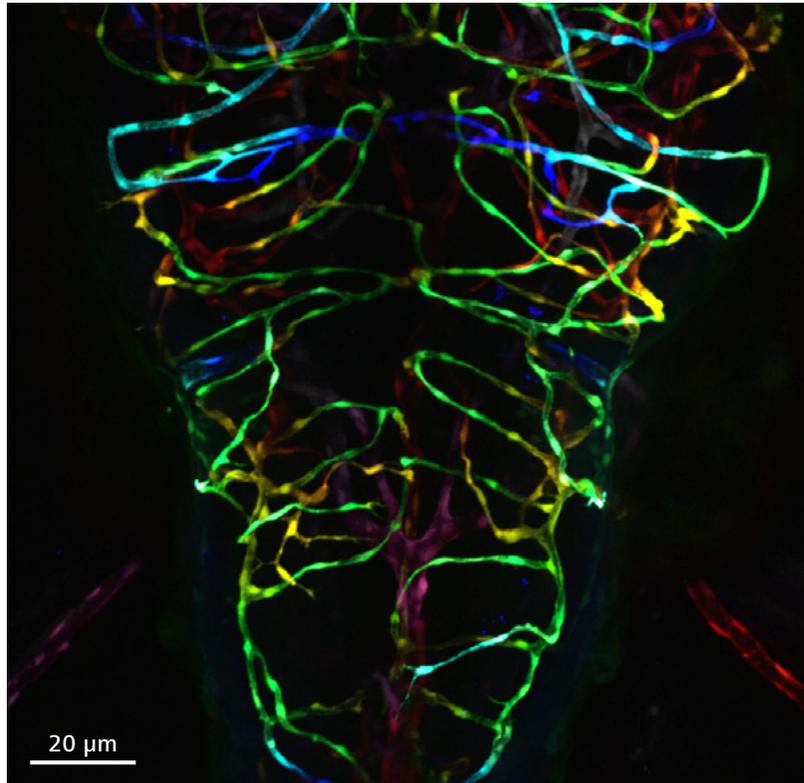
Add Airyscan 2 to combine deep tissue penetration with enhanced sensitivity, resolution, and speed—perfect for functional imaging experiments, large volume imaging, and screening applications. Several fluorophores can be excited with one or two independent NLO wavelengths, efficiently reducing the light exposure to the sample while speeding the acquisition process. Linear Unmixing of up to 7 non-descanned channels in

reflection or of a lambda scan utilizing the 34-channel Quasar detection allows a clear separation of all collected emission signals.

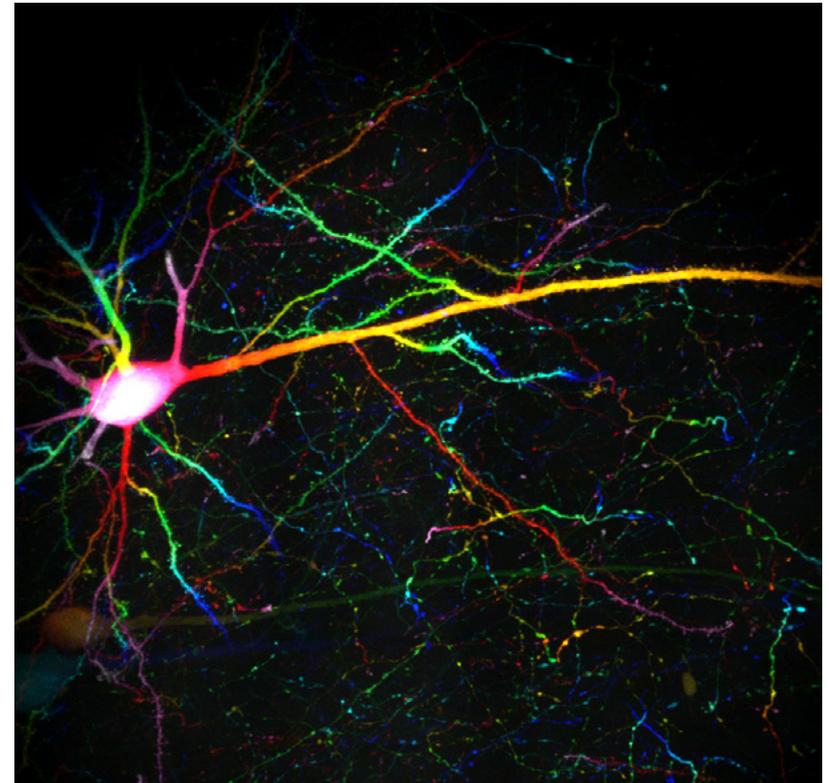
Even non-stained structures can be visualized with multiphoton excitation by second or third harmonic generation (SHG, THG). SHG effects occur on non-centrosymmetric molecules with predominantly periodic alignment, for example, in striated muscle and collagen.

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*Zebrafish hindbrain vasculature imaged in coronal orientation. Acquired with the two-photon laser excitation at 1,000 nm. The emission light was captured with the GaAsP BiG.2 non-descanned detector and processed with LSM Plus. Color coded 238  $\mu\text{m}$  z-stack.  
Sample courtesy of the Fish Facility, Leibniz-Institut für Alternsforschung – Fritz-Lipmann-Institut e.V. (FLI), Jena, Germany*



*Mouse brain slice with neuronal cytoplasmic GFP label. The 100  $\mu\text{m}$  volume was acquired with two-photon laser excitation at 1,000 nm with the GaAsP BiG.2 non-descanned detector. The dataset was colour coded for depth and an orthogonal projection was created with ZEN blue.  
Sample courtesy of Prof. J. Herms, LMU, Munich, Germany*

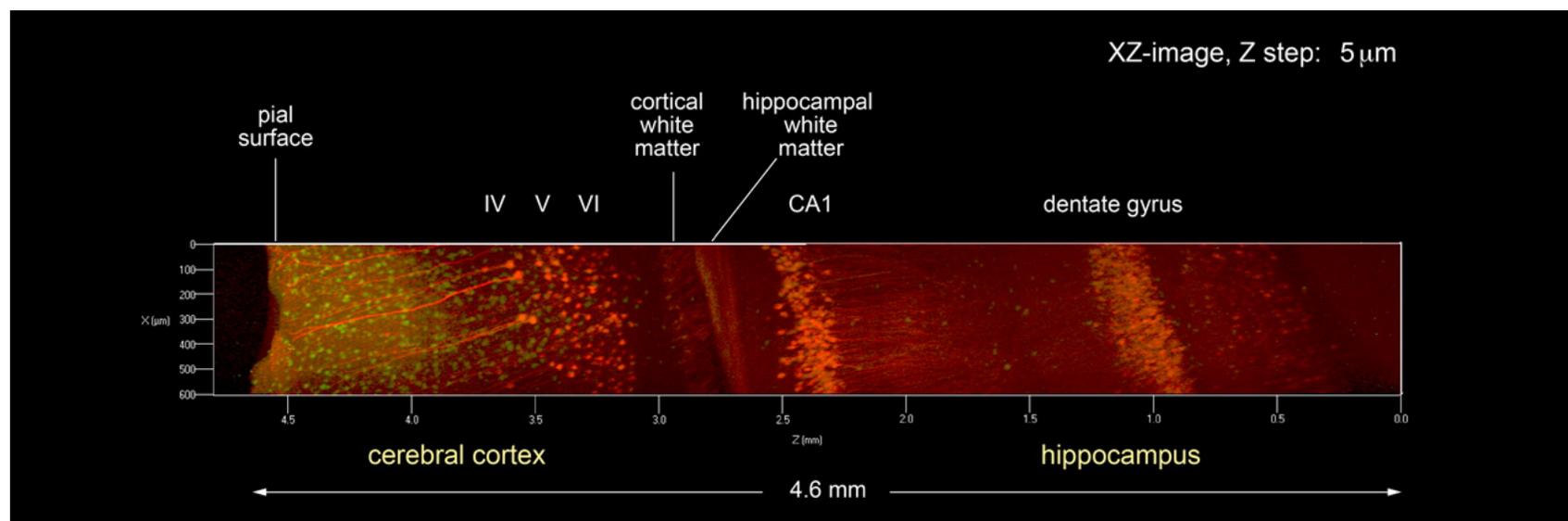
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## Image Large Cleared Samples

Tissue clearing opens up a new dimension of optical penetration depth into biological samples such as tissue sections, mouse brains, embryos, organs, spheroids or biopsies.

With Axio Examiner and special objectives—for example, Clr Plan-Apochromat 10x/0.5 nd=1.38, Clr Plan-Apochromat 20x/1.0 Corr nd=1.38 or Clr Plan-Neofluar 20x/1.0 Corr nd=1.45—you can look up to 5.6 mm deep into tissue that has been treated with clearing agents such as Focus Clear or Scale. The cleared tissue becomes almost transparent and the objectives provide the matching refractive index to the immersion medium, delivering crisp contrast. You can now image up to six times deeper than with a multiphoton microscope and up to 60 times deeper than with a conventional laser scanning microscope on uncleared samples. Get ready to be impressed by the quality of structural information you will retrieve from the deepest layers: expect a big push forward.



Maximum intensity projection, brain of 7-week old YFP-H mouse, fixed and cleared with Scale clearing technique (Hama et al, Nat Neurosci. 2011).  
Courtesy of H. Hama, F. Ishidate, A. Miyawaki, RIKEN BSI, Wako, Japan

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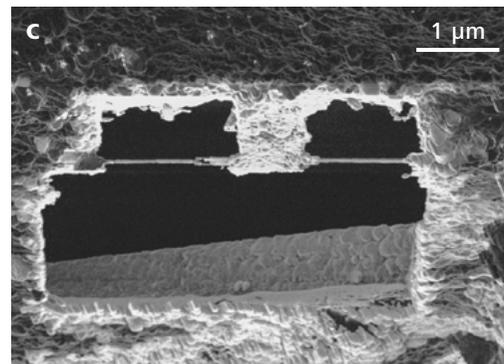
## Correlative Cryo Microscopy: Image the Near-to-native State

Spindle pole bodies are difficult to localize within yeast cells. They are small and rarely occurring structures. ZEISS Correlative Cryo Workflow lets you precisely identify and image such cellular structures in the near-to-native state.

The LSM with the Airyscan detector makes the identification of these structures even easier so further details can be imaged. All images – from a large overview of the entire cell to high-resolution images of these tiny structures – are organized in a ZEN Connect project, providing all data needed to re-locate these cellular structures in the FIB-SEM.

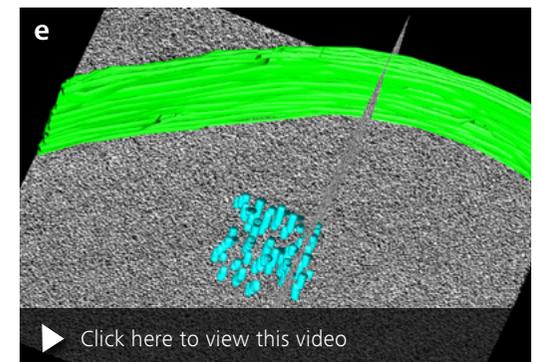
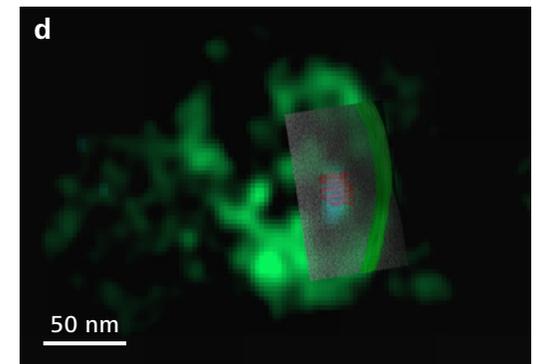
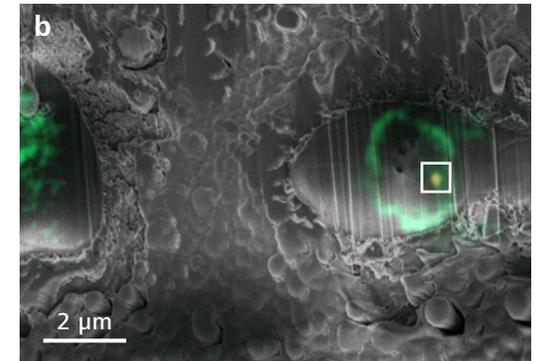
Using ZEISS Crossbeam, TEM lamella of the identified regions can be prepared for cryo electron tomography. Volume imaging is possible as well. Furthermore, the workflow solution allows you to reconnect all data after image acquisition. Images from the Crossbeam or tomograms from the TEM can be combined with the LSM data and can be rendered in three-dimensional context.

Learn more about ZEISS Correlative Cryo Workflow: [www.zeiss.com/cryo](http://www.zeiss.com/cryo)



Yeast cells labeled with NUP (nuclear pore complex)-GFP and CNM67-tdTomato. Sample and tomogram courtesy of M. Pilhofer, ETH Zürich, Switzerland

- ZEN Connect movie shows the overlay of an LM and EM dataset – from the grid overview to the region of interest identified for further TEM tomography.
- Early state of the milling process: Lamella is prepared around the marked region which was identified at the LSM.
- FIB image of the prepared lamella; lamella thickness: 230 nm
- 3D overlay of the reconstructed and segmented tomogram with LSM dataset (Spindle pole body is false-colored in cyan); nuclear membrane and microtubules were segmented using IMOD.
- Segmented and reconstructed tomogram



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As your needs grow, your LSM 980 grows with you, forming the basis for a number of enhancements. Like every system from ZEISS, open interfaces and a modular architecture guarantee the seamless interaction of all components now and in the future. These include:



*Combine your ZEISS Axio Observer 7 with integrated incubation modules to create the perfect environment for long-term live cell imaging with stable temperature conditions.*



*The upright fixed stage microscope ZEISS Axio Examiner.Z1 gives you ample specimen space and room for imaging of whole animals. This stable stand is ideally suited for your demanding multiphoton experiments with incubation for living specimens.*



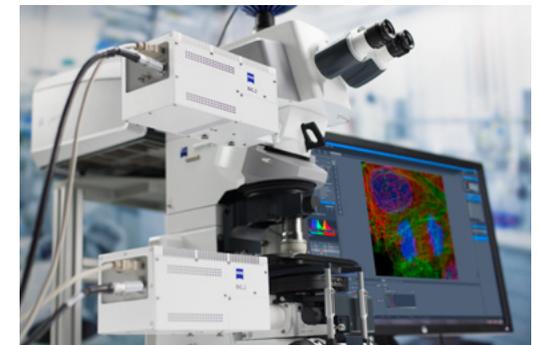
*AI Sample Finder automatically detects your sample carrier, adjusts the focus, and finds your sample regions on the coverslip. Even with low-contrast samples, you will access relevant regions with just a click and start your experiment right away.*



*Enhance your microscope with ZEISS Colibri 7. This flexible and efficient LED light source allows to screen and image your delicate fluorescent samples very gently. You profit from stable illumination and extremely long lamp life.*



*Add the BiG.2 module with its two GaAsP detectors for FCS, photon counting experiments and FLIM\* to your ZEISS LSM 980.*

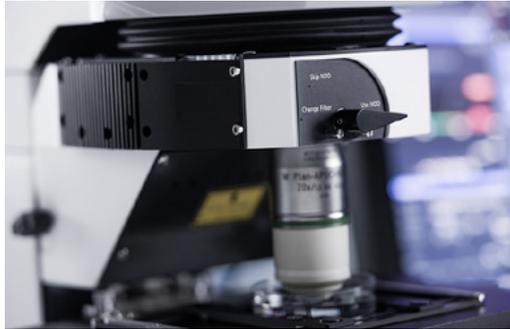


*Your BiG.2 works perfectly as a non-descanned detector, also providing a highly sensitive direct coupled detector for FLIM\*.*

\* available upon request

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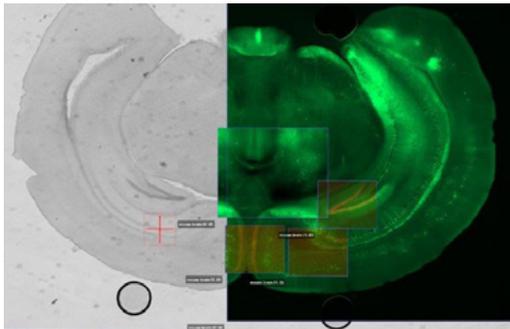
*The module GaAsP NDD 2 channels with flexible filter settings completes the ensemble of non-descanned detectors for ZEISS Axio Examiner.Z1.*



*With autocorr objectives and ZEN microscopy software it's easy to adjust your microscope optics to your sample. You get crisp contrast and better signal to noise – even in your most challenging samples.*



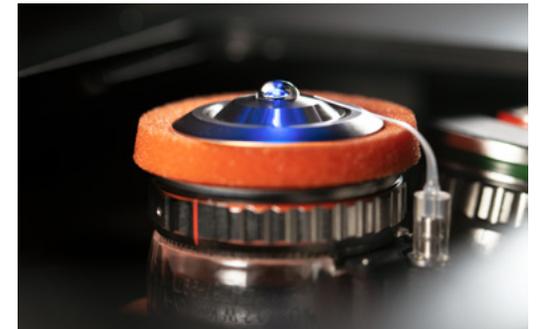
*You can add a choice of sensitive ZEISS Axiocams to your ZEISS LSM 980. It's very easy to acquire overview images for your multiposition experiments or to perform light efficient widefield imaging.*



*ZEN Connect 2D and 3D Add-on is your gateway to correlative light and electron microscopy (CLEM). Combine the specificity of functional fluorescence imaging with ultrastructural information.*



*Definite Focus 3 compensates Z-drift and stabilizes the focal position of your sample. You can now perform long-term multiposition and tiling experiments that can last for multiple days.*



*The Autoimmersion Module automates the application of immersion media for water immersion objectives. The immersion media is applied while maintaining objective focus and position, leaving your experiments undisturbed.*

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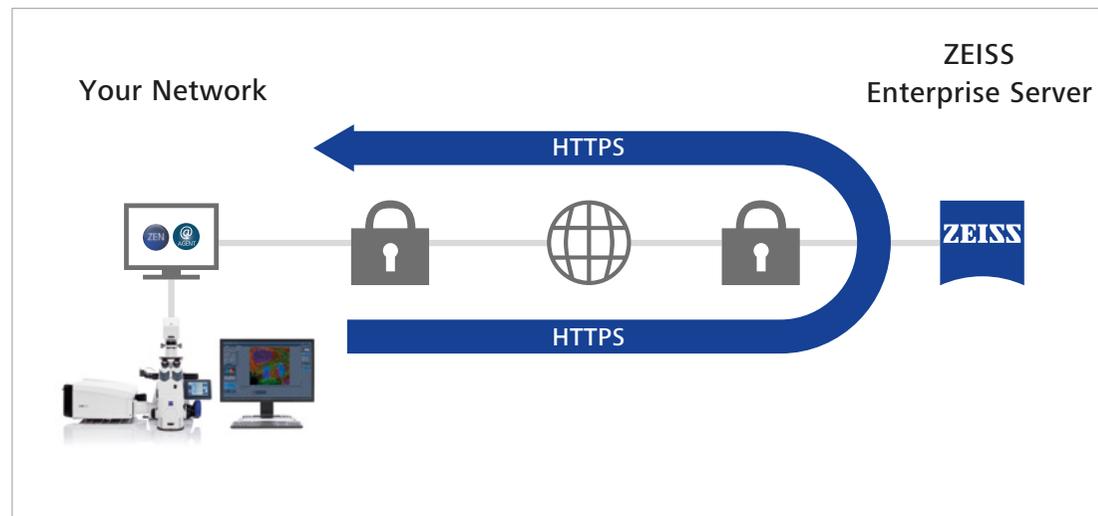
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## **ZEISS Predictive Service Maximizes System Uptime**

Once connected to your network and activated, this advanced technology will automatically track the health status of your instrument and collect system log files in the background to improve remote diagnosis.

Relevant technical data such as operating hours, cycle counts or voltages are periodically monitored via a secure connection to our data center. The ZEISS Predictive Service application evaluates the performance of your microscope as system data can be received and analyzed.

Our support engineers will diagnose any issues by analyzing data on the Enterprise Server – remotely and without interruption to your operation.



### ■ **Maintain highest system availability**

Increase your uptime through close monitoring of the system's condition as remote support can often provide immediate solutions.

### ■ **Data security**

Ensure highest data security standards using well established technologies like PTC Thingworx and Microsoft Azure Cloud. No personal or image data is uploaded, only machine data.

### ■ **Fast and competent support**

Use secure remote desktop sharing to easily get an expert connected.

### ■ **Optimum instrument performance**

As the status of your system is monitored, necessary actions can be planned before they become urgent.

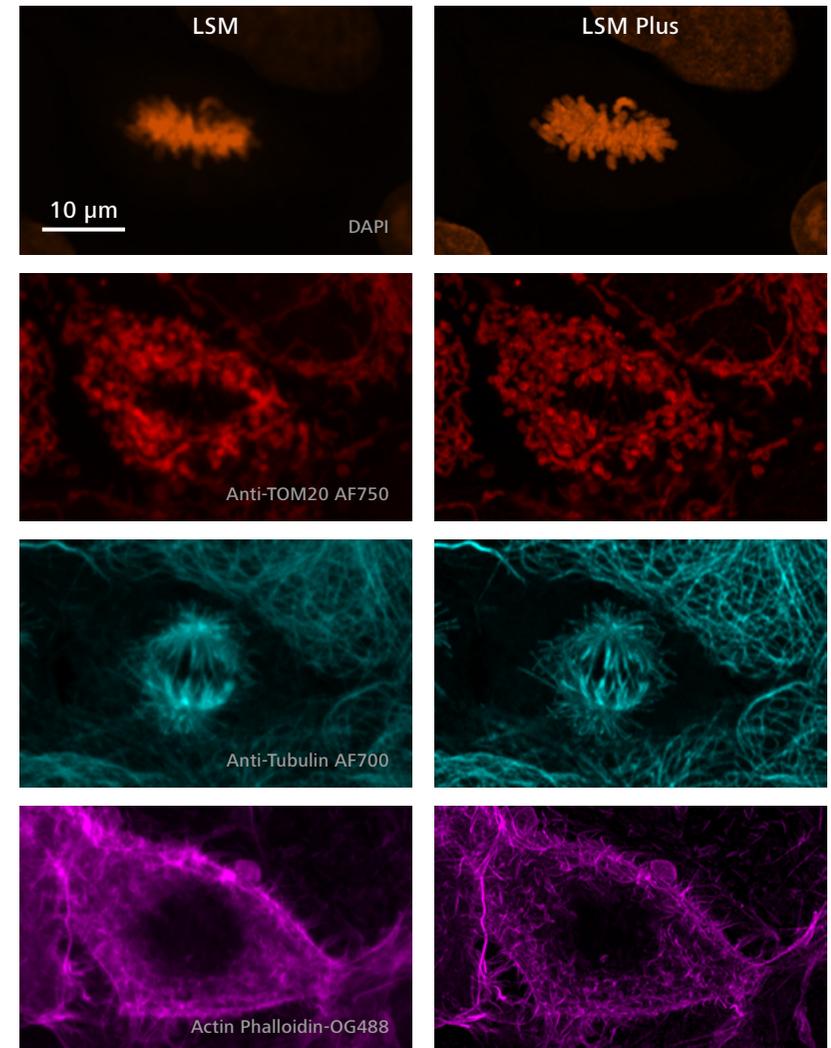
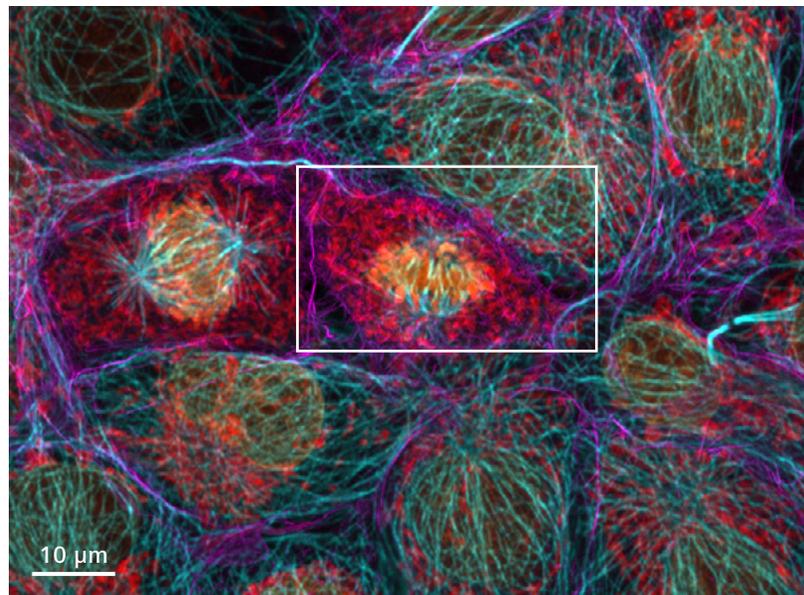
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## Expand the Number of Labels

To capture the complex world of biology, the ability to expand the number of labels is a great advantage. LSM 980 can image multiple labels simultaneously, covering a wide emission range up to 900 nm, making it the perfect tool for spectral multiplex experiments.

These Cos 7 cells were labelled with 4 different fluorophores, two of which have their emission peak in the near infrared range (NIR), Alexa 700 and Alexa 750. Utilizing the flexible LSM 980 Quasar and NIR detectors, all labels were imaged with optimal sensitivity. The zoom-in views illustrate how LSM Plus improves SNR and resolution.



Cos-7 cells Anti-TOM20 AF750 (red), Anti-Tubulin AF700 (cyan), Actin Phalloidin-OG488 (magenta), DAPI (orange). Imaged with LSM 980 with LSM Plus, including the ZEISS NIR detector in channel mode. The fluorescent signals were separated by Linear Unmixing, facilitating clear separation between the spectrally overlapping dyes Alexa 700 and Alexa 750. Sample courtesy of U. Ziegler and J. Doehner, University of Zurich, ZMB, Switzerland.

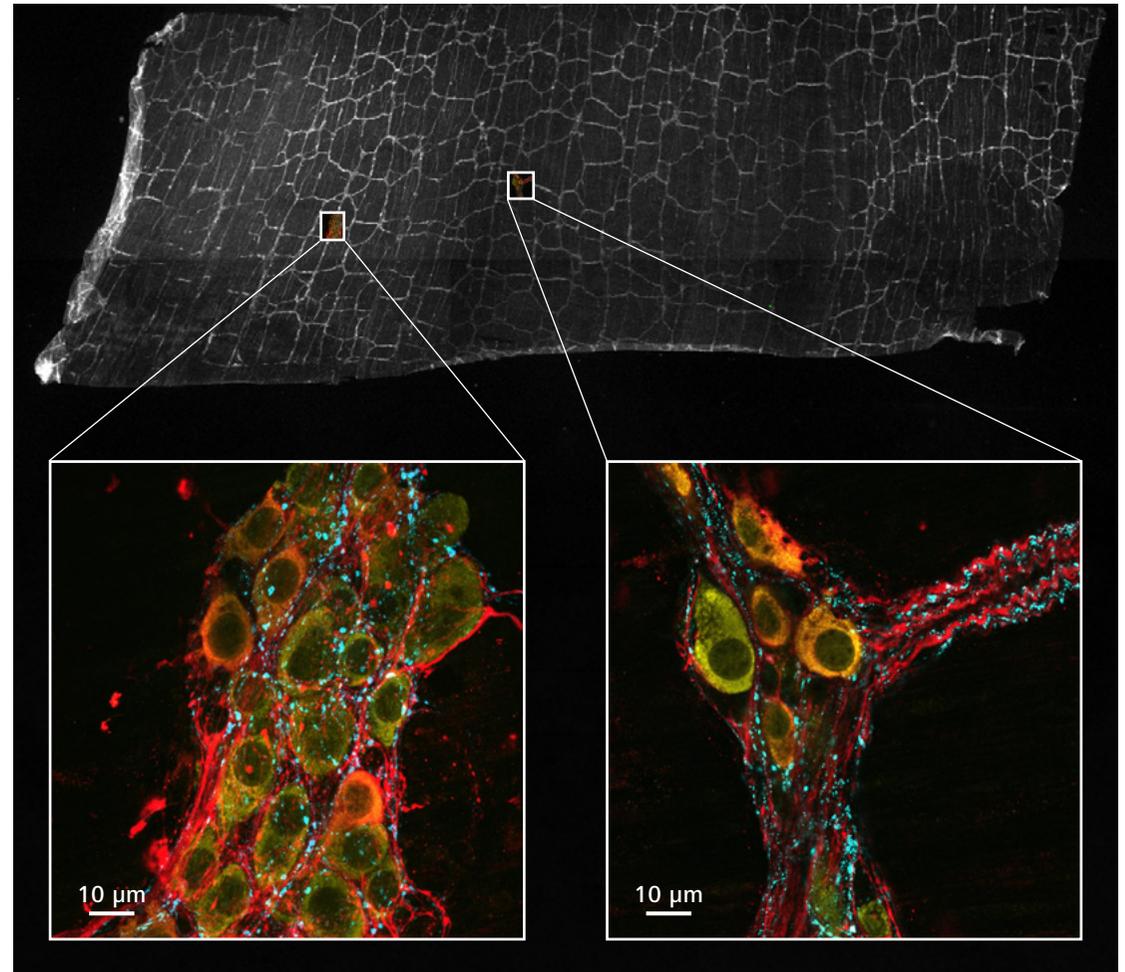
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## Navigate and Correlate with Ease

As the world of microscopy transitions gradually to larger samples, it becomes more important to maintain the positional context and keep a record of the areas captured. AI Sample Finder automatically classifies the sample carrier, identifies the sample, finds the focus, and creates a fast overview image using the T-PMT detector or camera. You can freely navigate using the overview image for orientation, and effortlessly move to the structures of interest. Making sure you only spend time imaging regions that hold information for your research. With the Connect Toolkit, you can correlate all data associated with the sample.

In this example, mouse intestinal tissue was labelled with three fluorophores covering an emission spectrum of 500 – 850 nm. AI Sample Finder automatically identified the carrier and created an overview image using the T-PMT to capture the Alexa 488 label. The overview image is used for sample navigation and identification of regions of interest. The ZEISS LSM 980 Quasar and NIR detectors were used to acquire images of the visible and invisible labels with optimal sensitivity.

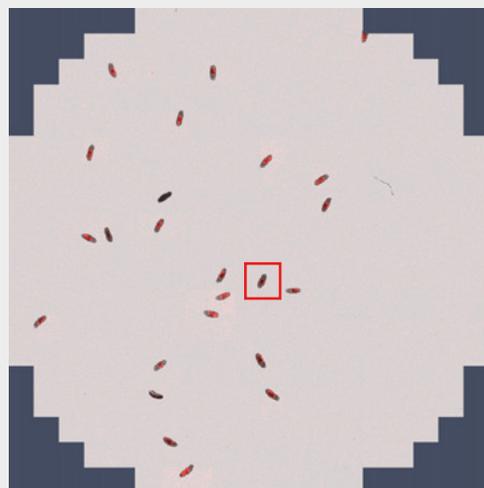


*Mouse intestine tissue section stained for Substance P (cyan, Alexa 488) labeling the presynaptic contacts in the enteric nervous system, HuC/D (yellow, Alexa 568) labeling the enteric neurons, and neuronal Nitric Oxide Synthase (nNOS, red, Alexa 750) labeling a sub-population of enteric neurons. Sample Courtesy of P. Vanden Berghe, LENS & CIC, University of Leuven, Belgium*

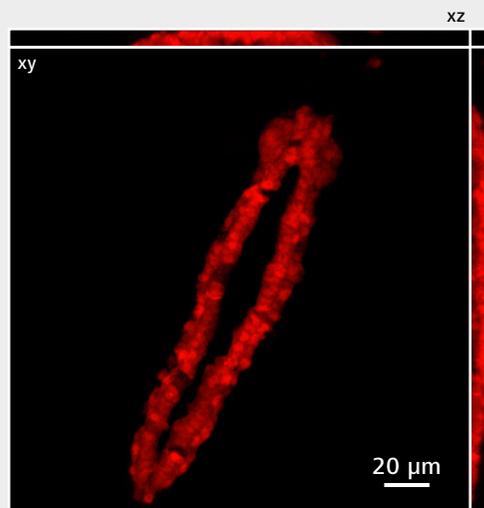
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## Live cell experiment



Overview scan

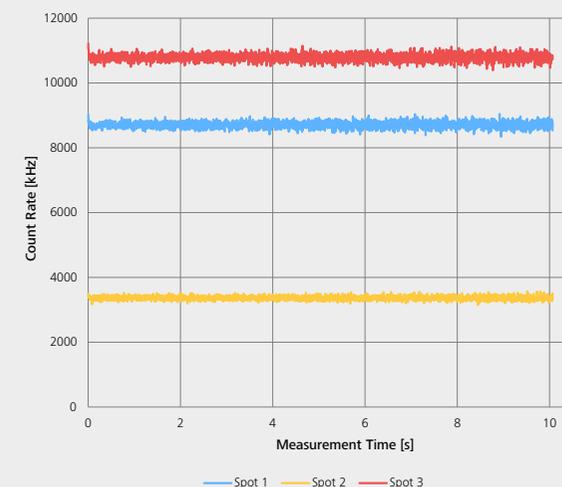
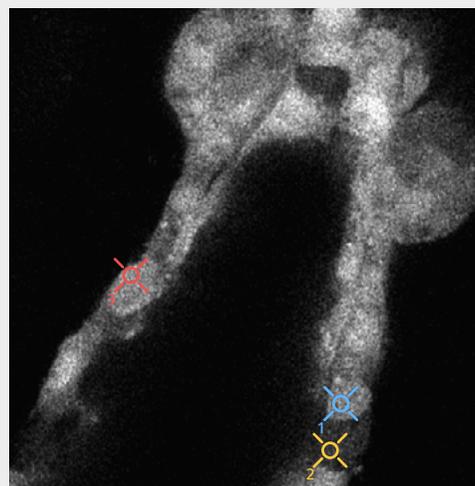


Z-stack with Airyscan 2 – Ortho Maximum Intensity Projection

**Dynamics Profiler** – Easily gain more information in your current imaging experiments. Fluorescent protein dynamics and concentration measurements can effortlessly be added to a confocal experiment. Dynamics Profiler uniquely allows these measurements even in bright and challenging samples. The sample shows a *Drosophila melanogaster* embryo expressing mCherry under control of the hand cardiac and hematopoietic enhancer (Han and Olson, 2005), located in the third intron. The expression of the reporter mimics endogenous hand expression in the heart and is maintained throughout embryogenesis in cardioblasts as well as in pericardial cells.

Sample Courtesy of Prof. Dr. Achim Paululat and Dr. Christian Meyer, Osnabrück University, Department of Zoology and Developmental Biology, Germany.

## Dynamics and concentration measurements

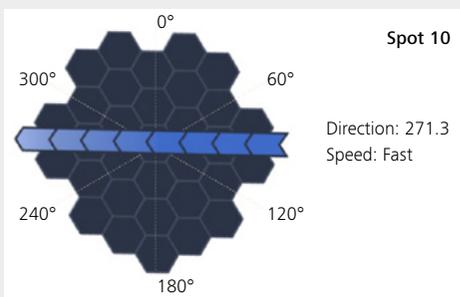
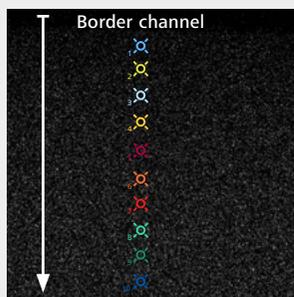


|        | Count Rate [kHz] | Concentration [nM] | Diffusion Coefficient [ $\mu\text{m}^2/\text{s}$ ] |
|--------|------------------|--------------------|--|
| Spot 1 | 8707.5           | 37263.1            | 242.9  |
| Spot 2 | 3365.5           | 13666.4            | 279.4  |
| Spot 3 | 10788.0          | 41997.8            | 239.9  |

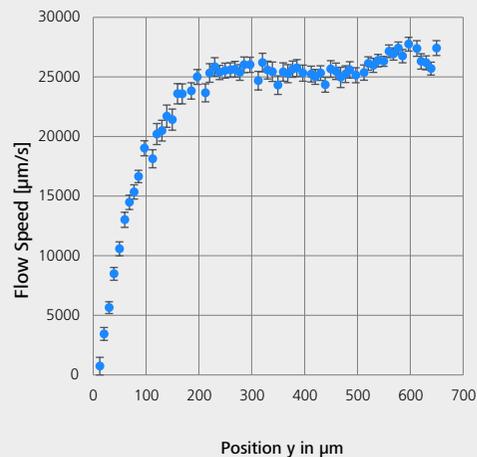
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## Flow

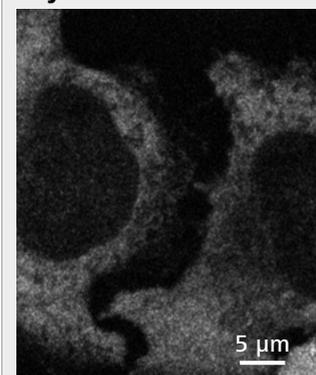


| ID | Flow Direction [°] | Flow Speed [ $\mu\text{m/s}$ ] |
|----|--------------------|--------------------------------|
| 1  | 315.62             | 740.00                         |
| 2  | 262.60             | 3441.79                        |
| 3  | 269.25             | 5647.59                        |
| 4  | 271.01             | 8480.16                        |
| 5  | 272.89             | 10579.27                       |
| 6  | 267.30             | 13014.14                       |
| 7  | 272.24             | 14479.63                       |
| 8  | 271.49             | 15351.73                       |
| 9  | 271.49             | 16648.21                       |
| 10 | 271.28             | 19016.89                       |

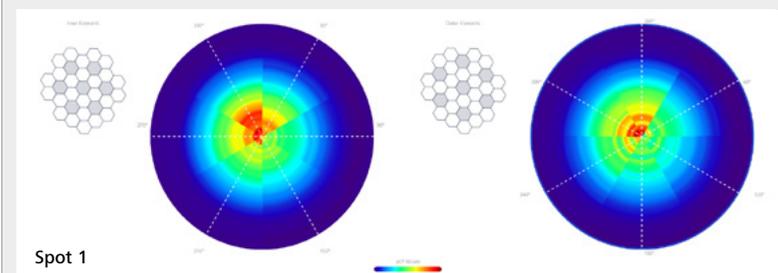
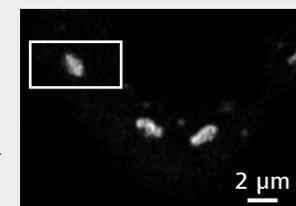


**Dynamics Profiler** – Spatial information collected with ZEISS Airyscan enables flow measurements that provide unique new data related to microfluidics. The Automated Sequential Injection System Aria by Fluigent was used to pump a rhodamine 110 solution through a microfluidic flow cell (1000  $\mu\text{m}$  channel width). Flow speed and direction of active movement can be determined for defined spots across a microfluidics channel. A reference image helps to position the spots and to orient within the sample. Laminar flow within such a channel can thus be characterized.

## Asymmetric Diffusion Behavior



Heat Shock



**Dynamics Profiler** – Spatial information collected with ZEISS Airyscan lets you characterize heterogenous diffusion behavior, ideal to investigate cellular condensates that form by liquid-liquid phase separation. A reference image helps to orient within the sample and to position the spots that indicate the actual area analyzed. This example shows GFP-labeled stress granules in HeLa cells.

*Sample courtesy of Dr. V. Bader und Prof. Dr. K. Winklhofer, Institut für Biochemie und Pathobiochemie, Ruhr-Universität Bochum, Germany.*

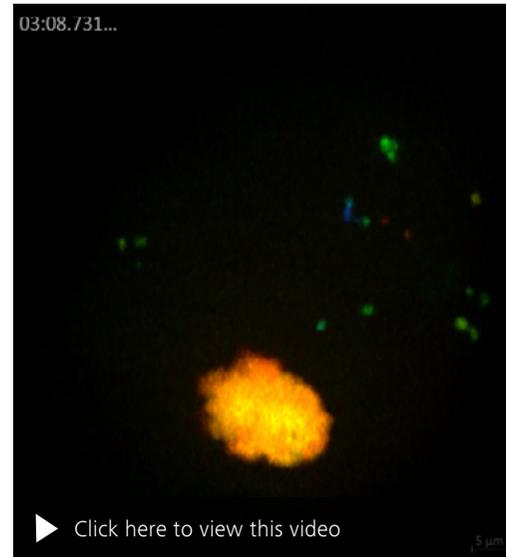
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Oocytes store all the nutrients to support early embryonic development, and are therefore very large cells with a large nucleus. Oocytes need to divide before fertilization. How to make cell division work in this very large cell is the topic investigated by P. Lenart's lab.

They have shown that, surprisingly, an actin network is required to collect chromosomes scattered in the oocyte nucleus. They are then handed over to microtubules, which capture chromosomes and align them on the spindle. The actin-driven and microtubule-driven transport phases have very different speeds and show other differentiating characteristics that can be distinguished by tracking chromosome motion.

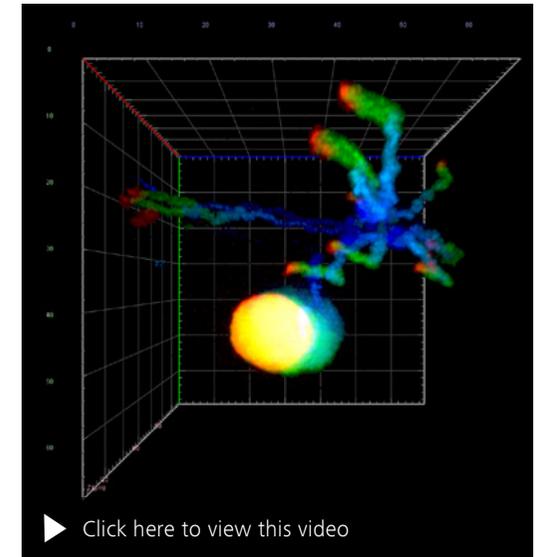
Peter Lenart says: "This is a nice imaging challenge, because chromosomes are scattered in the spherical nucleus with a diameter of 80  $\mu\text{m}$  and are transported over a period of approximately 15 minutes. Back in 2005 we could acquire stacks every 45 s, which was sufficient to distinguish actin- and microtubule-driven phases. Using the new, high resolution trajectories shown here we hope to learn about the details of the transport mechanism."



*Meiosis in starfish oocytes*

*The depth coding shows a subset of 52  $\mu\text{m}$ . The movie shows the transport of chromosomes, labeled by Histone 1-Alexa 568, in a starfish oocyte undergoing meiosis.*

*A z-stack of 67  $\mu\text{m}$  was acquired every 2.4 seconds with Airyscan CO-8Y mode. Concomitant with chromosome transport, the nucleolus (the large spherical structure) is disassembling. Courtesy of P. Lenart, MPI for Biophysical Chemistry, Göttingen, Germany*



*Meiosis in starfish oocytes*

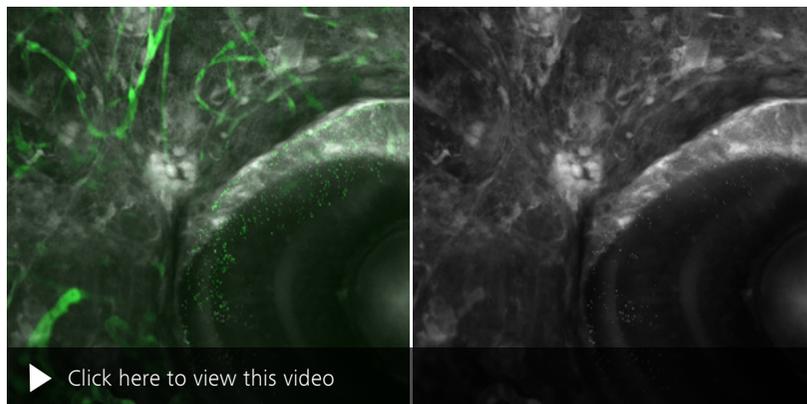
*The rendering is a projection of the process along z-axis (maximum intensity) and time (color-coded projection); to illustrate the movement of the chromosomes within the volume of the nucleus.*

## Reference:

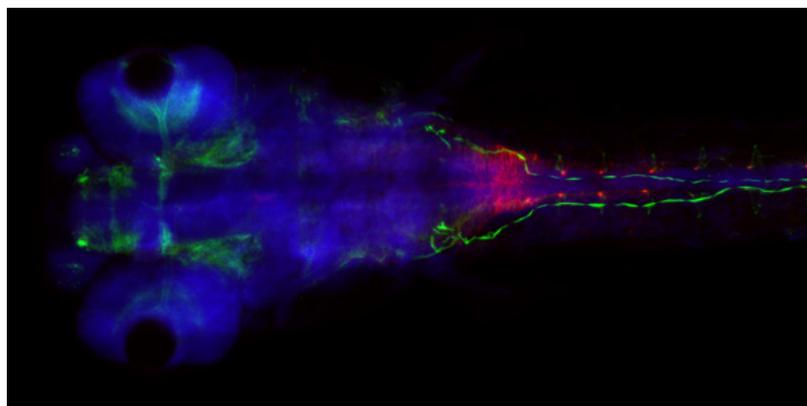
- Lenart P, et al. Nature. 2005 Aug 11;436(7052):812-8.
- Mori M, et al. Curr Biol. 2011 Apr 12;21(7):606-11.
- Bun P, et al. Elife. 2018 Jan 19;7. pii: e31469. doi:10.7554/eLife.31469.
- Burdykiuk M, et al. J Cell Biol. 2018 Aug 6;217(8):2661-2674.

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*Zebrafish brain and eye vasculature (green) and Second Harmonic Generation (grey) in sagittal orientation. A volume of 267  $\mu\text{m}$  was acquired with the two-photon laser at 1,000 nm and emission was detected with the GaAsP BIG.2 detector. SHG allowed the visualization of the tissue structures, such as the retinal cells and ocular muscles. Sample courtesy of the Fish Facility, Leibniz-Institut für Alternsforschung – Fritz-Lipmann-Institut e.V. (FLI), Jena, Germany*



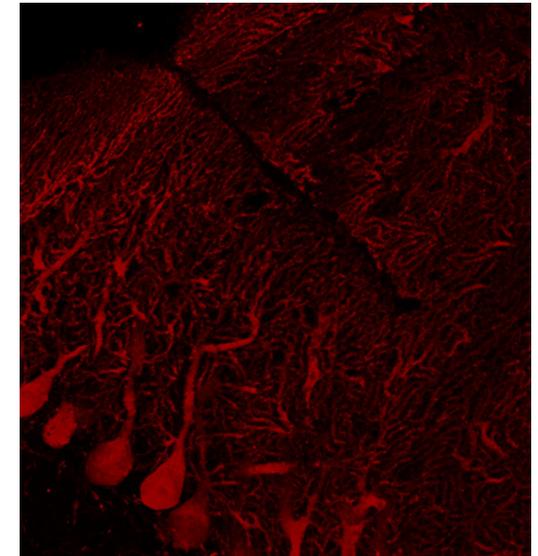
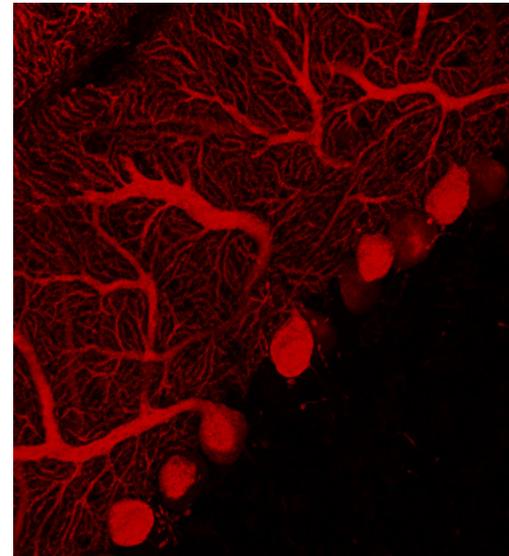
*Zebrafish larvae immunolabelled for nervous system (Alexa-488), cell nuclei (DAPI), acetylated tubulin (Alexa-633). All three dyes were excited with the two-photon laser at 780 nm and emission was detected with a lambda stack in 3D Tiling (157  $\mu\text{m}$ , 15 tiles). The fluorophore spectra were separated with Linear Unmixing and the 3D tiles were Stitched in ZEN Blue. This image is the orthogonal projection of the 3D dataset. Sample courtesy of Dr. H. Reuter, Molecular Genetics Group, Leibniz-Institut für Alternsforschung – Fritz-Lipmann-Institut e.V. (FLI), Jena, Germany*

Zebrafish is a well established model for studying development of the vascular system. Multiphoton imaging is a great way to capture the intricate vasculature patterns in zebrafish brain at great depth. Additionally, through the Second Harmonic Generation (SHG), structural information of the surrounding tissues can be captured without the need of additional labelling.

To minimize acquisition time and photobleaching it is useful that you can excite multiple fluorophores with a single two-photon laserline, allowing you to image multiple labels simultaneously. With LSM 980 and the 32-Channel Quasar detector you can perform lambda scans, collecting and unmixing the emitted signals over the whole visible spectrum. In this example, all 3 labels (Alexa-488, Alexa-633 and DAPI) were excited and detected simultaneously. 3D Tiling and Stitching was used to capture the large sample volume, maintaining the context in the tissue. LSM 980 offers you the best of both worlds, allowing you to combine the strength of the NLO excitation with the advantages of spectral detection, and all the imaging modalities of ZEN blue.

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*Mouse brain cerebellum labelled with anti-calbindin (Alexa-568) and anti-GFAP (Alexa-488). The fluorophores were both excited with the two-photon laser at 780 nm and the emission spectra were simultaneously collected by the BIG.2 detector. 3D Tiling and Stitching were used to cover whole structure, and an orthogonal projection was created in ZEN Blue. Specific areas of interest were imaged with the Airyscan 2 detector in order to acquire high resolution images of the Purkinje cells. The Airyscan 2 datasets were processed and orthogonal projections were created with ZEN Blue. The individual super-resolution images were aligned with the cerebellum using ZEN Connect. Sample courtesy of L. Cortes, University of Coimbra, Portugal*

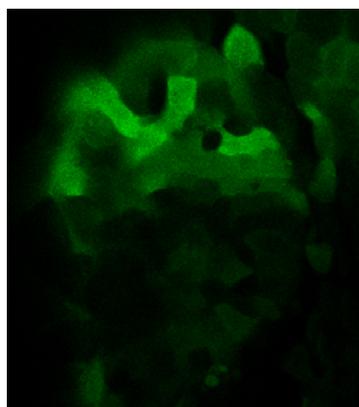
Multiphoton microscopy can be combined with 3D Tiling and Stitching in order to image large samples, such as this example of mouse cerebellum. Airyscan 2 imaging in super-resolution mode

can be used to acquire super-resolution images of specific areas of interest, and can be seamlessly combined with two-photon imaging. ZEN Connect can bring all the information from your

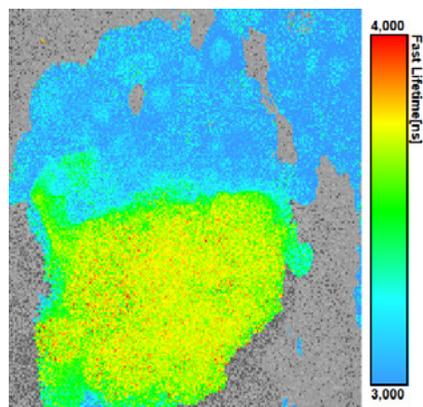
different experiments together, allowing you to map the high-resolution images on the larger structure, maintaining the context and simplifying your file organization.

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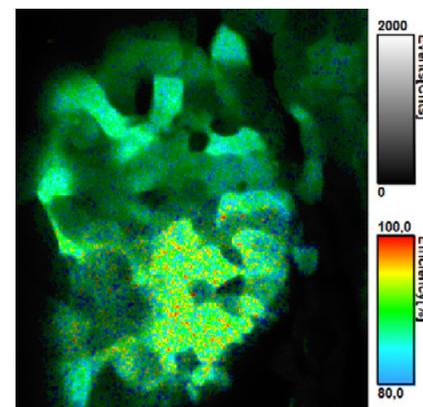
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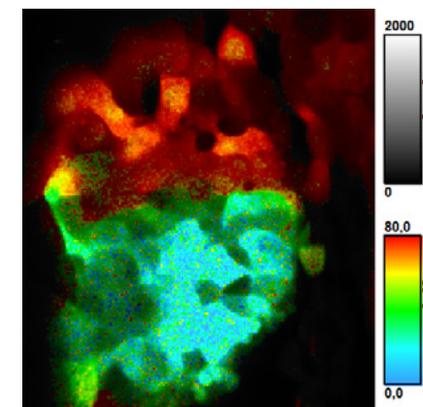
A) YFP intensity (bottom photobleached)



B) Donor / CFP lifetime (bottom photobleached)



C) FRET efficiency based on Donor lifetime (bottom photobleached)



D) Binding between CFP and YFP based on Donor lifetime (bottom photobleached)

## Fluorescence Lifetime Imaging (FLIM)

The lab of Marcos Gonzalez-Gaitan is investigating the role of small GTPase during Zebrafish embryonic development. The focus of their work lies in identifying when and where these GTPases are active during the oriented division of ectodermal progenitor (epiblast) cells. This activity can be monitored by using Förster Resonance Energy Transfer (FRET), in which energy transfer from one chromophore (Donor) to another (Acceptor) only occurs when the two chromophores are closer than <math><10\text{ nm}</math>. By measuring the fluorescent lifetime of the Donor (FLIM-FRET), relevant information can be collected.

In this example, the small GTPase Rac protein was fused to variants of CFP and YFP, as an

intramolecular FRET-pair biosensor to monitor GTPase activity. When the acceptor fluorophore is bleached (Fig. A: lower region of the image), the lifetime of the Donor fluorophore is increased in the same region (Fig. B). The FRET Efficiency is not influenced by the bleached Acceptor fluorophore and stays unchanged for the remaining FRET-pairs (Fig. C). Additional information is given in the Binding fraction (Fig. D), which holds quantitative spatial and temporal information of the currently active FRET pairs.

Quantitative FLIM-FRET analysis allows determining the spatial and temporal activity of two or more interacting molecules. In contrast to measuring FRET by photobleaching or intensity ratio

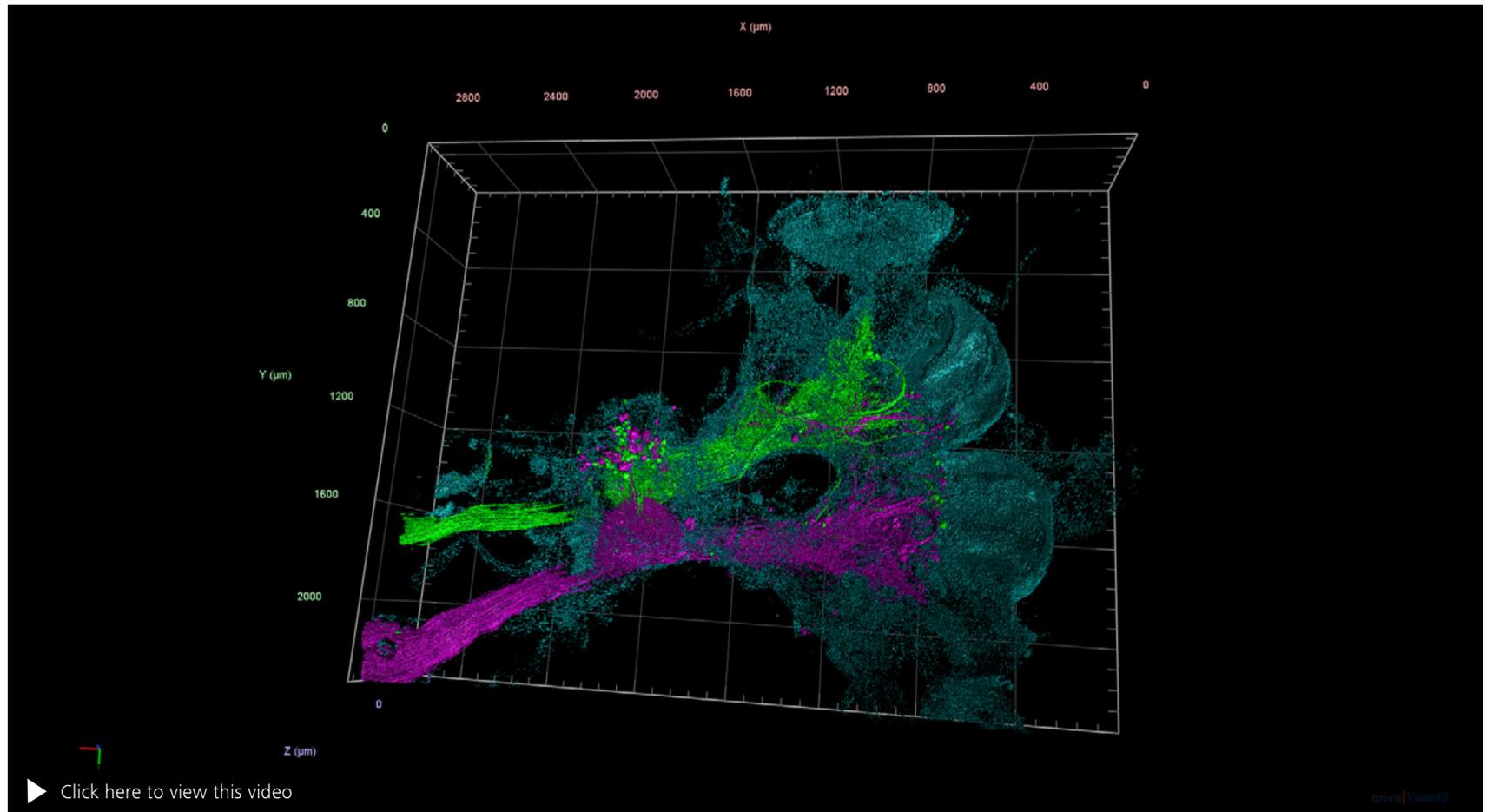
imaging, lifetime imaging allows precise quantification of FRET Efficiency. In addition, FLIM-FRET is used to quantify the binding fraction for a particular intermolecular FRET pair and the fraction of active sensors for an intramolecular FRET biosensor when using a suited FRET pair.

Data was obtained with LSM Systems equipped with PicoQuant FLIM\* & FCS kit and the PicoQuant FLIM module in ZEN microscopy software. Multiphoton excitation at 840 nm was used for lifetime measurements of CFP. Acceptor photobleaching was performed with 514 nm laser. The analysis was done within PicoQuant's SymPhoTime64.

\*available upon request

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The brain, thoracic and abdominal ganglia of the cockroach are joined together by bilateral connective bundles of ascending and descending interneurons forming the ventral nerve cord. In this preparation, left and right connectives were individually labelled (Alexa 488: green, Alexa 647: magenta) posteriorly to the suboesophageal ganglion to observe the extension of their innervation within the different neurophilis, and throughout the ipsi- and contralateral parts of the brain (DNA labelled with DAPI: cyan). Imaging was performed using Tiling and Stitching to capture the complete volume ( $3 \times 2.3 \times 0.26$  mm). 3D animation of the complete dataset was done with ZEISS arivis Pro, ideal for rendering and analyzing large datasets. The 4D viewer in ZEISS arivis Pro can be configured to adjust the appearance of individual channels independently to highlight specific features. These settings, along with clipping planes or the varying opacity of individual channels, can be stored into key frames which the software automatically interpolates between to produce a seamless animation.

These animations can be previewed and edited prior to producing high resolution video renders. Sample courtesy of M. Paoli, Galizia Lab, University of Konstanz, Germany

# Your Flexible Choice of Components

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## 1 Microscope

- Inverted stand: Axio Observer
- Upright stand: Axio Examiner, Axio Imager
- Port for coupling of Elyra 7 (Axio Observer)
- Camera port
- AI Sample Finder for Axio Observer
- Manual or motorized stages
- Incubation solutions
- Fast Z piezo inserts
- Definite Focus

## 2 Objectives

- C-Apochromat, C Plan-Apochromat
- Plan-Apochromat
- W Plan-Apochromat, Clr Plan-Apochromat,
- Clr Plan-Neofluar
- LD LCI Plan-Apochromat

## 3 Illumination

- V laser: 405 nm
- VIS + NIR laser: 445 nm, 488 nm, 514 nm, 543 nm, 561 nm, 594 nm, 639 nm, 730 nm
- Laser for multiphoton imaging: Ti:Sa (single-line laser), InSight X3 / X3+ and Discovery NX (dual-line laser)

## 4 Detection

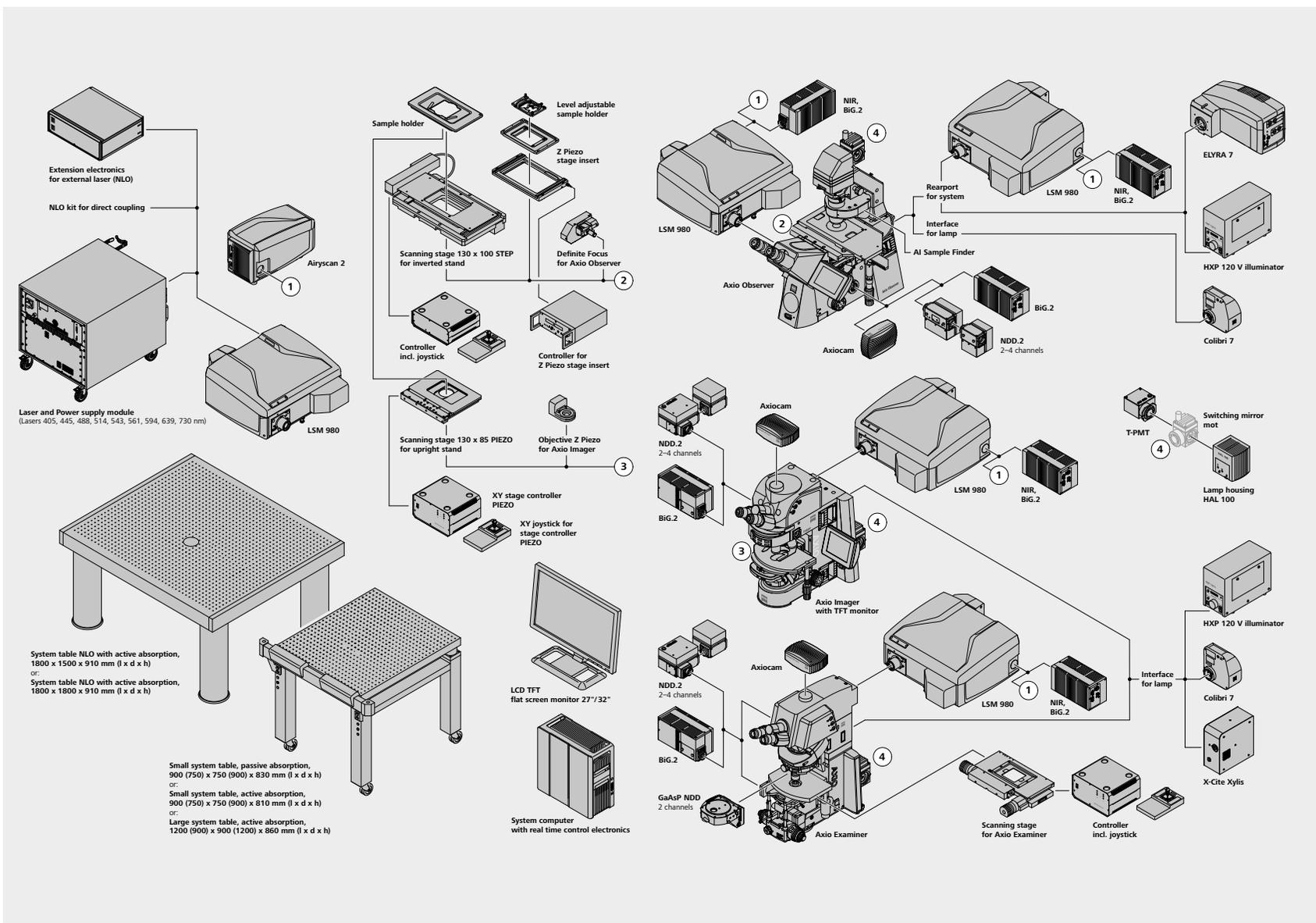
- 3, 6, or 34 descanned spectral channels (GaAsP and MA-PMT)
- NIR Detector (2 channels) with near infrared optimized GaAsP and GaAs detector
- 2 additional GaAsP channels (BIG.2)
- Up to 6 non-descanned GaAsP detectors
- Up to 12 non-descanned GaAsP and multi-alkali PMT detectors
- Airyscan 2 detector
- Transmitted light detector (T-PMT)

## 5 Software

- ZEN microscopy software, highlighted modules: LSM Plus, Airyscan Joint Deconvolution, Dynamics Profiler, Tiles & Positions, Experiment Designer, FRAP, FRET, FCS, RICS, Connect Toolkit, Direct Processing, 3D Toolkit

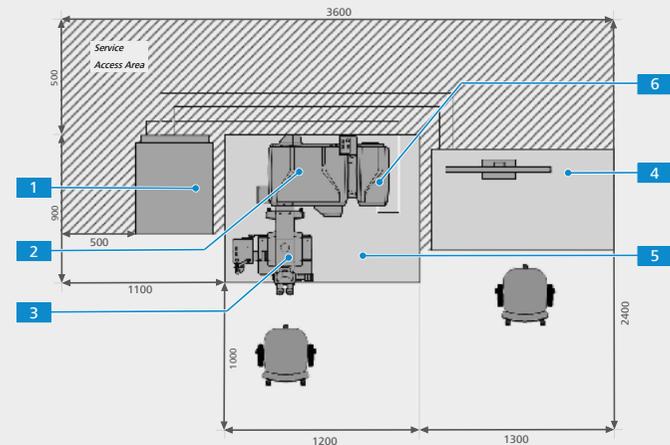
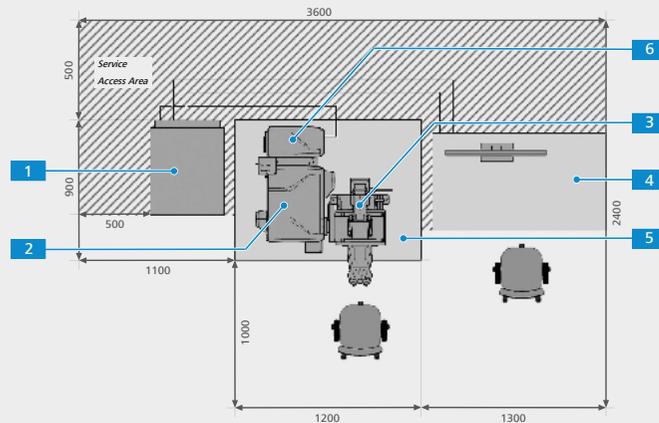
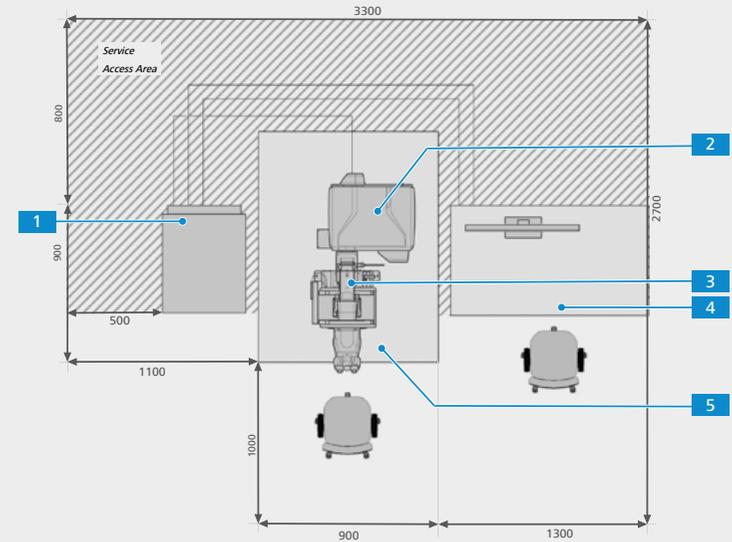
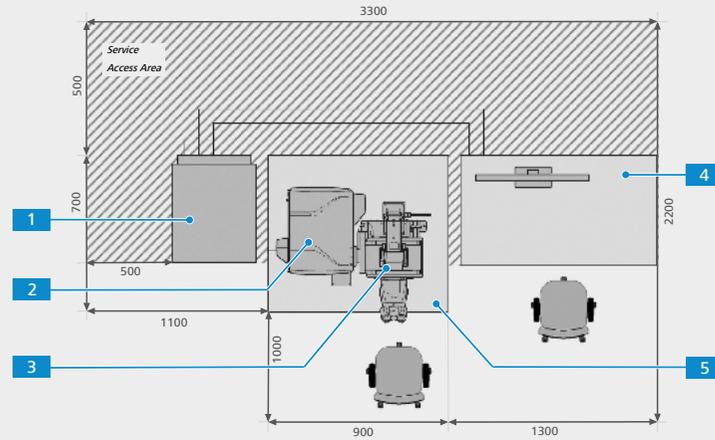
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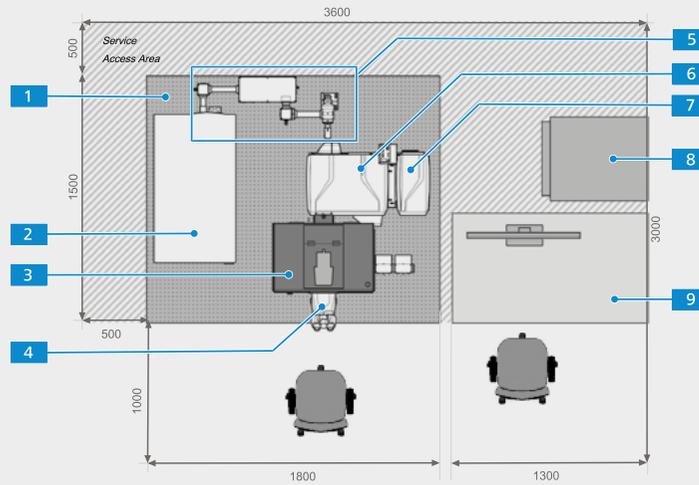
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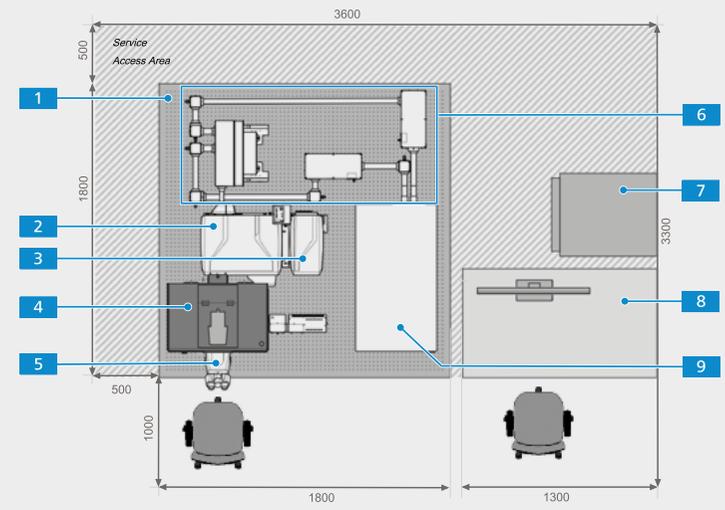
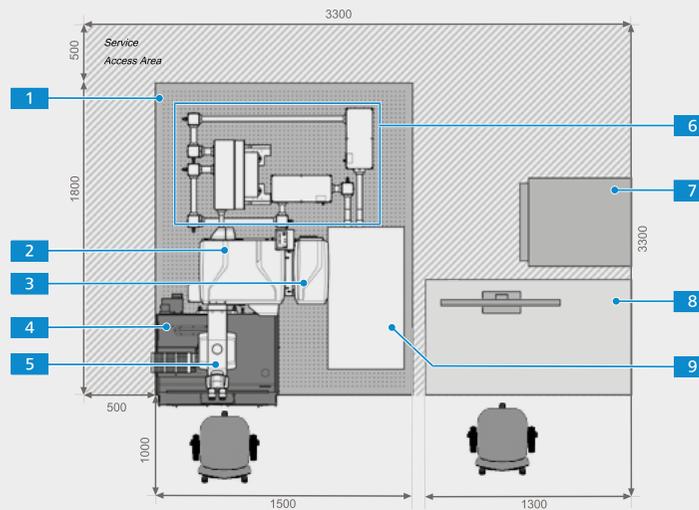
1 Laser and Power Supply Module 2 LSM 980 Scanhead 3 Microscope Stand (Axio Observer, Axio Imager or Axio Examiner) 4 Computer Table 5 System Table 6 Airscan 2

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- 3 Incubation
- 4 Microscope Stand (Axio Observer, Axio Imager or Axio Examiner)
- 5 Laser coupling with AOM for multiphoton laser
- 6 LSM 980 scanning module
- 7 Airyscan 2
- 8 Laser and Power Supply Module
- 9 Computer Table



- 1 System Table
- 2 LSM 980 scanning module
- 3 Airyscan 2
- 4 Incubation
- 5 Microscope Stand
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- 7 Laser and Power supply module
- 8 Computer Table
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| Physical Dimensions                 | Length (cm) | Width (cm) | Height (cm) | Weight (kg) |
|-------------------------------------|-------------|------------|-------------|-------------|
| Small Passively Damped System Table | 90          | 75         | 83          | 130         |
| Small Actively Damped System Table  | 90          | 75         | 81          | 130         |
| Large Actively Damped System Table  | 120         | 90         | 86          | 180         |
| Active Anti-Vibration Table (NLO)   | 180         | 150        | 91          | 475         |
| Active Anti-Vibration Table (NLO)   | 180         | 180        | 91          | 515         |
| Scanning Module LSM 980             | 55          | 45         | 22          | 27          |
| Microscope                          | 47–80       | 29–39      | 70–72       | 37–47       |
| Laser and Power Supply module       | 60          | 50         | 56          | 70          |
| Airyscan 2                          | 40          | 20         | 24          | 12          |
| Fiber Optic Cable, UV               | 400         |            |             |             |
| Fiber Optic Cable, VIS              | 400         |            |             |             |
| Cables                              | 250         |            |             |             |

| Microscopes         |   |
|---------------------|---|
| Stands              | Upright: Axio Imager.Z2, Axio Examiner.Z1<br>Inverted: Axio Observer 7 with side port or rear port, AI Sample Finder (optional)   |
| Z Drive             | Smallest increment Axio Imager.Z2: 10 nm;<br>Axio Observer 7: 10 nm;<br>Axio Examiner: 25 nm;<br>fast piezo objective or stage focus available; Definite Focus for Axio Observer 7                        |
| XY Stage (optional) | Motorized XY scanning stage, for Mark & Find function (XYZ) as well as Tile Scan (Mosaic Scan);<br>smallest increment of 0.25 µm (Axio Observer 7), 0.2 µm (Axio Imager.Z2) or 0.25 µm (Axio Examiner.Z1) |

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| <b>Scanning Module</b>   |  |
|--------------------------|--|
| Scanner                  | Two independent, galvanometric scanning mirrors with ultrashort line and frame flyback   |
| Scanning Resolution      | 32 × 1 to 8,192 × 8,192 pixels, also for multiple channels, continuously adjustable  |
| Scanning Speed           | At 512 × 512 pixels: confocal – up to 13 fps; 34-Channel Lambda Scan up to 5 fps; Airyscan SR – up to 4.7 fps; Multiplex SR-4Y – 25 fps; Multiplex SR-8Y – 47.5 fps; Multiplex CO-8Y – 34.4 fps<br>19 × 2 speed levels for confocal; 512 × 16 pixels up to 425 fps; up to 6830 lines/sec.<br>13 × 2 speed levels in Multiplex mode; up to 25 fps for 904 × 904 pixels; up to 17.8 fps at 1,024 × 1,024 pixels  |
| Scanning Zoom            | 0.6× to 40×; digitally adjustable in increments of 0.1 (Axio Examiner: 0.7 × to 40 ×)  |
| Scanning Rotation        | Can be rotated freely (360 degrees), adjustable in increments of 0.1 degree, freely adjustable XY offset   |
| Scanning Field           | 20 mm field diagonal (max. 17 mm for Axio Examiner) in the intermediate image plane, with full pupil illumination  |
| Pinholes                 | Master pinhole with preset size and position; can be adjusted as desired for multitracking and short wavelengths (such as 405 nm)  |
| Beam Path                | Exchangeable Twin Gate beamsplitter with up to 100 combinations of excitation wavelengths and outstanding laser line suppression; manual interface port for two external detection modules (such as NIR, BiG.2, Airyscan 2, third party detectors), internal detection with spectral signal separation and signal recycling loop for compensation of polarization effects  |
| <b>Detection Options</b> |  |
| Detectors                | 1, 4 or 32 GaAsP PMT combined with 2 multialkali PMT internal spectral detection channels (QE 45 % typical for GaAsP); LSM Plus: resolution down to 160* nm lateral, 500 nm axial with pinhole at 0.8 AU; resolution down to 120* nm lateral, 500 nm axial with pinhole at 0.3 AU<br><br>Additional Detection: 2ch NIR (GaAs and NIR GaAsP) detection or 2ch BiG.2 (UV-Vis GaAsP) detection<br><br>Airyscan 2 detector (32 channels GaAsP), delivers resolution down to 120* nm lateral, 350 nm axial; with jDCV: 90* nm lateral, 270 nm axial; Multiplex resolution: 140/160 nm lateral, 450 nm axial<br><br>Up to 12 non-descanned detection channels (PMT and/or GaAsP) depending on microscope stand<br><br>Transmitted light detector (PMT) |
| Spectral Detection       | 3, 6, or 34 + 2 NIR simultaneous, confocal reflected-light channels, GaAs, GaAsP (UV-Vis and NIR) and multialkali PMT based; freely adjustable spectral detection area (resolution down to 3 nm)   |
| Data Depth               | 8 bit or 16 bit available; up to 36 channels simultaneously detectable   |
| Real-time Electronics    | Microscope, laser, scanning module and additional accessory control; data acquisition and synchronization management through real-time electronics; oversampling read-out logic; ability to evaluate data online during image acquisition  |

\* Measured with respective nanoruler samples

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| <b>ZEN Microscopy Software</b>            |   |
|---|---|
| System Configurations                     | Workspace to conveniently configure all of the motorized functions of the scanning module, laser and microscope;<br>Save and restore application configurations as experiment settings or use acquired images (Reuse)   |
| Maintenance and calibration Tools         | Software tools and wizards to automatically test and calibrate the system   |
| Recording Modes, Smart Setup              | Spot, Line/Spline, Frame, Tiles, Z Stack, Lambda Stack, Time Series and all combinations (XYZ, lambda, t), online calculation and visualization of, average and summation (by line/image, adjustable), Step Scan (for higher image frame rates);<br>Quick set up of imaging conditions using Smart Setup by simply selecting the labelling dye                      |
| Crop Function                             | Easily select the scanning area by defining simultaneously zoom, offset and rotation  |
| Real ROI Scan, Line and Spline curve Scan | Scan multiple ROIs (regions of interest) as desired and pixel-by-pixel laser blanking;<br>Scan along a freely defined line  |
| ROI Bleaching                             | Localized bleaching in multiple bleach ROIs for applications such as FRAP (fluorescence recovery after photobleaching) or uncaging;<br>Use a speed or z-position different from imaging settings, use of different laser lines for different ROIs   |
| Multitracking                             | Rapidly change excitation lines when recording multiple fluorescences for the purpose of minimizing signal crosstalk and increasing dynamic range   |
| Multiplex Mode                            | Multiplex mode scan with 4x or 8x parallelisation in Y-direction, detection by Airyscan 2   |
| Lambda Scan                               | Parallel or sequential acquisition of multidimensional images with spectral information for every pixel   |
| Linear Unmixing                           | Acquisition of crosstalk-free, multiple fluorescence images using simultaneous excitation;<br>Online or offline and automatic or interactive unmixing;<br>Advanced unmixing logic with indication of reliability  |
| Visualization                             | 2D (XY); Split (XY-ch); Gallery (XY-ch, XY-Z), Orthogonal (XY, XZ, YZ) with adjustable cut lines, maximum intensity projection and 3D distance measurement; 2.5D viewing with various rendering options and animations; Histogram settings using channel specific brightness, gamma and contrast; color table selection and modification (LUT), various annotations |
| Image Analysis and Operations             | Colocalization and histogram analysis with individual parameters, number & brightness analysis, profile measurement along user-defined lines, measurement of lengths, angles, areas, intensities and much more; operations: addition, subtraction, multiplication, division, ratio, shift, filters (low-pass, median, high-pass, etc., also user-definable)         |
| Image Management                          | Features for managing images and the corresponding imaging parameters   |
| Advanced Acquisition Toolkit              | Z-stack and enhanced depth of focus functionality<br><br>Tiles & Positions: Scanning of predefined sample areas (tiles) and / or position lists<br><br>Software Autofocus: Determination of the optimal focus position in the sample  |
| 3D Toolkit                                | Combined 2D and 3D visualization in one screen<br><br>Rapid 3D and 4D reconstructions and animations<br><br>3D segmentation to quantify 3D microscopy data based on thresholding and machine learning models  |

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| Optional Software   |  |
|---|--|
| Direct Processing   | Processing of large data during acquisition by streaming, including e.g., Airyscan, LSM Plus; analysis and storage on second PC  |
| Deconvolution Toolkit                                       | 3D, GPU based image restoration based on calculated point-spread functions (modes: nearest neighbor, maximum likelihood, constrained iterative)  |
| HDR   | Imaging mode: High Dynamic Range, improvement of the dynamic signal range by combination of multiple images with ramped signal   |
| Molecular Quantification Toolkit                            | Physiology (Dynamics): Comprehensive evaluation software for online and offline ratio imaging with various pre-defined formulas<br>Acquisition of FRET (Förster resonance energy transfer) image data with subsequent evaluation<br>Acceptor Photobleaching and Sensitized Emission methods supported<br>Acquisition of FRAP (fluorescence recovery after photobleaching) experiments with subsequent evaluation of intensity kinetics |
| RICS Image Correlation                                      | Single molecule imaging and analysis using multialkali or GaAsP PMT detectors (publ. v. Gratton)   |
| Smart Acquisition Toolkit                                   | Experiment Designer: Definition of customized imaging configurations and procedures<br>Guided Acquisition: Automated and targeted acquisition of objects of interest   |
| Developer Toolkit   | Python scripting interface for automation & customization; experimental feedback for smart experiments and open interface to third party software (e.g. ImageJ)  |
| Connect Toolkit   | Exchange and alignment of image data from multiple image acquisition systems in 2D and 3D enabling correlative workflows   |
| AI Toolkit  | Image analysis and structure detection via computational self learning technology  |
| FCS/FCCS  | Fluorescence Correlation and Cross Correlation Spectroscopy for analysis of single molecule dynamics, concentration and number   |
| AI Sample Finder, Sample Navigator (requires additional HW) | Easy to perform sample overview scan with autofocus function using AxioCam or transmitted fluorescence with T-PMT (Finder requires Axio Observer)  |
| Bio Apps Toolkit  | Easy-to-use and modular image analysis for common assays   |
| Airyscan RAW data   | Optional export of complete Airyscan single channel data and the sheppard sum for external processing, e.g. correlations, deconvolution, AI etc.   |
| Airyscan Joint Deconvolution                                | Postprocessing joint deconvolution for Airyscan SR data, increased resolution down to 90 nm lateral  |
| LSM Plus  | Increased resolution for confocal/spectral datasets down to 160 nm lateral (120 nm with closed pinhole = 0.3 AU), preview and Auto strength  |
| Dynamics Profiler   | Easy-to-use Airyscan-based data collection that captures the underlying dynamics of living samples to provide molecular concentration, asymmetric diffusion, and flow information (Axio Observer)  |

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## Lasers

|  |  |
|--|--|
| Lasers   |  |
| Laser RGB (445, 488, 514, 543, 561, 594, 639 nm)   | <p>Single-mode polarization preserving fiber</p> <p>Laser beam attenuation for all lasers by VIS-AOTF</p> <p>Diode Laser 445 nm (30 mW nominal power; 7.5 mW ex fiber)</p> <p>Diode Laser 488 nm (30 mW nominal power; 10 mW ex fiber)</p> <p>Diode Laser 514 nm (30 mW nominal power; 10 mW ex fiber)</p> <p>DPSS Laser 543 nm (25 mW nominal power; 10 mW ex fiber)</p> <p>DPSS Laser 561 nm (25 mW nominal power; 10 mW ex fiber)</p> <p>DPSS Laser 594 nm (8 mW nominal power; 2.5 mW ex fiber)</p> <p>Diode Laser 639 nm (25 mW nominal power; 7.5 mW ex fiber)</p> |
| Laser V and NIR (405 and 730 nm), direct modulated | <p>Single-mode polarization preserving fiber</p> <p>Diode Laser 405 nm (30 mW nominal power; 14 mW ex fiber)</p> <p>Diode Laser 730 nm (20 mW nominal power; 9.5 mW ex fiber)</p>  |

## Power Requirements

LSM 980 has a main power supply cord and plug, either NEMA L5-15 (100V – 125V) 2pol (15A) + PE or CEE blue (200 – 230V) 2pol (16A) + PE.

|                |                         |                         |
|----------------|-------------------------|-------------------------|
| Line Voltage   | 1/N/PE 230 V AC (±10 %) | 1/N/PE 120 V AC (±10 %) |
| Line Frequency | 50...60 Hz              | 50...60 Hz              |

## ZEISS LSM 980 incl. VIS Laser

|   |              |               |
|---|--------------|---------------|
| Max. Current                            | 7 A at 230 V | 13 A at 120 V |
| Heat emission without Multiphoton Laser | 1500 W max.  | 1500 W max.   |
| Power Consumption                       | 1600 VA max. | 1600 VA max.  |

## Multiphoton Laser

Power consumption and heat emission varies depending on type of laser. See data sheet of laser from laser supplier for further information.

## EMC test

according to DIN EN 61326-1  
 – Emitted interference according to CISPR 11 / DIN EN 55011  
 – Interference immunity as specified in Table 2 (industrial)

Nominal power = power level of laser itself, leaving out necessary tolerances and losses due to laser steering and stability requirements

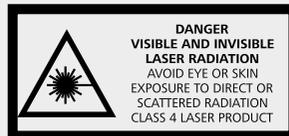
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## Environmental Requirements

For operation the system has to be placed in a closed room.

|  |  |
|--|--|
| 1. Operation, specified performance  | T = 22 °C ± 3 °C without interruption (24 h a day independently whether system is operated or switched-off)<br>It has to be ensured that the air-flow of the air-conditioning is not directed at the system. |
| 2. Operation, reduced performance  | T = 15 °C to 35 °C, any conditions different from item 1. and 5.   |
| 3. Storage, less than 16 h   | T = -20 °C to 55 °C  |
| 4. Temperature gradient  | ±0.5 °C/h  |
| 5. Warm up time  | 1 h, for high-precision and/or long-term measurements ≥3 h   |
| 6. Temperature gradient and range for continuous long-term image acquisition | ± 1.5 °C/12 h  |
| 7. Relative humidity   | <65 %  |
| 8. Operation altitude  | max. 2000 m  |
| 9. Loss of heat (without Multiphoton Laser)                                  | 1.5 kW   |
| 10. Vibrations under operation conditions (with system table)                | Vibration Class 12.5 µm/s VC-C<br>(IEST RP 12 and ISO 10811)   |
| 11. Shipping shock (LSM 980 box)   | <10 g  |



LSM 980 meets the requirements according to IEC 60825-1:2014

# ZEISS Service – Your Partner at All Times

Your microscope system from ZEISS is one of your most important tools. For over 175 years, the ZEISS brand and our experience have stood for reliable equipment with a long life in the field of microscopy. You can count on superior service and support - before and after installation. Our skilled ZEISS service team makes sure that your microscope is always ready for use.

- › In Brief
- › The Advantages
- › The Applications
- › The System
- › Technology and Details
- › **Service**

## Procurement

- Lab Planning & Construction Site Management
- Site Inspection & Environmental Analysis
- GMP-Qualification IQ/OQ
- Installation & Handover
- IT Integration Support
- Startup Training

## Operation

- Predictive Service Remote Monitoring
- Inspection & Preventive Maintenance
  - Software Maintenance Agreements
    - Operation & Application Training
    - Expert Phone & Remote Support
  - Protect Service Agreements
    - Metrological Calibration
      - Instrument Relocation
        - Consumables
        - Repairs

## New Investment

- Decommissioning
- Trade In

## Retrofit

- Customized Engineering
  - Upgrades & Modernization
- Customized Workflows via arivis Cloud



Please note: Availability of services depends on product line and location

>> [www.zeiss.com/microservice](http://www.zeiss.com/microservice)



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