



Product Information  
Version 1.1

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

Your Next Generation Confocal for Fast and Gentle Multiplex Imaging



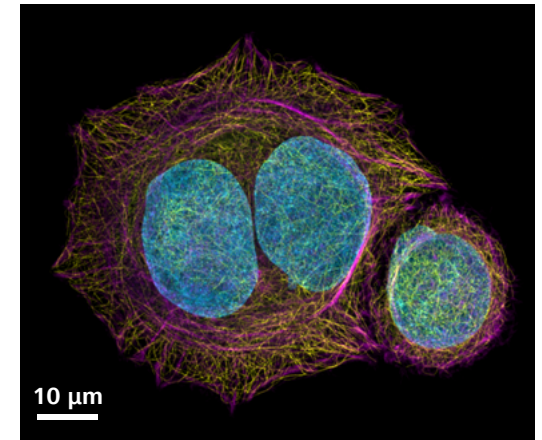
# Your Next Generation Confocal for Fast and Gentle Multiplex Imaging

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Life sciences research can be demanding, and if you are involved in neuroscience, cancer research or other cell- or organism-based disciplines, you'll often need microscopy data for your work. Emerging technologies such as CRISPR/Cas open up innovative ways of thinking and allow you to ask altogether new scientific questions, deeply affecting your imaging experiments. To monitor life as undisturbed as possible requires low labeling density for your biological models—for example, 3D cell culture, spheroids, organoids or even whole organisms—and this calls for 3D imaging that combines optical sectioning with low phototoxicity and high speed. Then there are the repeated experiment runs it takes to get statistically-valid data for your conclusions: it soon becomes apparent you will also need high throughput.

Your new LSM 980 with Airyscan 2 is the ideal platform for confocal 4D imaging. The entire beam path is optimized for simultaneous spectral detection of multiple weak labels with the highest light efficiency. Add Airyscan 2 with its new Multiplex mode to get more imaging options to enhance your experiments. You can now choose the perfect setup to gently image larger fields of view with superresolution in shorter acquisition times than ever before.

A number of software helpers will optimize your workflow and support efficient acquisition and data management. With ZEN Connect you can document and share all details of your experiments. You'll always keep the context as you combine overview images, ROIs and additional data, even across imaging modalities.



*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. Courtesy of A. Politi, J. Jakobi and P. Lenart, MPI for Biophysical Chemistry, Göttingen, Germany.*

See for yourself how the new Multiplex mode for Airyscan 2 gives you better data faster than ever before. Book a hands-on demonstration in one of our ZEISS Microscopy Labs now.

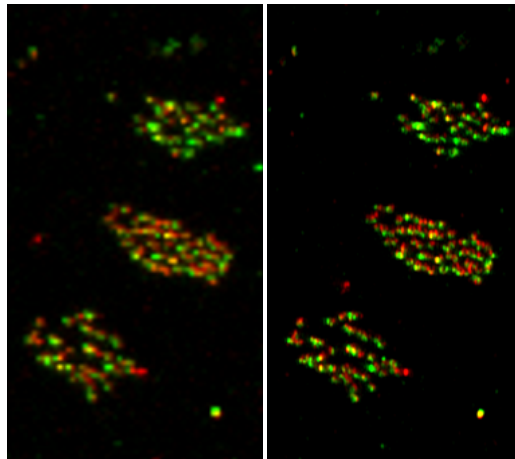
>> [www.zeiss.com/lsm980](http://www.zeiss.com/lsm980)

# Simpler. More Intelligent. More Integrated.

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## Get Better Data Faster

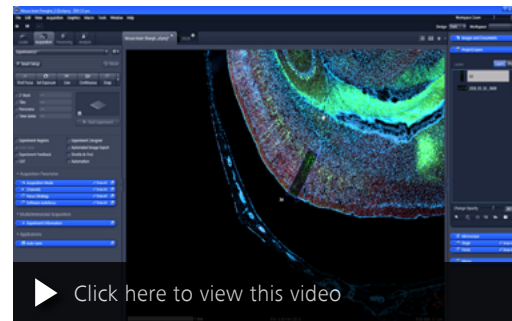
Use the new Multiplex mode for Airyscan 2 and get more information in less time. Smart illumination and detection schemes let you image your most challenging three-dimensional samples with high framerates beyond the diffraction limit and still treat your sensitive samples gently. By combining the full flexibility of a point scanning confocal with the speed and gentleness of the sensitive Airyscan area detector, it's now possible to answer your scientific questions up to ten times faster with superresolution.



Basal bodies (red) and basal feet (green) of ependymal cilia labeled by immunohistochemistry. Left side: imaged in conventional confocal mode; right side: imaging with the same frame time using the new Multiplex mode for Airyscan 2 clearly reveals the orientation of the cilia. Courtesy of S. Kapoor, Max Planck Institute for Biophysical Chemistry, Göttingen, Germany.

## Increase Your Productivity

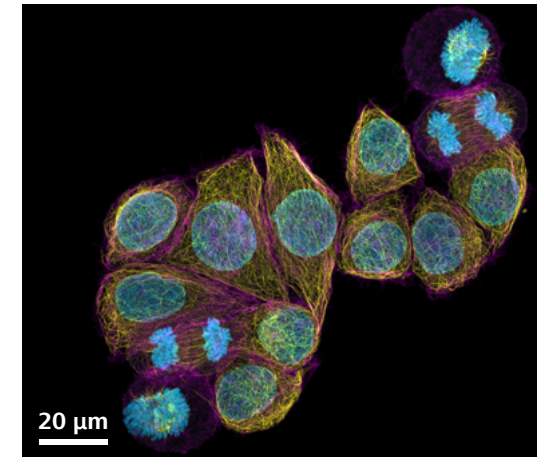
It's never been easier to set up complex confocal live cell imaging experiments. The latest version of ZEN imaging software drives the new LSM 980 with Airyscan 2 and puts a wealth of software helpers at your command. You'll work easier than ever before and faster, too, achieving reproducible results in the shortest possible time. Smart Setup and the new Sample Navigator let you find and image regions of interest quickly, leaving more time for the real work of acquiring data. 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.



See how ZEN Connect helps to always keep your context while imaging. From acquiring an overview image, to defining ROI's, and even when changing between different imaging systems. You save time and always stay on top of things.

## Image with More Sensitivity

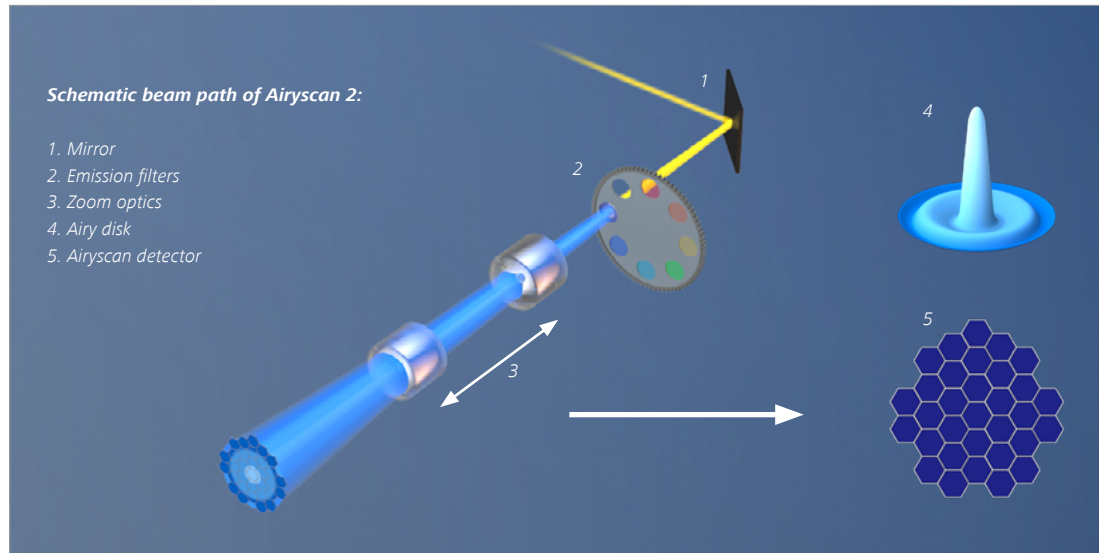
LSM 980 brings you the best of two worlds to image your most challenging samples. You get the light efficient beam path of the LSM 9 family with up to 34 simultaneous channels for full spectral flexibility. This lets you image faint signals with 4 – 8 times higher signal-to-noise ratio (SNR). Plus, when you combine it with Airyscan 2, this revolutionary area detector extracts even more information from your sample in less time. You don't need to close a pinhole to get superresolution, which makes your 3D imaging even more light efficient. Expect the very best data quality from all your samples.



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 superresolution imaging of a large field of view. Courtesy of A. Politi, J. Jakobi and P. Lenart, MPI for Biophysical Chemistry, Göttingen, Germany.

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## The Airyscan Principle

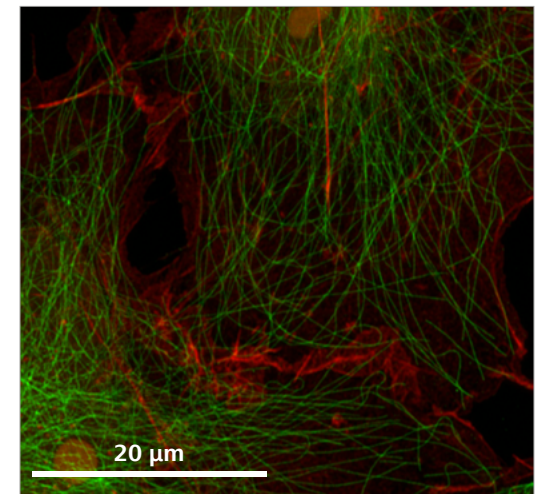
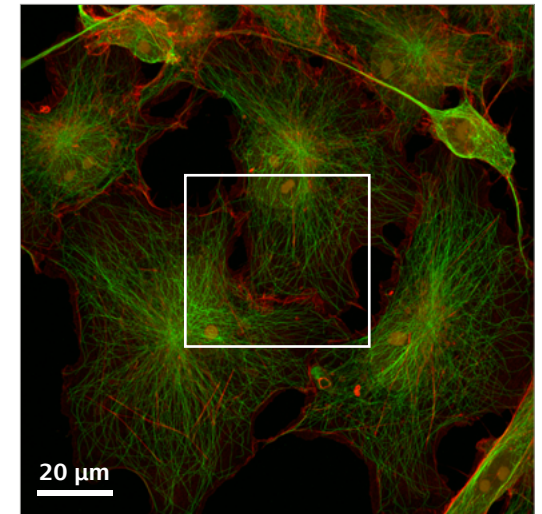
Classic confocal laser scanning microscopes use point illumination to scan the sample sequentially.

The microscope optics transform each point to an extended Airy disk (Airy pattern). A pinhole then spatially limits this Airy disk to block out-of-focus light from reaching the detector. Closing the pinhole gives higher resolution, but at the price of detecting fewer photons – and these photons cannot be brought back by e.g. deconvolution.

Airyscan 2 is an area detector with 32 concentrically arranged detection elements. This allows you to acquire more of the Airy disk at once. The confocal pinhole itself remains open and does not block light, thus more photons are collected. This produces much greater light efficiency while imaging. Airyscan 2 gives you a unique combination of gentle superresolution imaging and high sensitivity.

**For further information on the Airyscan principle please refer to:**

<https://zeiss.ly/airyscan-principle>



*Comparing the field of view you can image at superresolution in the same time using Airyscan SR (bottom) and Multiplex mode (top). COS 7 cells with labelled microtubules (alpha-tubulin 488, green) and actin (phalloidin 647, red).*

# Your Insight into the Technology Behind It

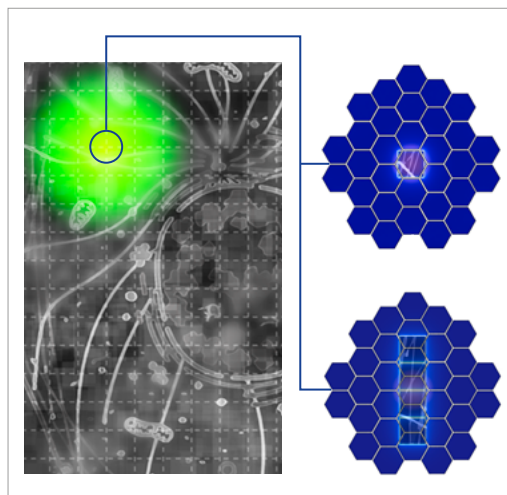
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## The New Multiplex Mode for Airyscan 2

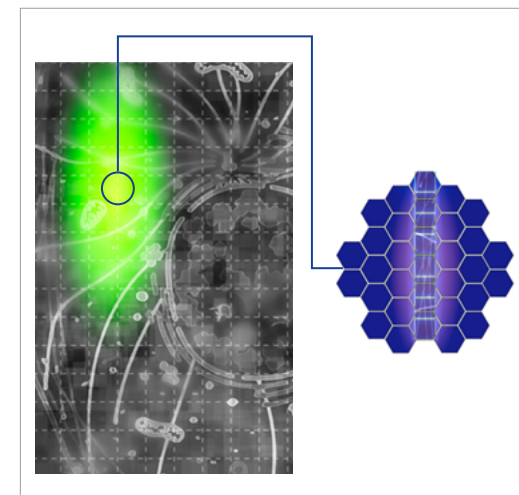
Do you want to image large fields of view and whole sample volumes in shortest possible time? And do you want to image with superb image quality at the same time? The LSM 9 family with Airyscan 2 from ZEISS now gives you more options to fit imaging speeds and resolution to your experimental needs. You combine an area detector with smart illumination and readout schemes, which let you choose from different parallelization options.

The new Multiplex mode uses knowledge about 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 to take bigger steps when sweeping the excitation laser over the field of view, improving your achievable acquisition speeds. In fact, the high amount of spatial information captured in the pinhole plane allows to reconstruct a final image with better resolution than the acquisition sampling. Airyscan 2 in Multiplex mode can acquire up to four superresolution image lines with high SNR in a single sweep. Your LSM 980 with Airyscan 2 allows to stretch the excitation laser spot to image eight lines in parallel. Use this speed advantage for ultrafast time series of single slices, for rapid tiling of large areas or for fast volumetric time-lapse imaging.

## Choose the perfect Airyscan Mode for Your Experiment



For each illumination position, Airyscan SR mode generates one superresolution image pixel. The spatial information provided by Airyscan 2 in Multiplex SR-4Y allows to scan 4 superresolution image lines in a single sweep.



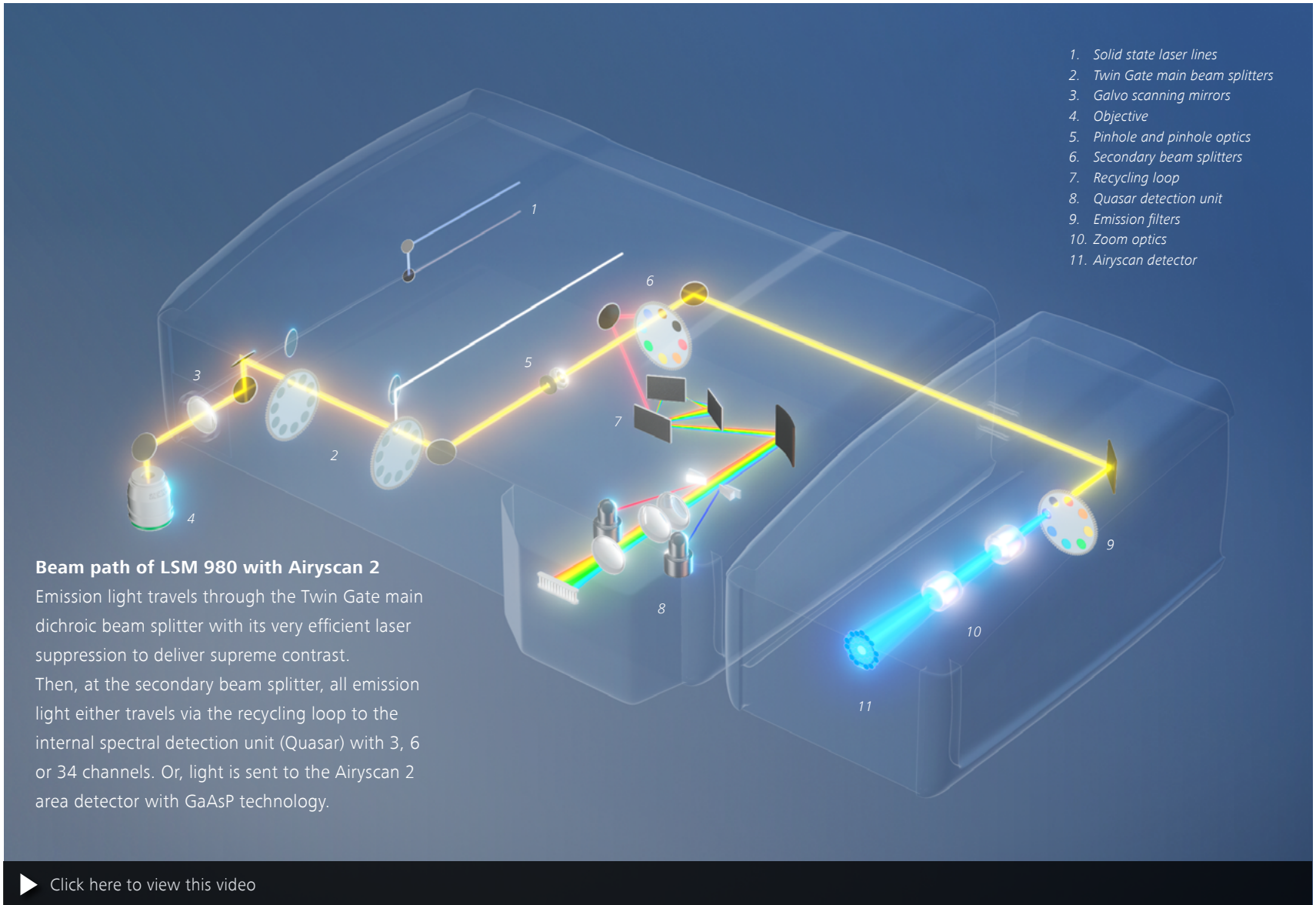
For Airyscan Multiplex SR-8Y and CO-8Y the illumination laser spot is vertically elongated which allows to capture 8 image pixels for each illumination position. Sampling can be done in superresolution (SR) or confocal (CO) resolution, depending on your experiment.

### LSM 980 with Airyscan 2

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

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## A Flexible and Sensitive System for Best Image Quality

Laser scanning microscopes (LSMs) bring a great deal of freedom to your experimental setup. The beam path design and every single component of your LSM 980 are optimized to deliver the highest sensitivity and flexibility for your experiments.

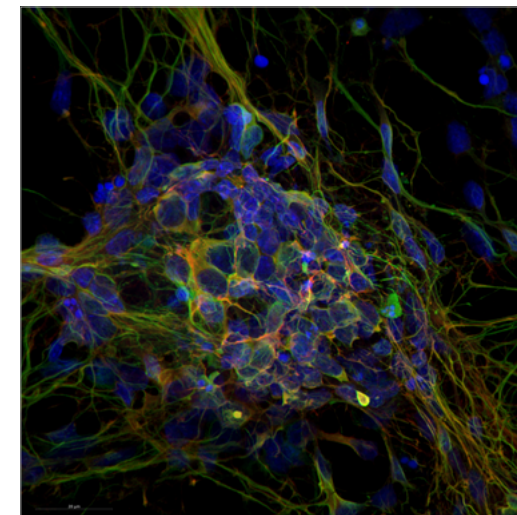
Take your choice of solid-state lasers to generate your excitation light. The linear scanners illuminate your sample evenly for efficient signal collection during more than 80 % of the frame time. The emission light travels through the Twin Gate main dichroic beam splitter. The special low angle orientation suppresses stray light and gives you crisp contrast in all situations. You can even extend your emission detection range over the excitation laser line to make sure you collect all of those precious emission photons.

Nowadays you can employ a wealth of fluorescent labels to analyze multiple sample structures simultaneously. Your LSM 980 directs all emission light

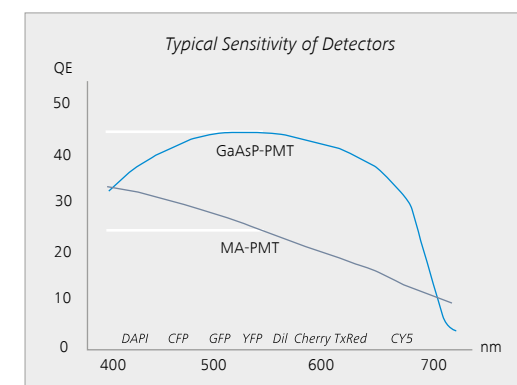
to the 3, 6 or 34 channel Quasar detector where all signals from your dye combination are measured. The unique recycling loop maximizes your detection efficiency. Depending on which labels you choose, you will define the emission detection bands with nanometer precision. You can acquire overlapping labels or autofluorescence in a single lambda scan with 34 channels and then separate them with Linear Unmixing. This allows you to keep your sample's light exposure to a minimum while also speeding up your imaging.

With LSM 980, you always have the advantage of the enhanced quantum efficiency of sensitive GaAsP detectors. You can switch to photon counting readout to analyze the weakest labels efficiently.

Then add Airyscan 2 for enhanced sensitivity, speed and superresolution – and combine all imaging modes into one single experiment.



Maximum intensity projection of neurosphere, multi-color label with DAPI (blue), Tubulin-Cy2 (green), DCX-Cy5 (red). Acquired with ZEISS Airyscan 2 in Multiplex mode. Sample courtesy of H. Braun, LSM Bioanalytik GmbH, Magdeburg, Germany.



Typical spectral quantum efficiency (QE) of multi-alkali (MA-PMT) and GaAsP-PMT detectors.

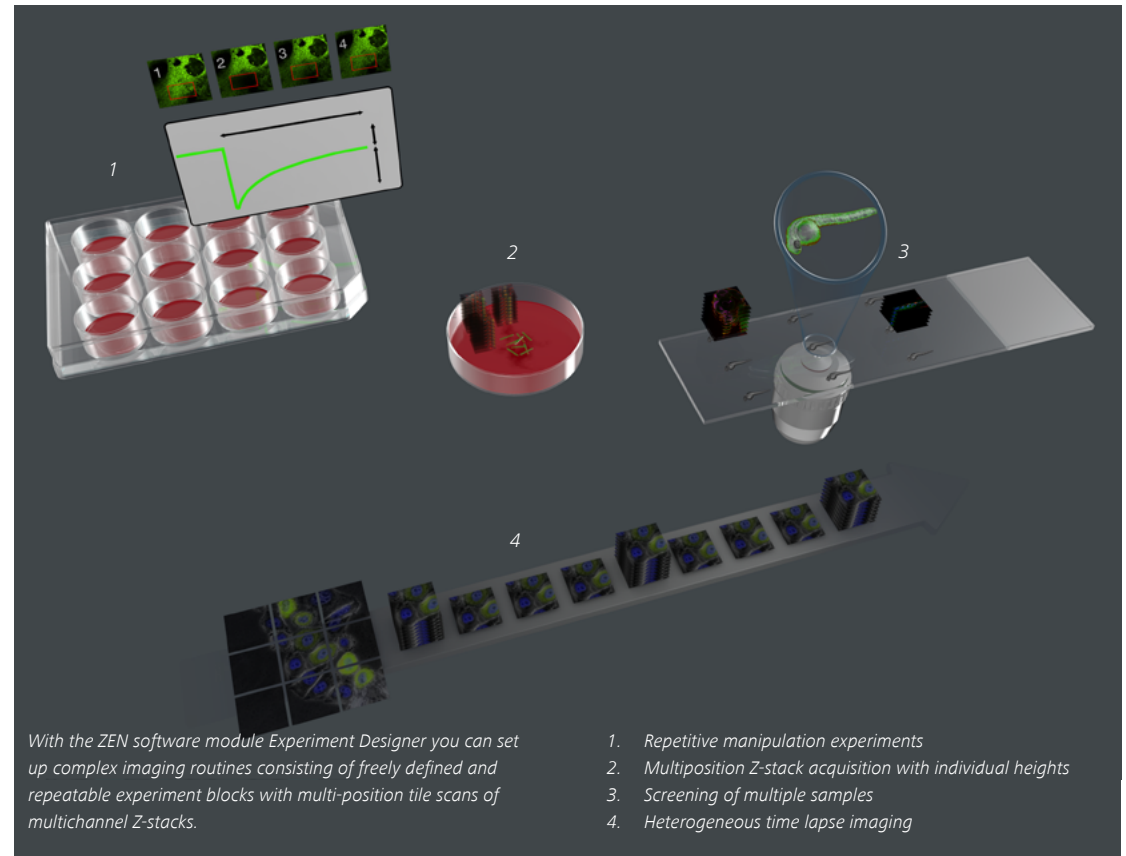
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## Acquire Reproducible Data with Ease

With all its various aspects and workflows, your research leaves you with no time to waste. That's why ZEN imaging software was created—to make your confocal imaging both efficient and enjoyable. ZEN – ZEISS Efficient Navigation – is the only user interface you will ever see on all imaging systems from ZEISS. This familiar and easy-to-learn interface will help you get reproducible results in the shortest possible time.

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. The new Sample Navigator makes quick work of finding and imaging the regions of interest (ROI) on your specimen. The fast Autofocus lets you quickly acquire an overview image of your whole sample using the AxioCam or T-PMT. It takes less time to illuminate your sample and leaves you more of the precious time you've booked on the system for imaging. In addition, you can use the overview image to document all steps of your experiment and load it in ZEN Connect to 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 .



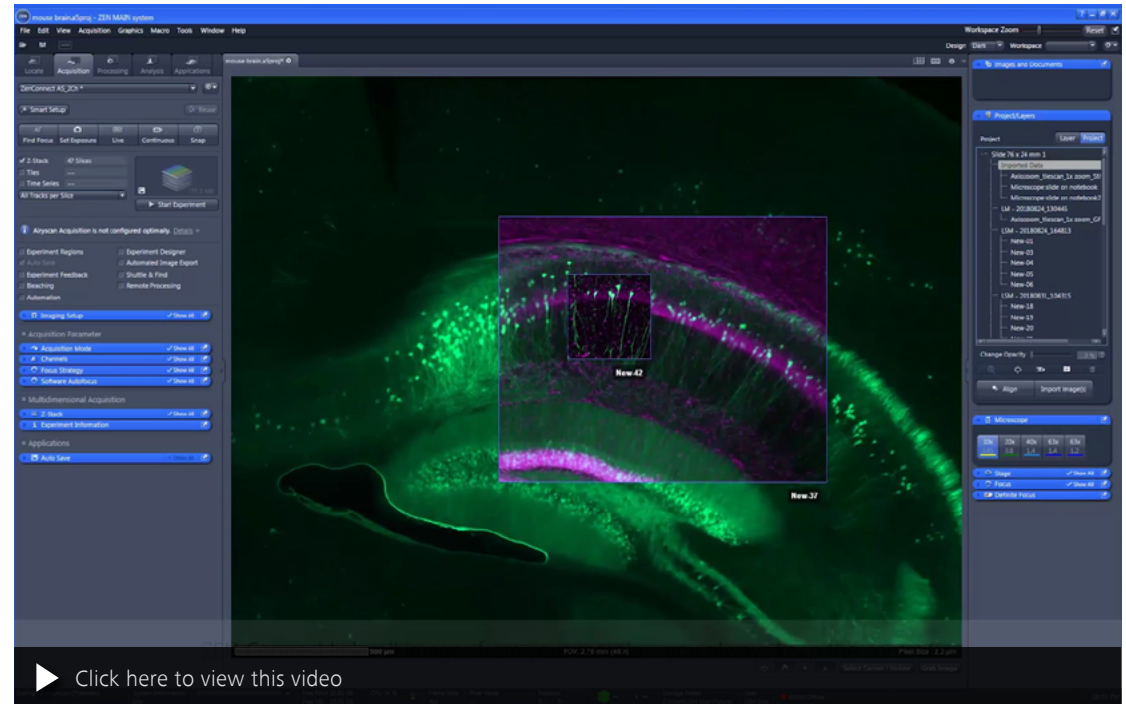
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## See More Details

Sometimes you need to see and assess your multi-modal images during acquisition in order to plan your next steps. ZEN imaging software gives you multiple options. You can sit at your connected computer to start the new 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. ZEN Connect 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 co-work with others as a team.



*Connect all your imagery: With ZEN Connect 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 arivis®, is optimized to render the large 3D and 4D image data you have acquired with the fast new LSM 980. You can create impressive renderings

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

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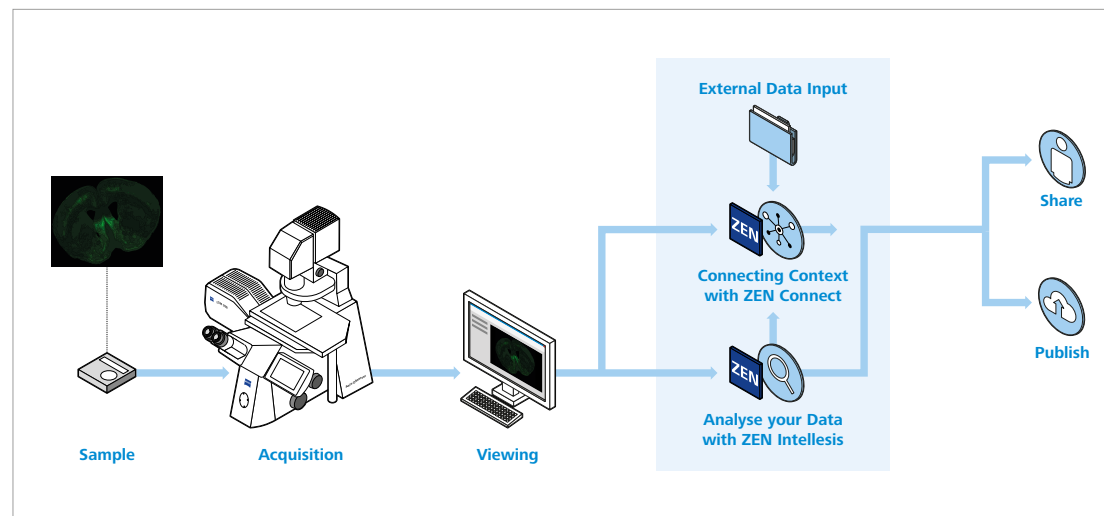
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## Get More Data from Your Sample

As enjoyable as microscopic images are, their real value is in the data they provide. The CZI file format of ZEN imaging 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.

You can perform FRET analysis based on sensitized emission or acceptor photobleaching. Or analyze dynamic processes with ratiometric imaging or photomanipulation experiments such as FRAP or FLAP. Raster image correlation spectroscopy (RICS) gives you access to information on single molecule dynamics and correlation data, based on conventional LSM images.

ZEN Intellesis lets you segment complex multi-modal images. Just use your own expertise to train the software on a few images. Then powerful deep learning algorithms will take over and do all the time-consuming segmentation steps on the hundreds of similar images. Integrate the individual segmentation models seamlessly into your ZEN image analysis workflow.



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



*From beautiful images to valuable data  
Use the power of deep learning to easily segment your images.  
A smooth workflow helps to analyze multimodal images from many sources.*

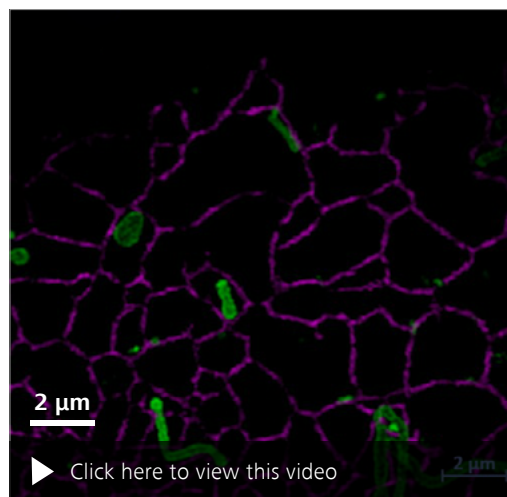
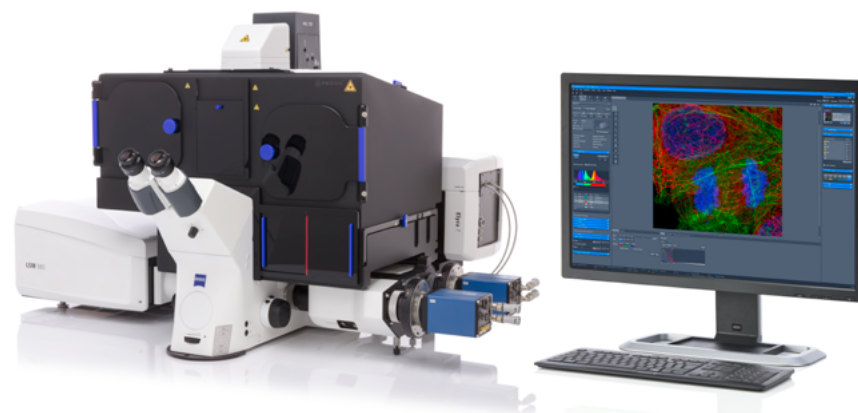
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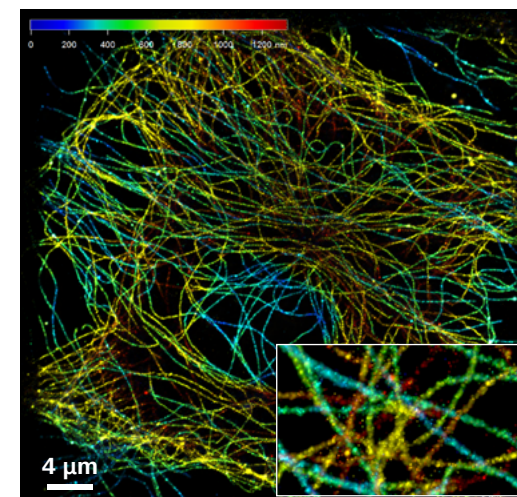
## Combine Multiple Superresolution Techniques

Combine your LSM 980 with Elyra 7 and Lattice SIM and you can always choose the best superresolution technique for your experiment at hand. The new Lattice SIM technology brings structured illumination microscopy (SIM) to a new level. Groundbreaking light efficiency gives you gentle superresolution imaging with incredibly high speed – at 255 fps you will get your data faster than ever before. 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 superresolution in one single system.



*Lattice SIM: In this movie you can see the ER membrane visualized with dTomato and Mitochondria with Tomm20-mEmerald in MEF cells. With ZEISS Elyra 7 equipped with the dual camera option, you can acquire two channels simultaneously.*



*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 alpha-tubulin color coded for depth. Sample courtesy of M. W. Davidson, Florida State University, USA.*

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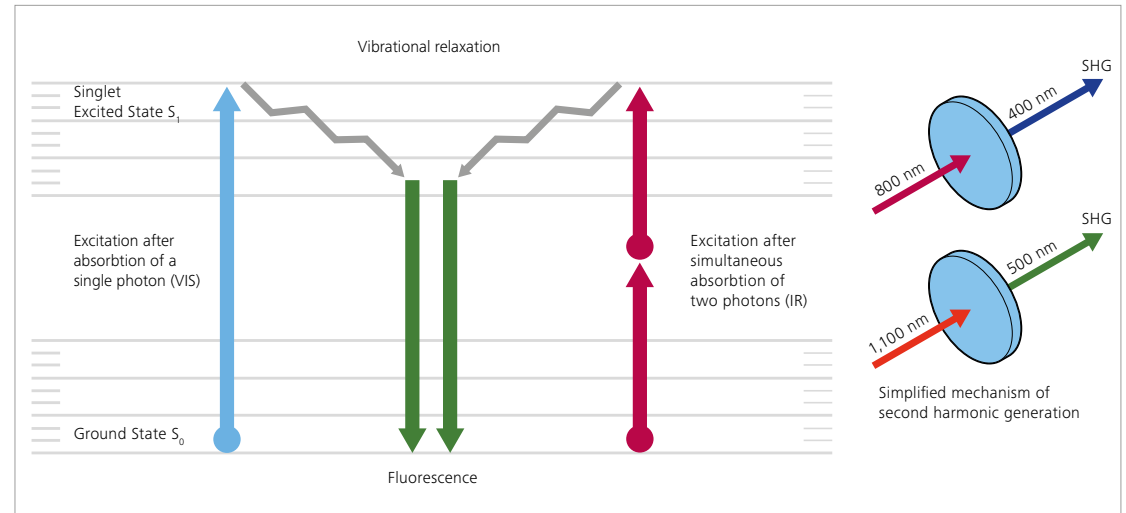
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## Multiphoton Microscopy

Multiphoton microscopy lets you acquire optical sections of deep tissue layers. This imaging method makes use of these basic principles:

- The longer the wavelength of light, the less it is scattered when entering tissue. Light of a wavelength between 600 and 1300 nm experiences the lowest absorption in tissue, making it nearly transparent in this spectral range.
- A fluorescent dye with an excitation maximum at 500 nm can be excited with one photon of this wavelength or with two photons of the doubled wavelength –1000 nm – that arrive simultaneously.
- A powerful pulsed tunable laser of 700 to 1300 nm makes sure that enough photons arrive simultaneously to excite the fluorescent dye. Outside the focal plane, the laser intensity drops exponentially and produces no emission light.

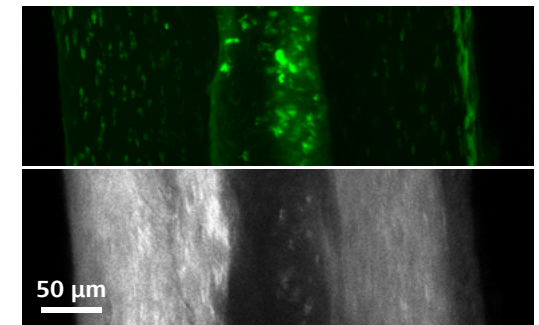
Emission light, created by multiphoton excitation, can be captured efficiently with non-descanned detectors. Using Airyscan detection with multiphoton excitation combines deep tissue penetration with increased sensitivity, resolution and speed.



Energy diagram of 2 Photon Microscopy

You can make use of these Airyscan advantages for functional imaging experiments, large volume imaging and screening applications.

Even non-stained structures can be visualized with multiphoton high intensity excitation by the non-linear effect of frequency doubling. This second harmonic generation (SHG) on non-centrosymmetric molecules with predominantly periodic alignment occurs, for example, in striated muscle and collagen.



Confocal microscopy and SHG helped to reveal dormant tumor cells (DTCs) in mice. Top: disseminated tumor cells labeled with NeonGreen. Bottom: long bones were subsequently cleared with Bone CLARITY protocol (Greenbaum et al., 2017) and SHG imaged. Sample courtesy of S. Stewart, Cell Biology & Physiology, Washington University School of Medicine in St. Louis, USA

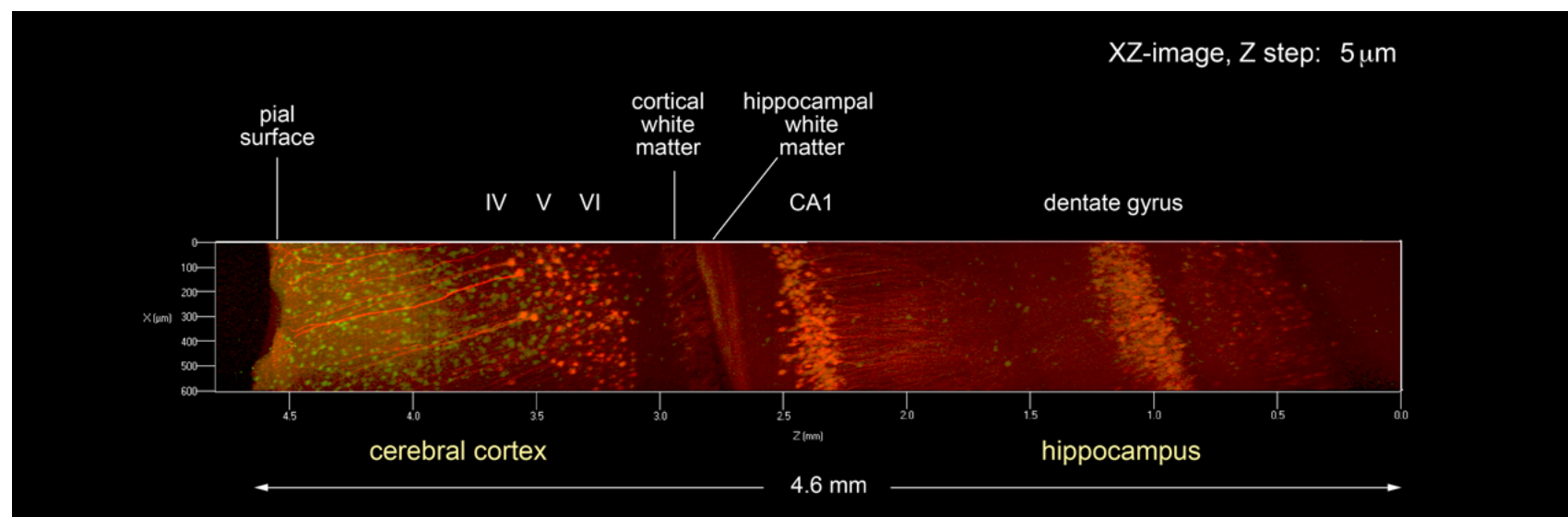
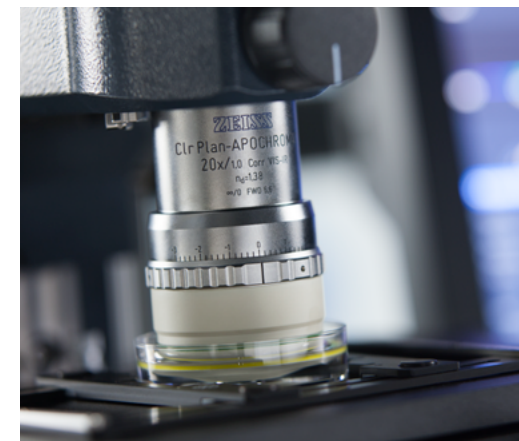
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## Image Large Cleared Samples

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 10×/0.5 nd=1.38, Clr Plan-Apochromat 20×/1.0 Corr nd=1.38 or Clr Plan-Neofluar 20×/1.0 Corr nd=1.45—you can look 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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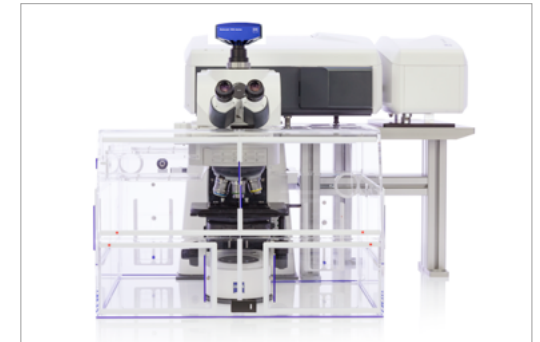
As your needs grow, your LSM 980 grows with you, forming the basis for a number of enhancements. Like every system from ZEISS, open interfaces and a modular architecture guarantee the seamless interaction of all components now and in the future. These include:



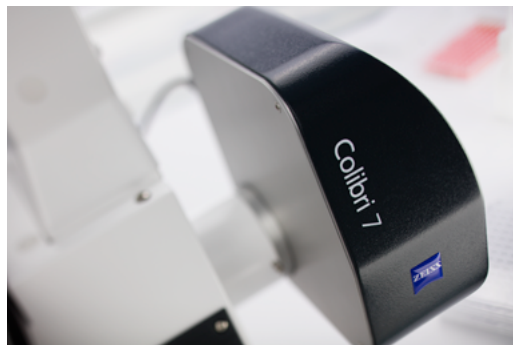
*Combine your ZEISS Axio Observer 7 with integrated incubation modules to create the perfect environment for long-term live cell imaging with stable temperature conditions.*



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



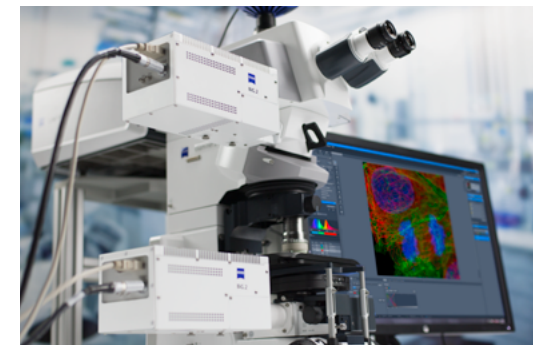
*You can also combine your ZEISS LSM 980 with Airyscan 2 and the upright research microscope ZEISS Axio Imager Z2. The optional incubation keeps even your most sensitive samples happy.*



*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 photon counting experiments and FLIM on your ZEISS LSM 980.*

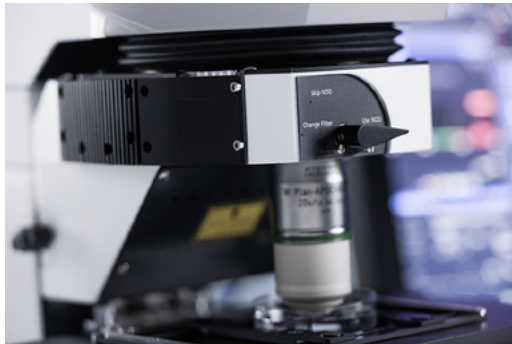


*Your BiG.2 works perfectly as a non-descanned detector, also providing a highly sensitive direct coupled detector for FLIM.*

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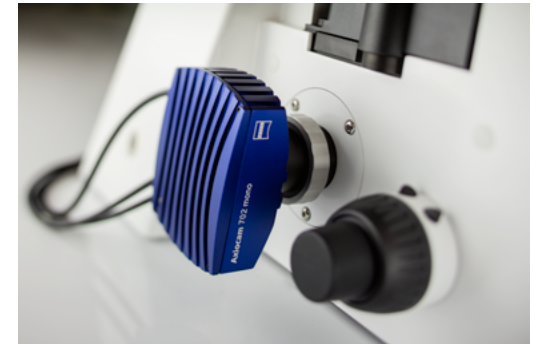
As your needs grow, your LSM 980 grows with you, forming the basis for a number of enhancements. Like every system from ZEISS, open interfaces and a modular architecture guarantee the seamless interaction of all components now and in the future. These include:



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



*With Autocorr objectives and ZEN imaging 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.*



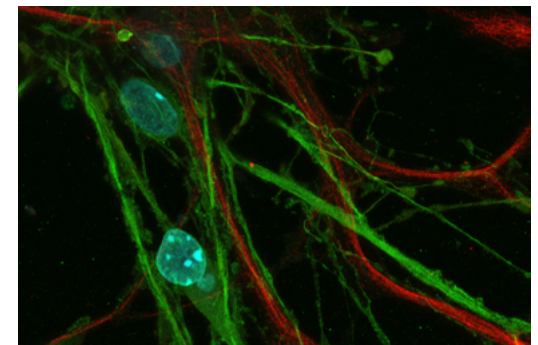
*You can add a choice of sensitive ZEISS Axiocams to your ZEISS LSM 980. It's very easy to acquire overview images for your multiposition experiments or to perform light efficient widefield imaging.*



*Shuttle & Find is your gateway to correlative light and electron imaging (CLEM). Combine the specificity of functional fluorescence imaging with ultrastructural information.*



*Definite Focus.2 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.*



*Collect all labels simultaneously with the numerous channels of ZEISS LSM 980 and accelerate the deconvolution process significantly with the CUDA enabled GPUs. Add enhanced resolution and signal to noise to the multi-channel flexibility of imaging with your ZEISS LSM 980.*

# Expand Your Possibilities

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- › The Applications
- › The System
- › Technology and Details
- › Service

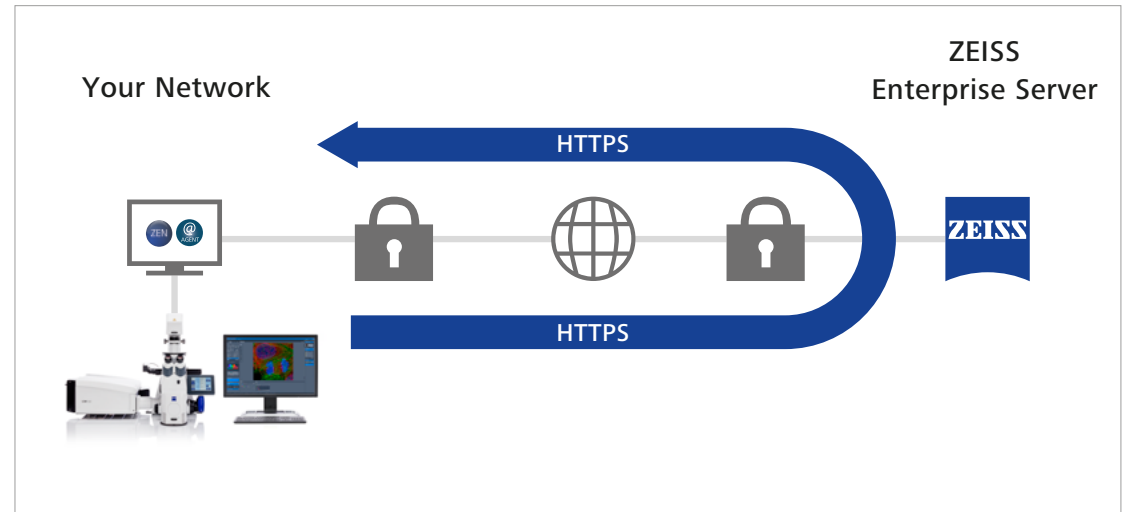
## ZEISS Predictive Service

### 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



# Tailored Precisely to Your Applications

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Typical Applications, Typical Samples	Task	ZEISS LSM 980 Offers
Antibody stained tissue slices	Document morphological relations of structures with resolution of down to 120 nms (XY)/ 350 nm (Z) at 488 nm excitation.	Airyscan 2 with SR and Multiplex mode for efficient superresolution imaging
	Acquire large fields of view and tiling experiments	
Cleared tissue	Image cleared tissue with up to 5.6 mm in Z	Special objective corrected for immersion medium of refractive index 1.38 or 1.45 working with confocal or multiphoton imaging on Axio Examiner
Live cell culture	Study the motility of vesicles and organelles	Airyscan 2 in Multiplex mode for gentle imaging with high frame rates
	Follow fast processes such as Calcium waves, muscle contractions, blood flow, cilia beating while keeping structural information.	Airyscan 2 with Multiplex mode for gentle imaging at very high frame rates at confocal resolution
	Screen and document cells expressing the desired fluorescent label in response to pharmacological treatment	Widefield imaging using AxioCam
Live cell culture with two labels	Study the motility of subcellular structures	Airyscan 2 with GaAsP detector to image 2 colors with time lapse imaging in 2D or 3D at 2.4 frames per second and up to 23 frames per second in Multiplex mode
	Explore the interaction of two proteins with fluorescent lifetime microscopy	BiG.2 as detector for FLIM and third party electronics and software*
	Explore the interaction of two proteins exploiting the Förster Resonance Energy Transfer effect	FRET analysis tool
Live cells with multiple labels	Image over long time in an automated way	Experiment Designer to acquire complex experiments. Combination of different acquisition modes of the LSM system, e.g. spectral imaging, tiling with Airyscan 2 Multiplex mode at superresolution. Combine all findings with ZEN Connect.
Fixed and living cultured specimens	Document cellular structures in superresolution in 3D with 2x the resolution of a confocal	Structured illumination with ELYRA 7
Live or fixed cells with multiple labels and overlapping emission signals	Examine the interplay of multiple proteins	Parallel acquisition of all signals with spectral imaging at 5 full frames per second and online or post processed linear unmixing
Cellular structures with weak labels	Image subcellular structures at physiological expression levels	LSM 980 with Airyscan 2 with GaAsP detectors
Living organisms/ animals	See the interaction of cells within living tissue	Multiphoton extension of LSM 980
	Imaging of live tissue with cells expressing multiple different fluorescent proteins	Extension of LSM 980 NLO

(\* available upon request)

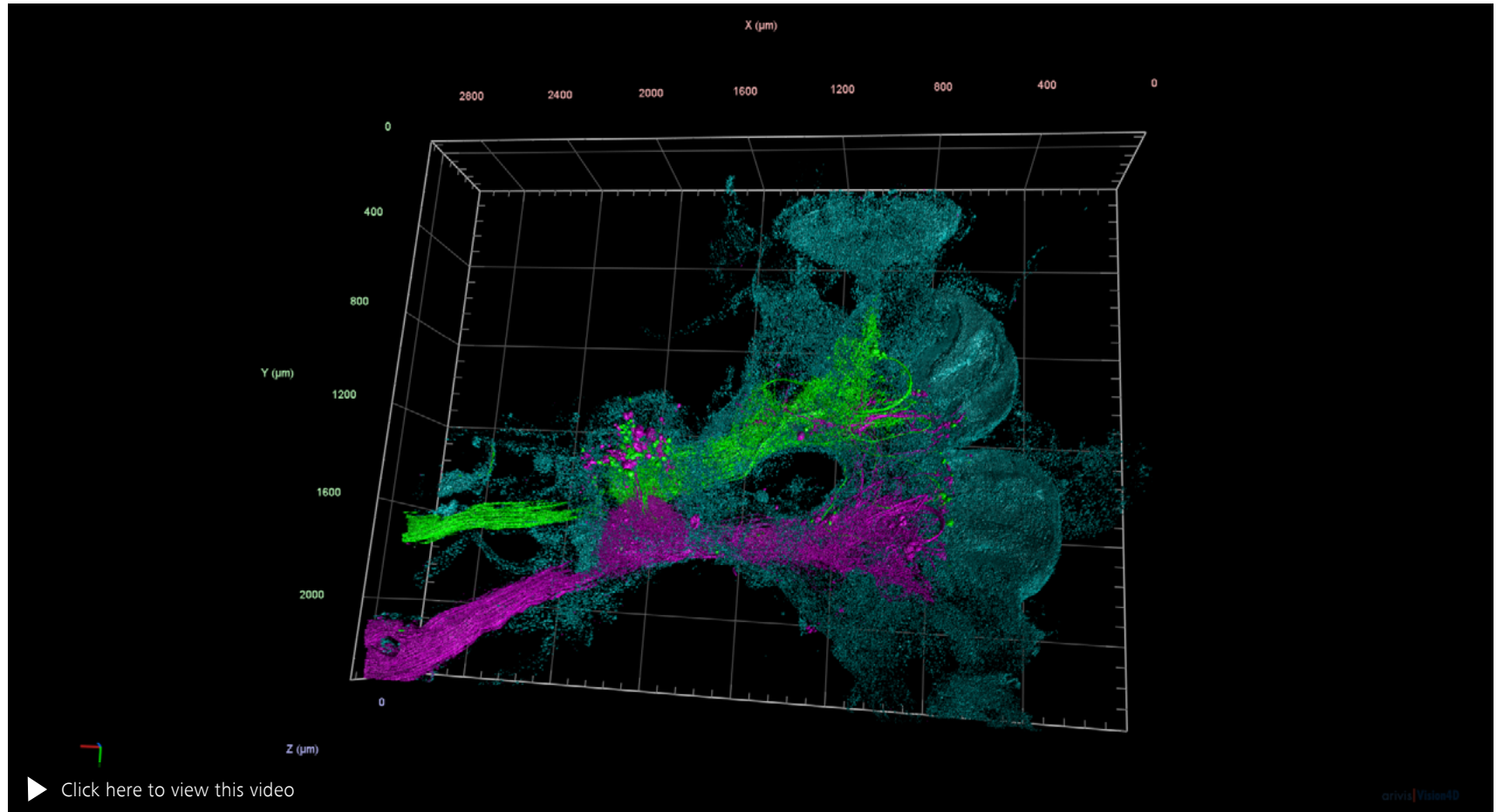
# Tailored Precisely to Your Applications

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Typical Applications, Typical Samples	Task	ZEISS LSM 980 Offers
Plant roots	Follow the changes of subcellular structures over time with a high resolution	Airyscan 2 with GaAsP detector for superresolution imaging beyond 40 µm deep into tissue with up to 47 frames per second (512x512)
Model organisms, e.g. Zebrafish, <i>Drosophila</i> or <i>C. elegans</i>	See fine details of the organisation and dynamics of endogeneously expressed FP proteins	Airyscan 2 with GaAsP detector for superresolution imaging beyond 40 µm deep into tissue
	Image large fields of view at high volume rate to capture developmental processes	Airyscan 2 with Multiplex mode for high frame rates at confocal resolution

# ZEISS LSM 980 at Work

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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 arivis Vision 4D, ideal for rendering and analyzing large datasets. The 4D viewer in arivis Vision 4D 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

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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."

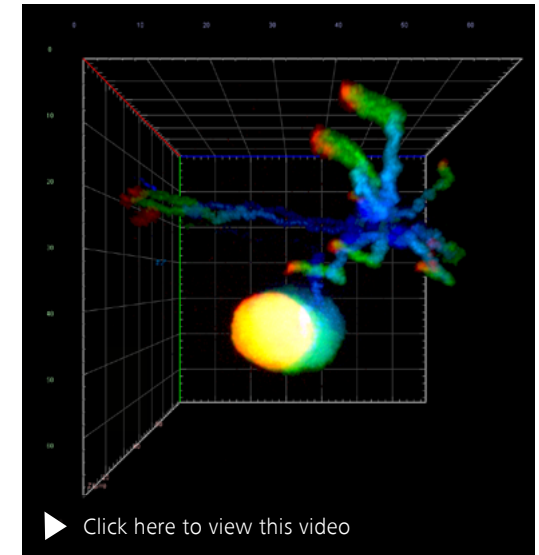


### *Meiosis in starfish oocytes*

*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*

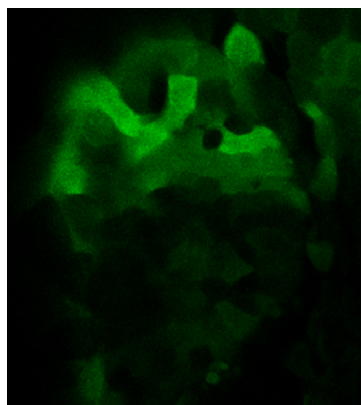
*The rendering is a projection of the process along z-axis (maximum intensity) and time (color-coded projection); to illustrate the movement of the chromosomes within the volume of the nucleus.*

### Reference:

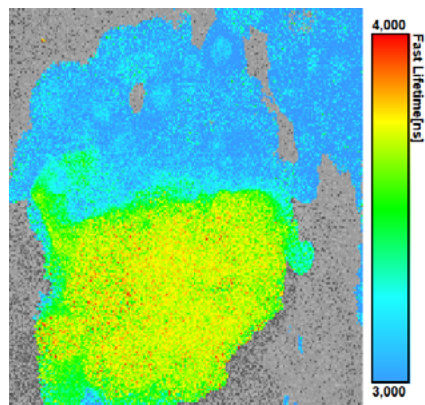
- Lenart P, et al. Nature. 2005 Aug 11;436(7052):812-8.
- Mori M, et al. Curr Biol. 2011 Apr 12;21(7):606-11.
- Bun P, et al. Elife. 2018 Jan 19;7. pii: e31469. doi:10.7554/eLife.31469.
- Burdyniuk M, et al. J Cell Biol. 2018 Aug 6;217(8):2661-2674.

# ZEISS LSM 980 at Work

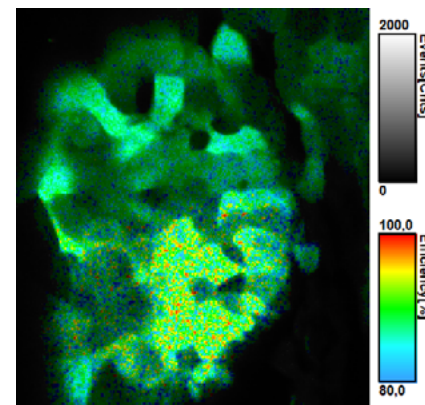
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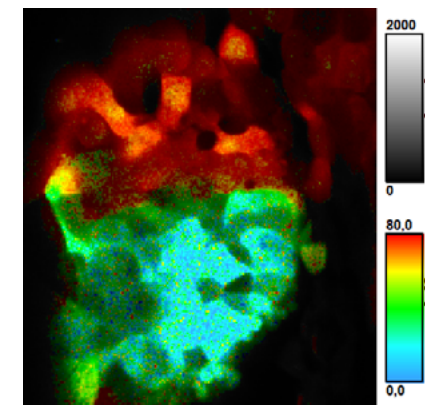
A) YFP intensity (bottom photobleached)



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)

## Fluorescent 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  $<10$  nm. 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 intramo-

lecular 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.

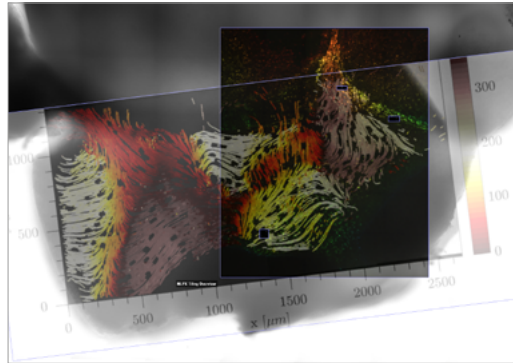
Quantitative FLIM-FRET analysis allows determining the spatial and temporal activity of two or more interacting molecules. In contrast to measuring FRET by photobleaching or intensity ratio

imaging, lifetime imaging allows precise quantification of FRET Efficiency. FLIM-FRET also allows quantification of the binding fraction for a particular intermolecular FRET pair and the fraction of active sensors for an intramolecular FRET biosensor using a suited FRET pair.

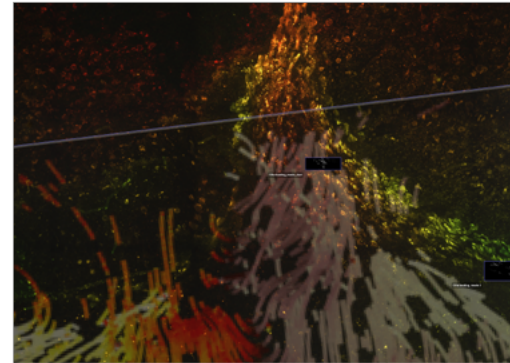
Data was obtained with LSM Systems with a PicoQuant FLIM & FCS upgrade kit using the PicoQuant FLIM module in ZEN imaging software. Lifetime measurements of CFP were performed with the NLO laser at 840 nm wavelength and 80 MHz repetition rate; acceptor photobleaching was performed with the 514 nm laserline; analysis was performed within PicoQuant's SymPhoTime64.

# ZEISS LSM 980 at Work

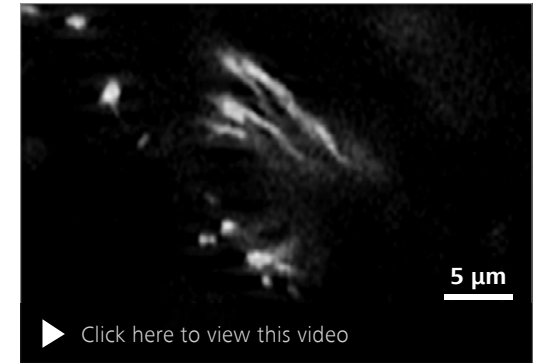
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This ZEN Connect project documents the experiment performed with the tissue explant of ependyma from the ventricular system of a mouse brain. All acquired data of the experiment session is kept in context. The overview images by camera and LSM allow to precisely record the localization of the acquired ciliary beating within the sample. The flow map of cilia generated flow along the ependymal wall is added as a reference.



An overview of fluorescently labeled motile cilia on ependyma tissue explant from the mouse brain is quickly acquired by tiling with Airyscan 2 in Multiplex CO-8Y mode to find regions of interest. Z-Stack displayed in colored depth coding. The exact position of the recorded motile cilia is documented.



Live imaging with 143 frames per second of fluorescently labeled motile cilia of brain ependyma. Acquired with Airyscan CO-8Y mode combining image quality and speed; for detailed analysis of ciliary beating direction and frequency.

## Reference for all images:

G. Eichele, Department of Genes and Behavior, Max Planck Institute for biophysical Chemistry, Göttingen, 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
- Camera port
- Manual or motorized stages
- Incubation solutions
- Fast Z piezo inserts
- Definite Focus

## 2 Objectives

- C-APOCHROMAT
- Plan-APOCHROMAT
- W Plan-APOCHROMAT, Clr Plan-APOCHROMAT, Clr Plan-NEOFLUAR
- LCI Plan-APOCHROMAT

## 3 Illumination

- UV laser: 405 nm
- VIS laser: 445 nm, 488 nm, 514 nm, 543 nm, 561 nm, 594 nm, 639 nm
- NIR laser for multiphoton imaging: Ti:Sa, InSight DeepSee\*

## 4 Detection

- 3, 6, or 34 descanned spectral channels (GaAsP and multialkali PMT)
- Airyscan 2 detector with optional Multiplex module
- 2 additional GaAsP channels (BiG.2)
- Up to 6 non-descanned GaAsP detectors
- Up to 12 non-descanned GaAsP or PMT detectors total
- Transmitted light detector (T-PMT)

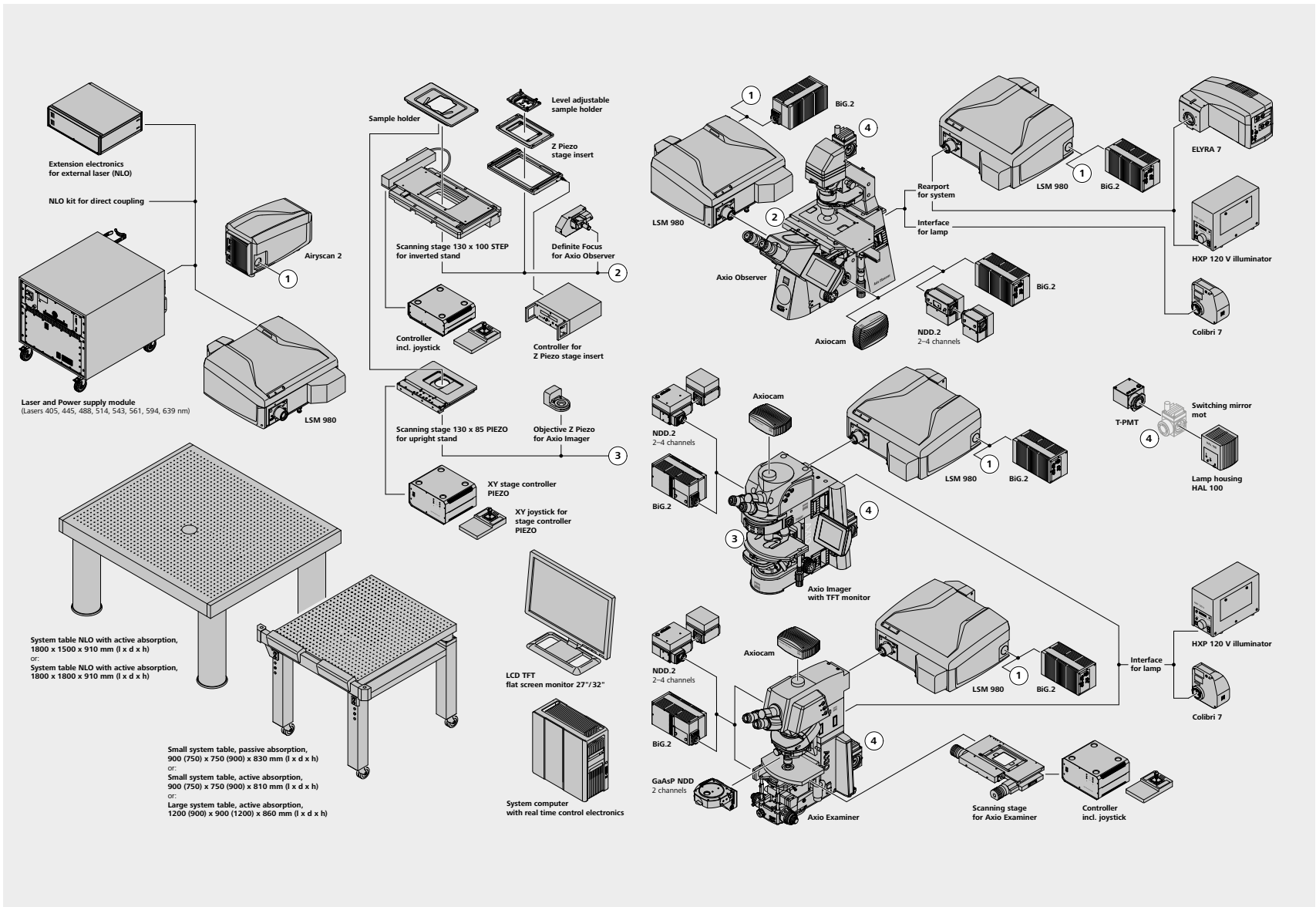
## 5 Software

- ZEN imaging software, highlighted modules: Tiles & Positions, Experiment Designer, FRAP, FRET, RICS, Deconvolution, 3Dxl Viewer – powered by arivis®

(\* available upon request)

# System Overview

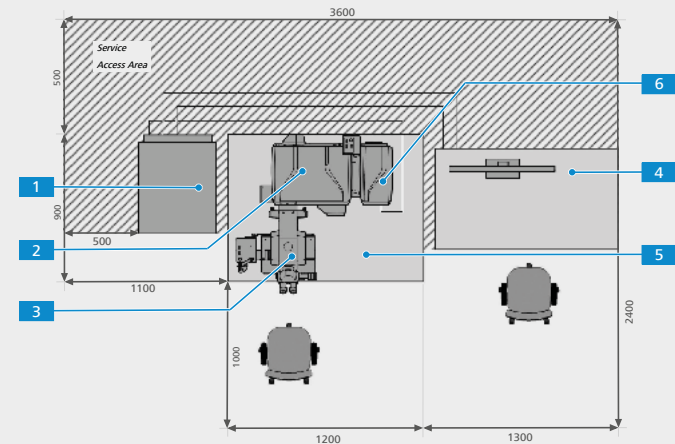
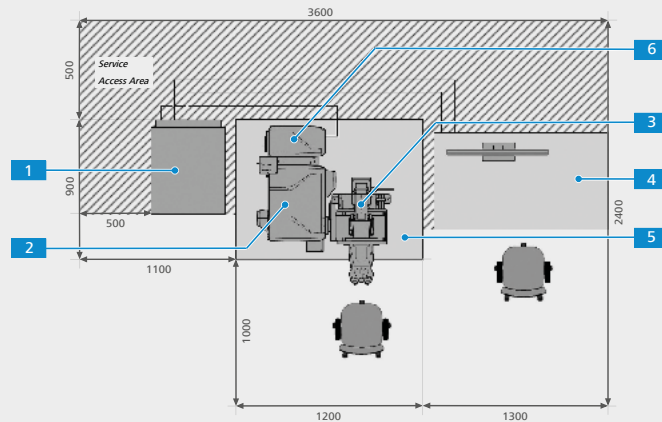
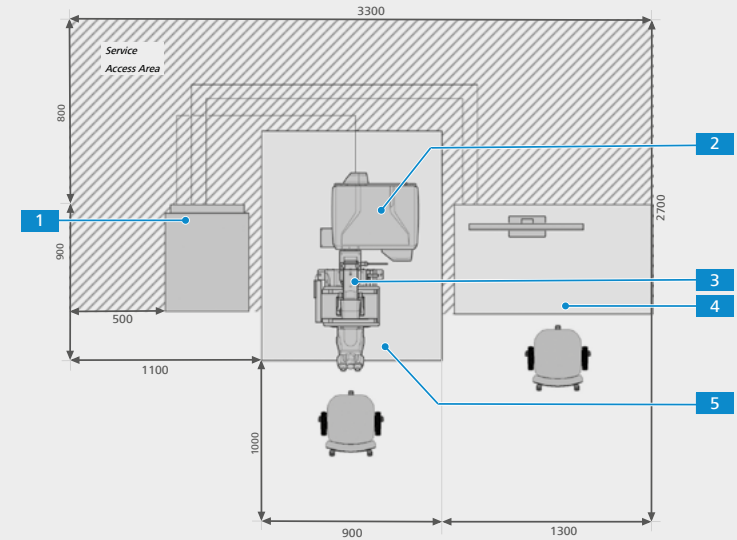
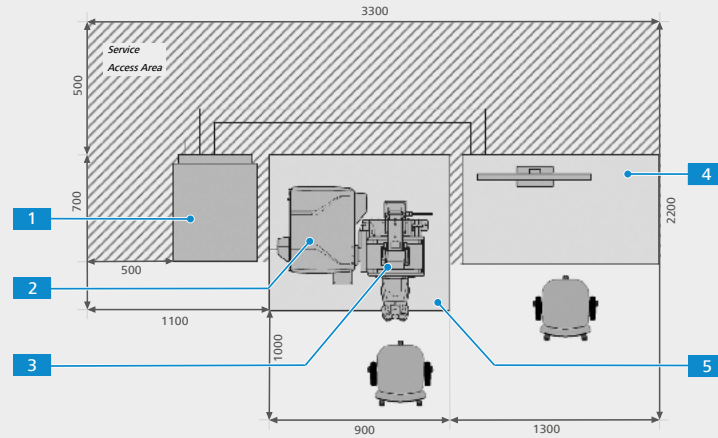
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# Technical Specifications

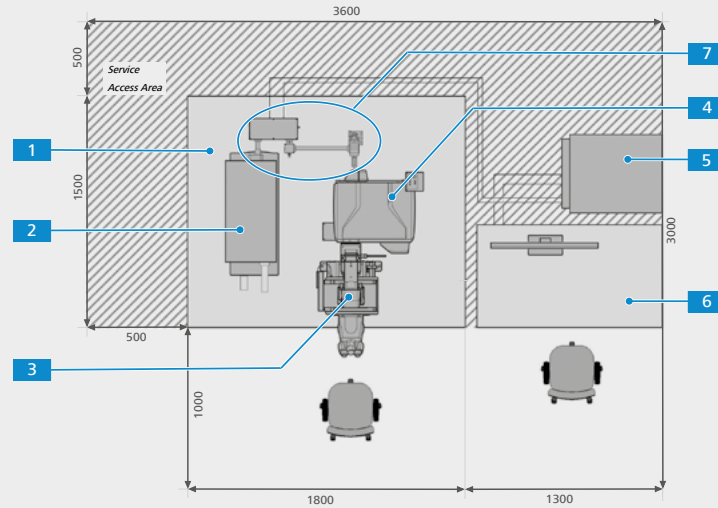
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1 Laser and Power Supply Module 2 LSM 980 Scanhead 3 Microscope Stand (Axio Observer, Axio Imager or Axio Examiner) 4 Computer Table 5 System Table 6 Airyscan 2

# Technical Specifications

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- 1 System Table
- 2 Multiphoton Laser
- 3 Microscope Stand (Axio Observer, Axio Imager or Axio Examiner)
- 4 LSM 980 Scanhead
- 5 Laser and Power Supply Module
- 6 Computer Table
- 7 Laser coupling with AOM for Multiphoton laser

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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) for Mai Tai Laser or Chameleon	180	150	75	200
Active Anti-Vibration Table (NLO) for Two-Microscope Configuration	250	150	75	400
Scanning Module LSM 980	50	45	22	27
Microscope	50	35	70	40
Plug-in Unit External Laser	70	55	25	10
Laser Module UV	80	60	45	40
Airyscan 2	40	20	24	12
Fiber Optic Cable, UV	200			
Fiber Optic Cable, VIS	250			
Cables	250			

Microscopes	
Stands	Upright: Axio Imager.Z2, Axio Examiner.Z1 Inverted: Axio Observer 7 with side port or rear port
Z Drive	Smallest increment Axio Imager.Z2: 10 nm; Axio Observer 7: 10 nm; Axio Examiner: 25 nm; 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); 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; Airyscan SR – up to 4.7 fps; Multiplex SR-4Y – 25 fps; Multiplex SR-8Y – 47.5 fps; Multiplex CO-8Y – 34.4 19 × 2 speed levels for confocal; 512 × 16 pixels up to 425 fps; up to 6830 lines/sec. 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: 0.67 × 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. 18 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 external detection modules (such as 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 spectral detection channels (QE 45% typical for GaAsP) 2 additional GaAsP detection channels (BiG.2) Airyscan 2 detector (32 channels GaAsP), delivers resolution up to 120 nm lateral, 350 nm axial; Multiplex resolution: 140 / 160 nm lateral, 450 nm axial Up to 12 non-descanned detection channels (PMT and/or GaAsP) Transmitted light detector (PMT)
Spectral Detection	3, 6 or 34 simultaneous, confocal reflected-light channels, GaAsP and PMT based freely adjustable spectral detection area (resolution down to 3 nm)
Data Depth	8 bit or 16 bit available; up to 35 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

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<b>ZEN Imaging Software</b>	
System Configurations	Workspace to conveniently configure all of the motorized functions of the scanning module, laser and microscope; Save and restore application configurations (Reuse)
Calibration Tools	Calibration and testing tools 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 ratio images, average and summation (by line/image, adjustable), Step Scan (for higher image frame rates); Quick set up of imaging conditions using Smart Setup by simply selecting the labelling dye
Crop Function	Easily select scanning areas (simultaneously select zoom, offset, rotation)
Real ROI Scan, Spline Scan	Scans multiple ROIs (regions of interest) as desired and pixel-by-pixel laser blanking; 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; Use of different speeds for bleaching and imaging, 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 module
Lambda Scan	Parallel or sequential acquisition of image stacks with spectral information for every pixel
Linear Unmixing	Acquisition of crosstalk-free, multiple fluorescence images using simultaneous excitation; Online or offline and automatic or interactive unmixing; Advanced unmixing logic with indication of reliability
Visualization	XY, orthogonal (XY, XZ, YZ), Cut (3D section); 2.5D for time series of line scans, projections (maximum intensity); animations; Depth coding (inverse colors), brightness, gamma and contrast settings; color table selection and modification (LUT), character functions
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
3DxL Viewer powered by Arivis	Rapid 3D and 4D reconstructions and animations (available modes: shadow projections, transparency projection, surface rendering)

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## Optional Software

Direct Processing	Processing of large datasets during acquisition by streaming technology, including analysis and storage on second computer
Deconvolution	3D, GPU based Cuda 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
Physiology	Comprehensive evaluation software for online and offline calibration of ion concentrations
FRET	Acquisition of FRET (Förster resonance energy transfer) image data with subsequent evaluation; Acceptor Photobleaching and Sensitized Emission methods supported
FRAP Efficiency Analysis	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)
Experiment Designer	Defintion of customized imaging configurations and procedures
Open Application Development	Python scripting interface for automation & customization; experimental feedback for smart experiments and open interface to third party software (e.g. ImageJ)
ZEN Connect	Exchange and alignment of image data from multiple image acquisition systems
ZEN Intellesis	Image analysis and structure detection via computational self learning technology

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## Lasers

Laser RGB (445, 488, 514, 543, 561, 594, 639 nm)	Single-mode polarization preserving fiber
	Laser beam attenuation for all lasers by VIS-AOTF
	Diode Laser 445 nm (30 mW nominal power; 7.5 mW ex fiber)
	Diode Laser 488 nm (30 mW nominal power; 10 mW ex fiber)
	Diode Laser 514 nm (30 mW nominal power; 10 mW ex fiber)
	DPSS Laser 543 nm (25 mW nominal power; 10 mW ex fiber)
	DPSS Laser 561 nm (25 mW nominal power; 10 mW ex fiber)
	DPSS Laser 594 nm (8 mW nominal power; 2.5 mW ex fiber)
Laser V (405 nm)	Single-mode polarization preserving fiber
	Laser beam attenuation via direct modulation
	Diode Laser 405 nm (30 mW nominal power; 14 mW ex fiber)

## Power Requirements

LSM 980 has a main power supply cord and plug, either NEMA L5-15 (100V - 125V) 2pol (15A) + PE; CEE blue (200–230V) 2pol (16A) + PE; CEE red (200–230V) 4pol (16A) + PE (Phase L2/L3 not used) and the matching mains socket outlet.

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	12 A at 120 V
Heat emission without Ti:Sa	1,400 W max.	1,400 W max.
Power Consumption	1,500 VA max.	1,500 VA max.

## Multiphoton Laser

### Power Consumption

Ti:Sa laser	800 VA max.	800 VA max.
-------------	-------------	-------------

## EMC test

according to DIN EN 61326-1:2013

1. Noise emission according to CISPR 11 / DIN EN 55011:2011
2. Noise immunity according to table 2 (industrial sector)

\* 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) 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. Storage, less than 6 h	T = -20 °C to 55 °C
5. Temperature gradient	±0.5 °C/h
6. Warm up time	1 h, for high-precision and/or long-term measurements ≥3 h
7. Temperature gradient and range for continuous long-term image acquisition	± 0.5 °C/h, ± 1.5 °C/ 12 h
8. Relative humidity	<65 %
9. Operation altitude	max. 2,000 m
10. Loss of heat (without Ti:Sa)	1.4 kW
11. Vibrations under operation conditions (with system table)	max. 5 µm pp at 0 – 5 Hz max. 10 µm pp at 5 to 20 Hz
12. Shipping shock (LSM 980 box)	10 g



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



# Count on Service in the True Sense of the Word

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

Because the ZEISS microscope system is one of your most important tools, we make sure it is always ready to perform. What's more, we'll see to it that you are employing all the options that get the best from your microscope. You can choose from a range of service products, each delivered by highly qualified ZEISS specialists who will support you long beyond the purchase of your system. Our aim is to enable you to experience those special moments that inspire your work.

## **Repair. Maintain. Optimize.**

Attain maximum uptime with your microscope. A ZEISS Protect Service Agreement lets you budget for operating costs, all the while reducing costly downtime and achieving the best results through the improved performance of your system. Choose from service agreements designed to give you a range of options and control levels. We'll work with you to select the service program that addresses your system needs and usage requirements, in line with your organization's standard practices.

Our service on-demand also brings you distinct advantages. ZEISS service staff will analyze issues at hand and resolve them – whether using remote maintenance software or working on site.

## **Enhance Your Microscope System.**

Your ZEISS microscope system is designed for a variety of updates: open interfaces allow you to maintain a high technological level at all times. As a result you'll work more efficiently now, while extending the productive lifetime of your microscope as new update possibilities come on stream.



*Profit from the optimized performance of your microscope system with services from ZEISS – now and for years to come.*

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



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