

VivaTome

Discover the Dynamics of Life



The Entry-level System that Captures Dynamic Processes with Outstanding Image Quality.



We make it visible.

Innovative Technology Captures Dynamic Processes with Outstanding Image Quality.

In collaboration with Aurox Ltd., Carl Zeiss introduces a pioneering technology: VivaTome, an economical entry-level solution for high resolution, artifact-free and light efficient imaging of highly dynamic processes in living samples in optical sections.

New technology for fast optical sectioning

VivaTome is the solution of choice when expanding your microscope system to observe dynamic processes in living samples. The technology behind VivaTome is as innovative as efficient: It combines the speed of a spinning disk with the light efficiency of structured illumination, resulting in rapid frame rates that allow the capture of fast processes in living samples.

• Artifact-free optical sections of dynamic processes

This innovative technology eliminates motion artifacts even at high frame rates.

• High temporal resolution

Up to 30 images per second can be captured using VivaTome, which is fast enough to capture all stages of dynamic events.

• Optimal signal-to-background ratio

Two different grids are available to ensure optimal sectioning strength and a good signal-to-background ratio with various objectives and samples.

• Simultaneous sectioned and widefield images

VivaTome can simultaneously acquire both widefield and sectioned images, increasing the amount of image information from your sample.

• Flexible choice of excitation wavelengths

Since VivaTome works with a white light source, it allows for a range of wavelengths. Different filter units are available for a wide variety of dyes.

• Compatible with many Carl Zeiss microscope stands

With its flexible optical concept and compact form, VivaTome is designed to be easily integrated into a variety of Carl Zeiss microscope stands such as Axio Observer, Axio Imager and Axio Examiner.



- **Easy to use**

You don't have to be an expert to use VivaTome for optical sectioning. It is as easy to operate as any wide-field microscope.

- **Driven by AxioVision**

VivaTome is fully integrated in the Carl Zeiss AxioVision software. AxioVision allows the user to control all of the system's components such as microscope objectives and cameras necessary for the task at hand. Images can be analyzed in detail using AxioVision as well.

- **Integration into multimodal systems**

VivaTome can be combined with other imaging modalities on the same microscope, resulting in flexible systems that are useful for a variety of applications.



A New Direction for Imaging Technology. Impressive Performance in Every Detail.

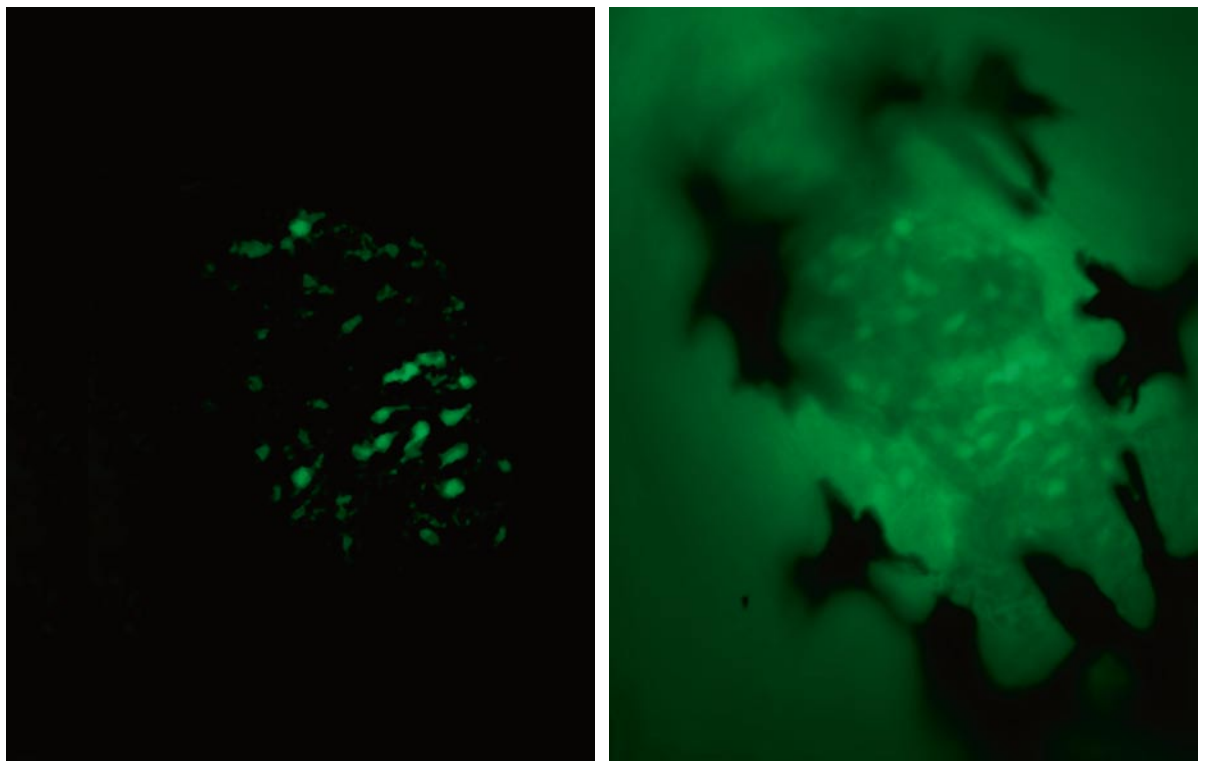
VivaTome is a new approach to an established scientific process: taking optical sections to distinguish weak fluorescent signals from the background. With VivaTome, this can now be accomplished at high frame rates using simple instrumentation, for blur-free images of dynamic processes.

Comprehensive image information: widefield and optical section

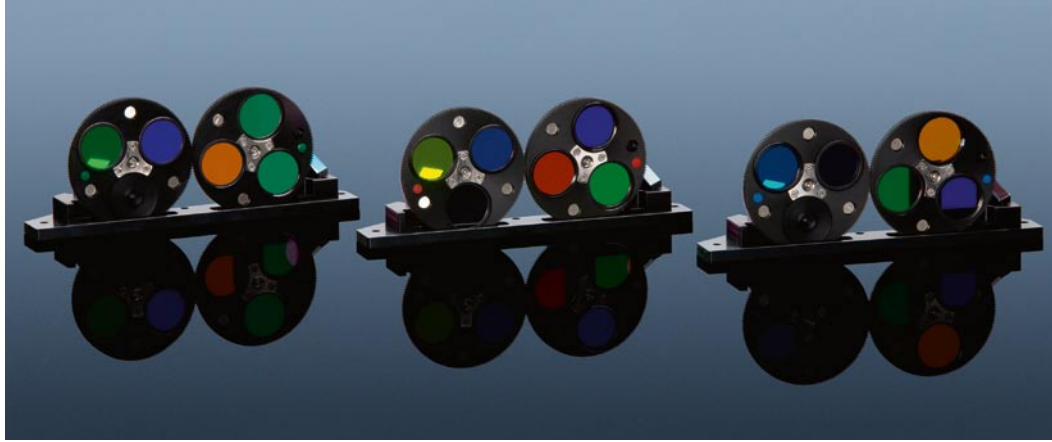
VivaTome collects all the data necessary for calculating an optical section in a single image. Any motion artifacts that may arise due to sample movements are eliminated. The system can simultaneously deliver both widefield images and optical sections, guaranteeing comprehensive image information. The data from both techniques can be compared, analyzed, and processed without interrupting the workflow.

Those crucial fractions of a second: high frame rates to capture living processes

Capturing dynamic processes in living samples requires an imaging system that is both precise and fast. VivaTome can achieve frame rates of up to 30 images per second, depending on exposure time and camera. With the right settings even the fastest processes can be captured at full temporal resolution. Whether used for examining tissue growth, cellular transport, or signal transduction – VivaTome delivers high-resolution optical sections free of background fluorescence.



Transgenic 26-day old zebrafish larva TG (flk1: GFP) showing fluorescent cardiac endothelial cells in the contracting ventricle. 20x/0.8 Plan-APOCHROMAT. Left: optical section, right: widefield image. The corresponding raw data image is shown on the title page. TinChung Leung, North Carolina Central University, USA



A variety of filtersets is available for VivaTome which can be easily exchanged.

The right resolution for every sample

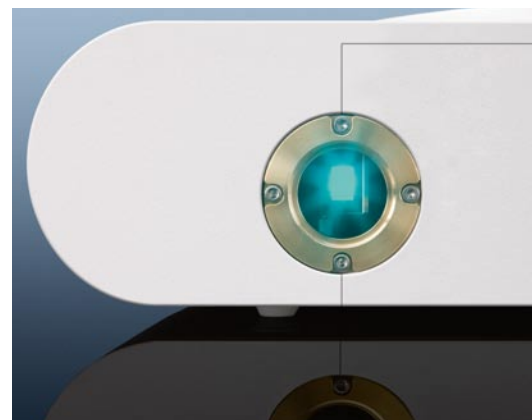
Resolution requirements differ from application to application. For instance, precise localization requires high z-resolution. And for weak signals in thick samples with a lot of background fluorescence, an optimized signal-to-background ratio may be more important. With VivaTome you can choose between two different grid patterns to fit your applicational needs.

Multiple wavelengths for greater versatility

VivaTome uses a white light source making the entire visible spectrum available for excitation. Since its filter units can be changed with minimum effort, the system is configured easily to image various common dye combinations, with configurations for new dyes or combinations added as they are needed.

Modular and compatible: fully integrated in AxioVision

The integration of VivaTome in the AxioVision system software provides full control of the entire experiment and precise command of the microscope system. Different image acquisition modes, numerous evaluation and analysis functions are available, which can be added when necessary. The same is true for the microscope stand: VivaTome is quickly mounted on the camera interface of many Carl Zeiss microscopes. As the applications' demands grow, your system grows along with it.



The VivaTome Principle: Aperture Correlation

VivaTome's technical innovation is based on the principle of aperture correlation, which combines the speed of a spinning disk with the light efficiency of structured illumination.

Optical sectioning made fast and easy

Optical sectioning can be achieved by a variety of technical approaches. The structured illumination technique utilized with ApoTome from Carl Zeiss demonstrates one of the easiest and most efficient ways to implement optical sectioning capabilities into a normal white light widefield system. VivaTome pushes structured illumination into the domain of live cell imaging, with frame rates high enough to capture even fast events in detail. The underlying principle, which allows high acquisition rates while maintaining most of the ease-of-use and light efficiency of a structured illumination system, was pioneered by Tony Wilson and coworkers from Oxford University^{1, 2, 3}. After founding Aurox Ltd. they have now turned this idea into a product – VivaTome.

A disk with a grid structure for maximum light efficiency

At the heart of VivaTome is a spinning disk in a conjugate image plane. Excitation light passes through this disk before reaching the sample and the emission light passes through the disk again. The disk in VivaTome is optimized for light efficiency: Instead of individual pinholes, like in Nipkow disk systems, VivaTome uses a grid pattern. With this structure up to 50% of the excitation light from the economical white light source reaches the sample. And the white light means that the full spectrum is available, overcoming limitations normally in place when using laser lines for excitation.

Two grid frequencies for outstanding results

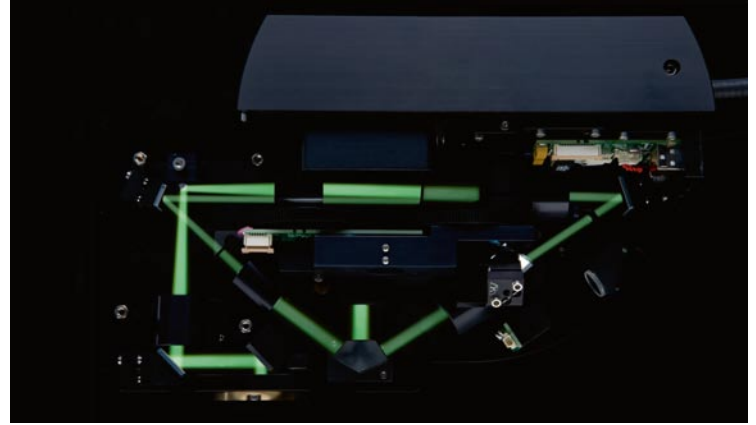
Two different grid patterns optimize results from various objectives. Both grid patterns are arranged on a single disk, and switching between grids is automated.



Two different grid patterns are arranged on a single disk to easily change easily when using different objectives.



A high precision prism is used to combine both images on one single CCD chip.



Excitation (left) and emission (right) beams in the open VivaTome unit.

Single shot for optical sectioning

The emission detected through the disk is not an optical section: In addition to information from the focal plane, a significant amount of fluorescence from outside the focal plane also passes through the disk. To calculate an optical section from this image, a second image is needed, containing only the out-of-focus information. By making the side of the disk that faces the sample reflective, the remaining out-of-focus signal is collected, instead of being blocked by the disk. The light reflected by the disk is exactly the information needed to calculate the optical section. Since both the emission light detected through the disk and the light reflected by the disk are imaged with a single camera, all the information necessary to calculate an optical section is contained in a single image.

Optical section calculation

The image detected through the disk (I_1) contains signal from the focal plane and a portion of signal from outside the focal plane. To calculate an optical section, this signal from outside the focal plane must be subtracted. The second image reflected by the disk (I_2) contains only intensities from outside the focal plane and is used to subtract the out-of-focus information from the image

detected through the disk. An optical section is obtained by simple scaled subtraction of the two images:

$$I_1 - k \cdot I_2 = I_{\text{sectioned}}$$

Scaled addition of the two images in turn yields a conventional widefield image:

$$I_1 + k \cdot I_2 = I_{\text{widefield}}$$

(*k = a constant that corrects differences in the light efficiency of the two imaging pathways)

Calibration

It is crucial to precisely overlay the two images to ensure that artifacts are not introduced, when the images are subtracted from one another. The two imaging beam paths are thus calibrated to sub-pixel precision. An internal calibration target with a dense random point pattern is used to identify relative differences between the two image halves. These two image halves are then registered, resulting in artifact-free, optically sectioned images.

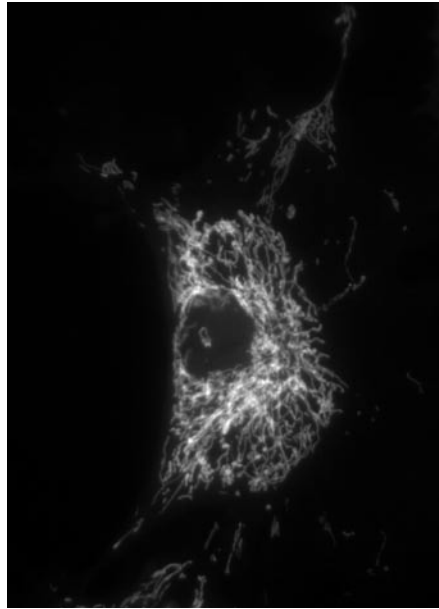
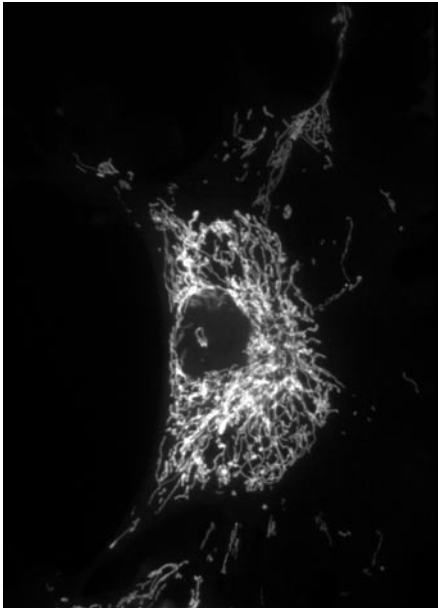
¹ Juskaitis, R., Wilson, T., Neil, M. A., Kozubek, M., Efficient real-time confocal microscopy with white light sources. Nature 1996, 383, 804-6.

² Wilson, T., Juskaitis, R., Neil, M. A., Kozubek, M., Confocal microscopy by aperture correlation. Opt. Lett. 1996, 21, 1879-1881.

³ Neil, M. A., Wilson, T., Juskaitis, R., A light efficient optically sectioning microscope. J. Microsc. 1998, 189, 114-117.



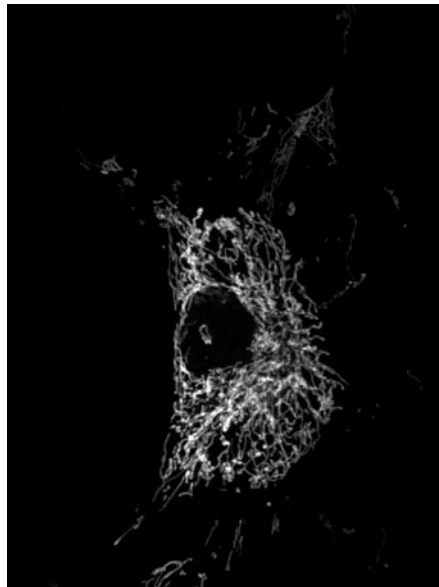
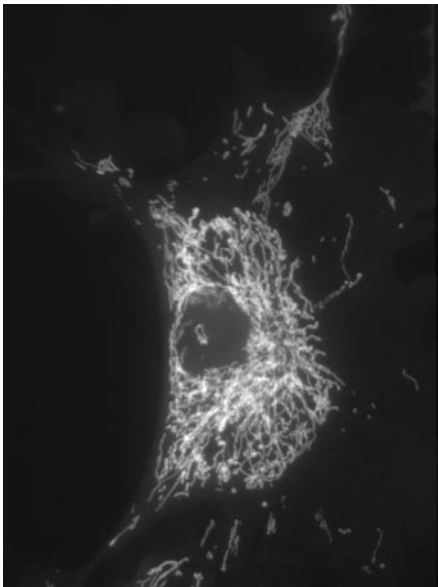
The raw data image captured by VivaTome is a side-by-side view of an image detected through the disk (left side) and the image reflected at the disk (right side).



In the first step of image calculation, both images are extracted from the raw data image, one image is mirrored and both images are registered.

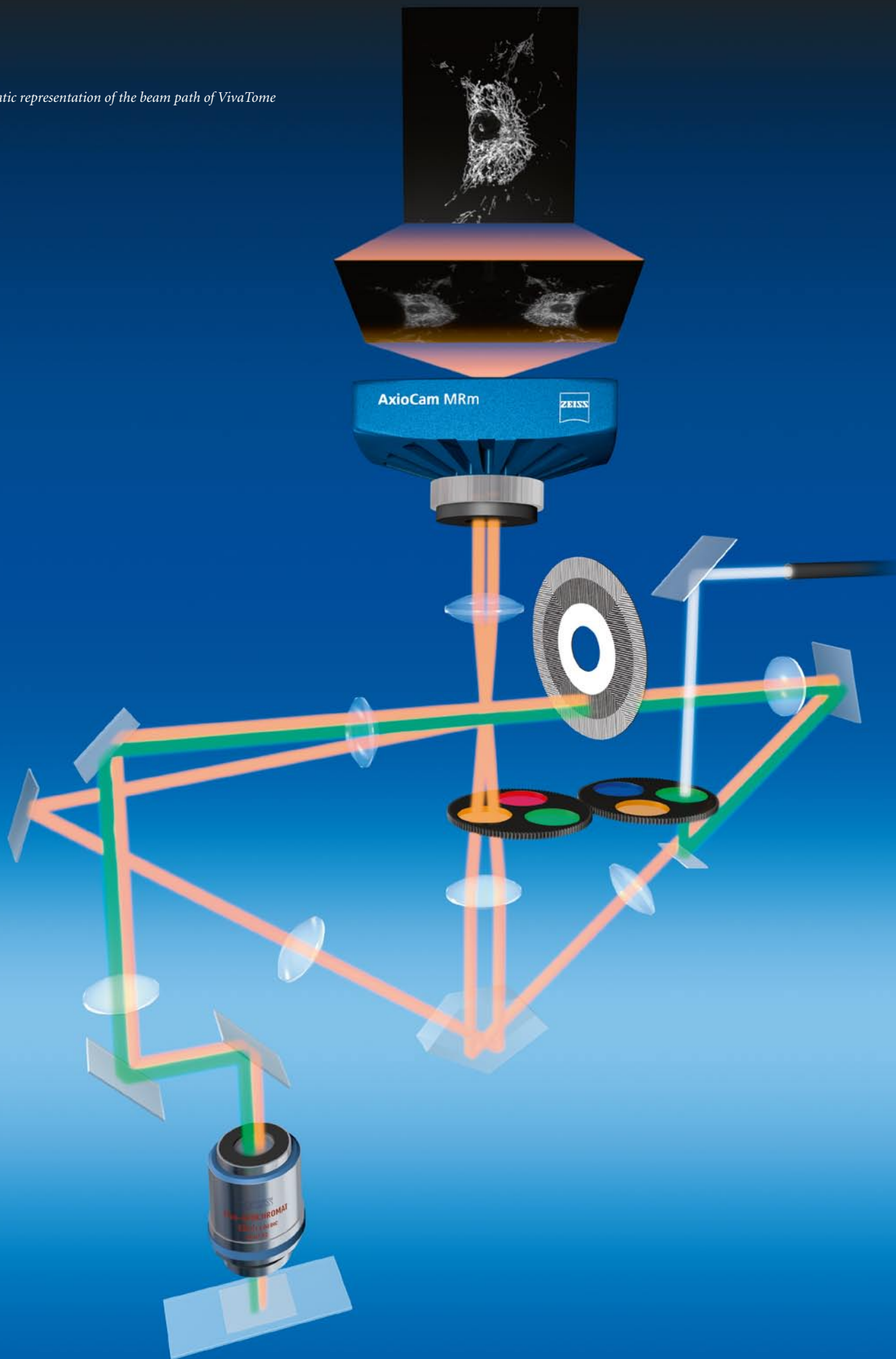
Scaled addition

Scaled subtraction



A scaled subtraction of both images completely removes the out-of-focus intensity and results in an optical section (right), while a scaled addition results in a normal widefield image (left).

Schematic representation of the beam path of VivaTome



What You Need Is What It Gives. VivaTome at Work

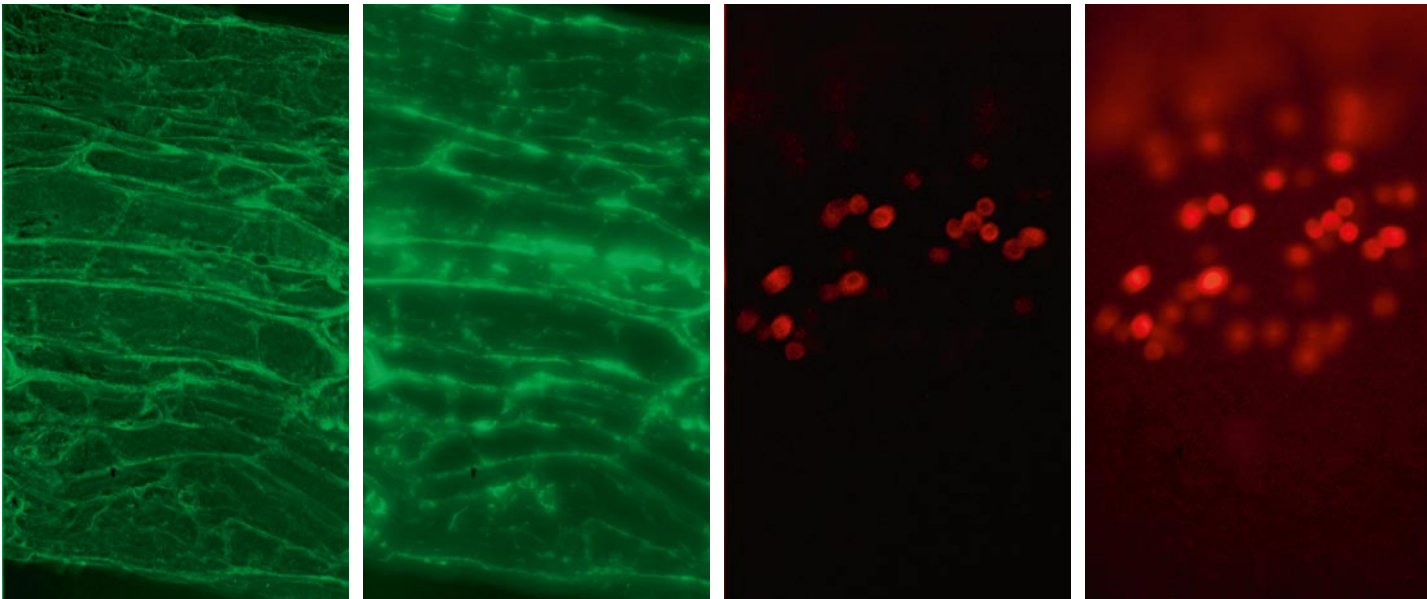
Capturing high-quality optical sections of rapid processes is mandatory for a large number of applications. VivaTome's flexibility means it can do this and much more: Since it is compact and compatible with all types of Carl Zeiss microscopes, it is integrated quickly and easily into existing imaging systems.

Developmental biology

The development of an organism is a highly dynamic process whose imaging requires high temporal resolution while maintaining excellent out-of-focus light rejection. Besides, since matching the grid to the objective's magnification is so easy with VivaTome, a range of model organisms and developmental stages can be observed on the same system with minimal hardware changes. Regardless of whether the task is to examine cell division and migration during early development or studying morphogenetic processes at later stages – VivaTome will always deliver outstanding images.

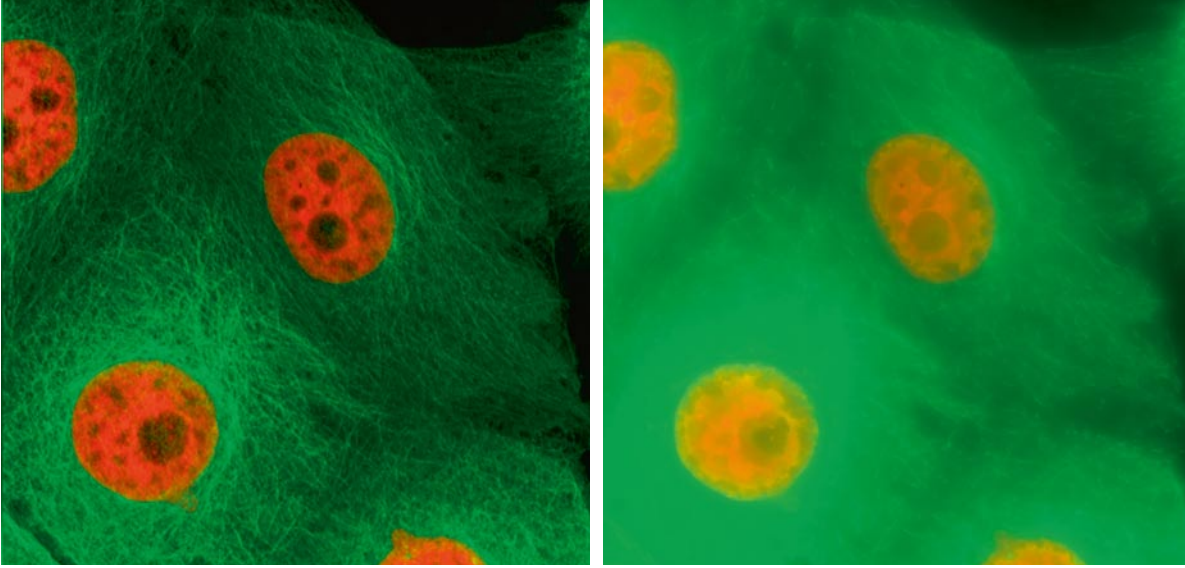
Cell biology

Cellular processes can be highly dynamic and the imaging system must be able to keep up with these processes. Applications such as observing the cytoskeleton, membrane transport or organelle dynamics often require three-dimensional image series as well as maximum resolution for reliable analysis. These applications also need optimal environmental conditions for the living sample: Minimal phototoxicity and perfect incubation ensure successful experimental results. VivaTome provides both: The high light efficiency ensures gentle illumination and the Carl Zeiss incubation functions are fully integrated in AxioVision for combination with the VivaTome system. Optimal culturing conditions + maximum resolution = outstanding results.



EGFP-labeled Arabidopsis thaliana, 10x/0.45 Plan-APOCHROMAT, maximum intensity projection of a 3D stack, left sectioned, right widefield. Scott Olenych, David H. Murdock Research Institute Kannapolis, USA

Fluorescent erythrocytes (RED) in a two-day-old zebrafish embryo TG (gata1: dsRed), 20x/0.8 Plan APOCHROMAT, left sectioned, right widefield. TinChung Leung, North Carolina Central University, USA

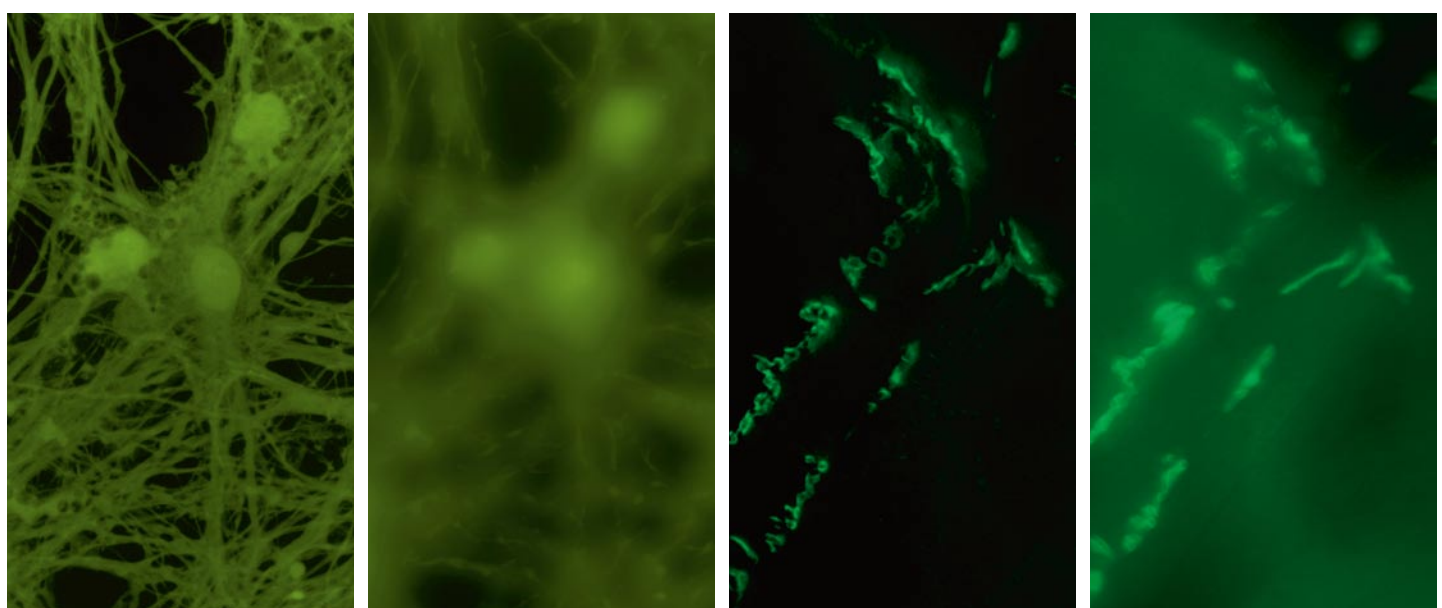


Tubulin-Emerald, H2B-Cherry, 63x/1.4 Plan-APOCHROMAT, maximum intensity projection of a 3D image stack, left sectioned, right widefield.

Physiology

Today, fluorescence can be used to directly monitor a variety of intracellular processes in real time. Ion concentrations, membrane potential or membrane organization can be probed with suitable reporter dyes and changes can be recorded at a high temporal resolution. VivaTome allows fast acquisition of optical sections, allowing unambiguous interpretation of such time lapse sequences from cells or tissues. The fixed-stage microscope Axio Examiner

from Carl Zeiss is the ideal platform for electrophysiological and physiological applications. When combined with VivaTome it is the ideal entry-level solution. With this imaging system, signal transduction processes, for instance, can be recorded and three-dimensional reconstructions of the surrounding tissue can be captured in precise detail – reliable, simple to operate and with optimal image quality.



*Cultured neuronal cells labeled with BCECF, 63x/1.0 W Plan-APOCHROMAT, maximum intensity projection of a 3D stack, left sectioned, right widefield.
Eva Ruusuvaori, University of Helsinki, Finland*

*Drosophila 3rd instar larvae with CD8-GFP-Sh labeled neuromuscular junctions, 40x/1.3 Plan-NEOFLUAR, maximum intensity projection of a 3D stack, left sectioned, right widefield.
Anne-Marie Lüchtenborg, University of Konstanz, Germany*

Optimal Performance from Optimal Components

Different tasks in life science require different microscope systems. Carl Zeiss offers a wide range of high-performance components, which, when combined with VivaTome, can meet even the toughest demands.

Top shelf microscopy:

Axio Observer, Axio Imager and Axio Examiner

Optimal fluorescence technology and unsurpassed optics – these performance features make the microscopes from Carl Zeiss the gold standard in scientific research. The upright Axio Imager microscope is perfectly suited for applications in developmental biology. The inverted microscope Axio Observer is the right platform when monitoring living cell cultures. And the fixed-stage Axio Examiner is ideal for electrophysiological studies. VivaTome is compatible with each of these solutions: It fits on every upright, inverted and fixed-stage microscope from Carl Zeiss.

- Fully motorized microscopes for high-end research
- Corrected apochromatic fluorescence beam path for brilliant image quality in multichannel image acquisition
- Optimized ergonomics for comfortable operation
- Rapid vibration-free motorization
- Unsurpassed stability and extendibility

Designed for optimal performance: objectives from Carl Zeiss

Complex applications pose tough demands on the optical performance of any imaging system. The objectives from Carl Zeiss make even the weakest signals clearly visible. Combined with VivaTome high numerical aperture objectives and water immersion objectives provide optimal results.

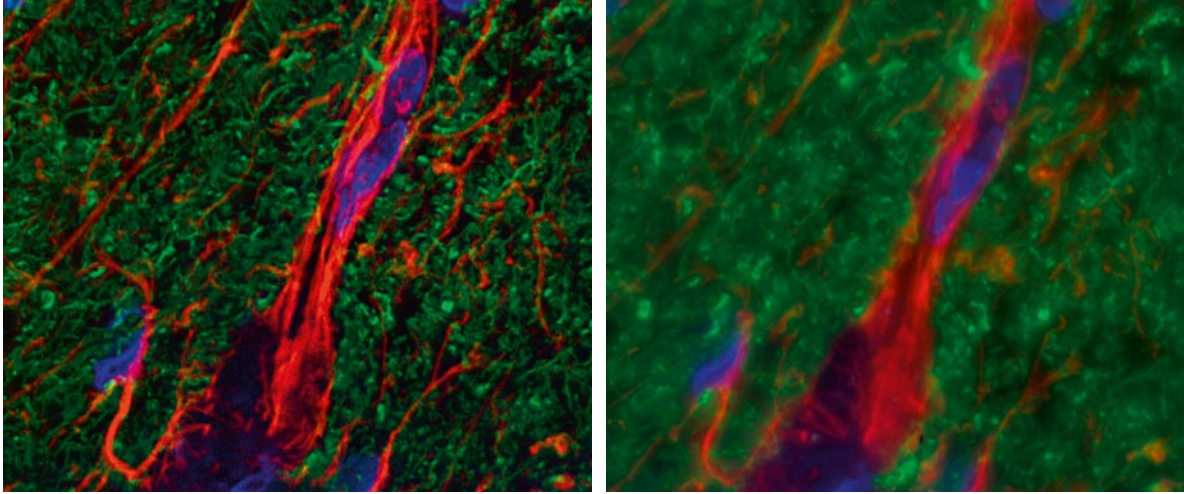
• EC Plan-NEOFLUAR

EC Plan-NEOFLUAR objectives are designed for applications where flexibility over a broad range of methods is required. The optimized IC²S optics allow contrast-rich imaging with perfect homogeneity and high resolution.

• Plan-APOCHROMAT

With their optimal color correction and very high numerical apertures, Plan-APOCHROMAT objectives deliver brilliant images of unparalleled resolution and clarity.





Triple stained mouse brain sections, 63x/1.4 Plan-APOCHROMAT, maximum intensity projection of a 3D image stack, left sectioned, right widefield.

- **LCI Plan-NEOFLUAR**

The multi-immersion LCI Plan-NEOFLUAR objectives are used for live cell imaging (LCI). A correction collar allows them to be adapted to different sample conditions.

- **LD LCI Plan-APOCHROMAT**

This objective meets the very highest requirements in live cell imaging. It offers an extremely long working distance, enabling researchers to focus through thick specimens.

- **C-APOCHROMAT**

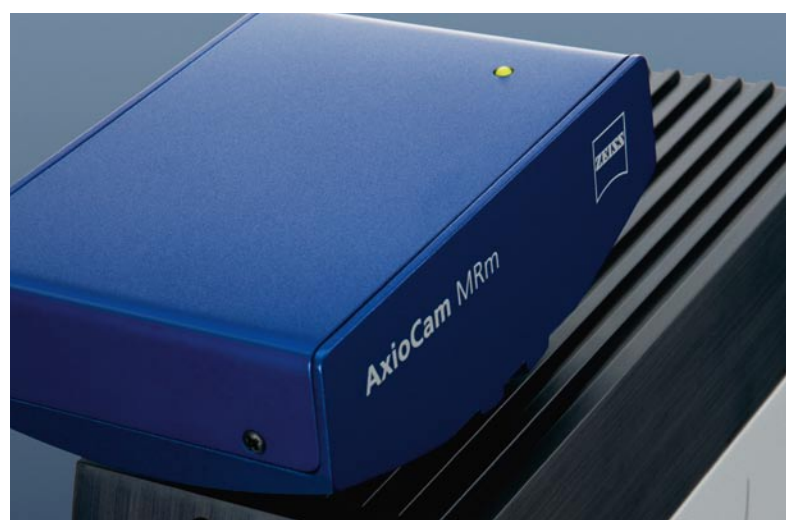
These high-performance objectives are able to compensate optically for different cover slip thicknesses and different temperatures. They are perfectly suited to extremely demanding applications in research of living organisms.

Sensitive and reliable: the AxioCam family

AxioCam is the latest generation of Peltier-cooled digital cameras from Carl Zeiss. These cameras offer maximum sensitivity and a range of resolutions: 1.4 megapixels with AxioCam MRm up to 13 megapixels with AxioCam HRm. A dynamic range of up to 1:2200 or 1:2500 ensures optimal image quality, especially when combined with structured illumination techniques.

Gentle yet durable: the light sources

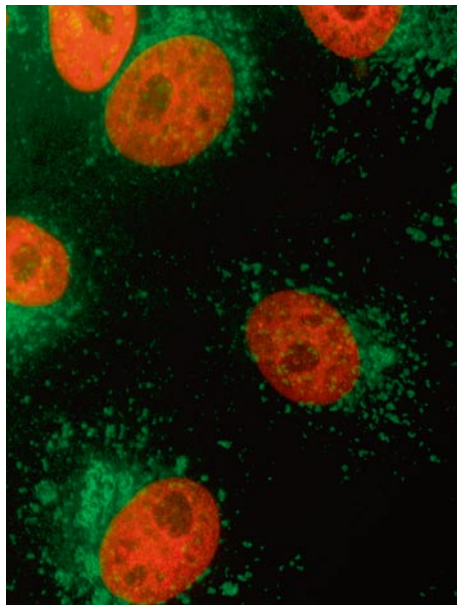
VivaTome works with a fiber-coupled white light source. The new HXP 120 C from Carl Zeiss is an efficient, homogeneous, long-lasting metal halide light source. It requires no adjustment and is completely integrated into the software.



AxioVision Microscope Software.

Digital Intelligence That Grows with Your Tasks.

AxioVision is the system software from Carl Zeiss. In addition to controlling and synchronizing all microscope components, AxioVision offers an unsurpassed variety of modules. From simple image acquisition to complex analytical procedures, AxioVision has an impressive range of functions that you can adapt to your individual demands any time they arise – quickly and economically.



Multichannel Fluorescence

Freely combine different fluorescence channels with transmitted-light images. One image per channel, with optimal exposure time, can be acquired for each excitation wavelength.

Z-Stack

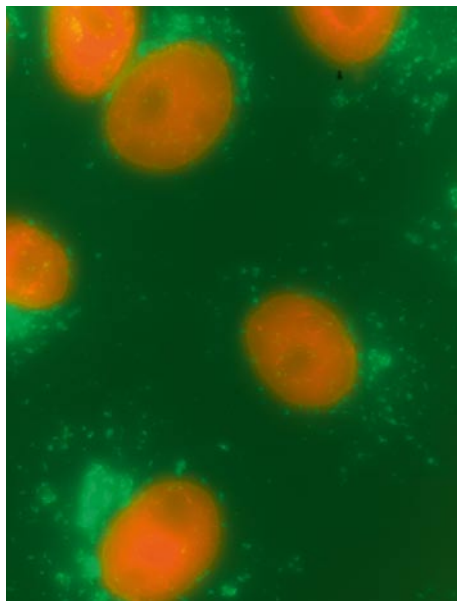
With a software-driven z-drive you can acquire and display different focal planes from a sample as an image stack that can be reproduced accurately and with great precision.

Time Lapse

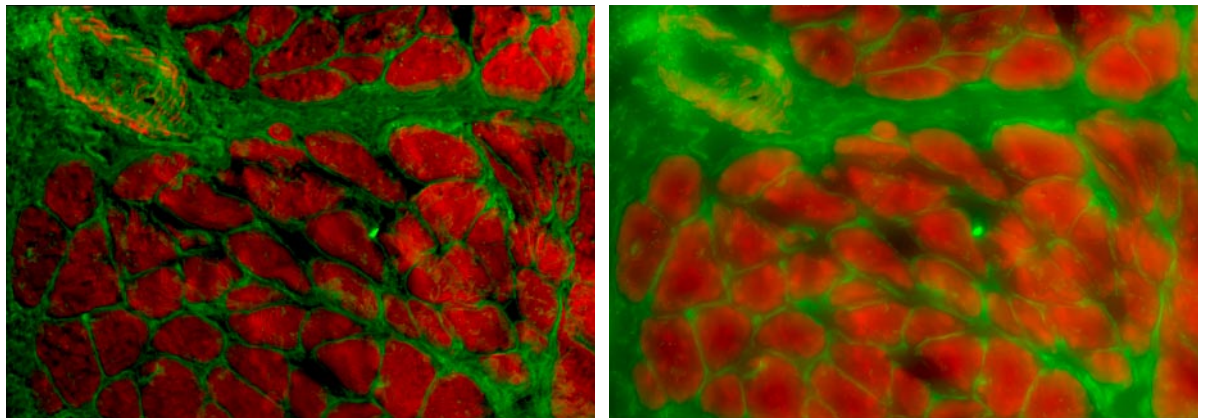
A time lapse is a series of images captured over time. This allows you to document physiological processes and the dynamics of living cells with a series of images. The software automatically controls all the important microscope components during acquisition.

MosaiX

With this module and a motorized x, y-stage you can image large sample preparations at high resolution. Large areas of your preparation are scanned, recorded as tiles and stitched together.



Lyso-Emerald, H2B-Cherry, 63x/1.4 Plan-APOCHROMAT, maximum intensity projection of a 3D image stack, upper image sectioned, lower image widefield.



Rat esophagus, WGA-Alexa 488, Phalloidin-Alexa 568, 63x/1.4 Plan-APOCHROMAT, maximum intensity projection of a 3D stack, left sectioned, right widefield.

Mark&Find

The Mark&Find module is used to mark, store and automatically locate different positions of interest in your slide, culture or well plate. You can then image these areas of interest repeatedly and greatly increase the throughput and efficiency in prolonged experiments.

Multidimensional image acquisition

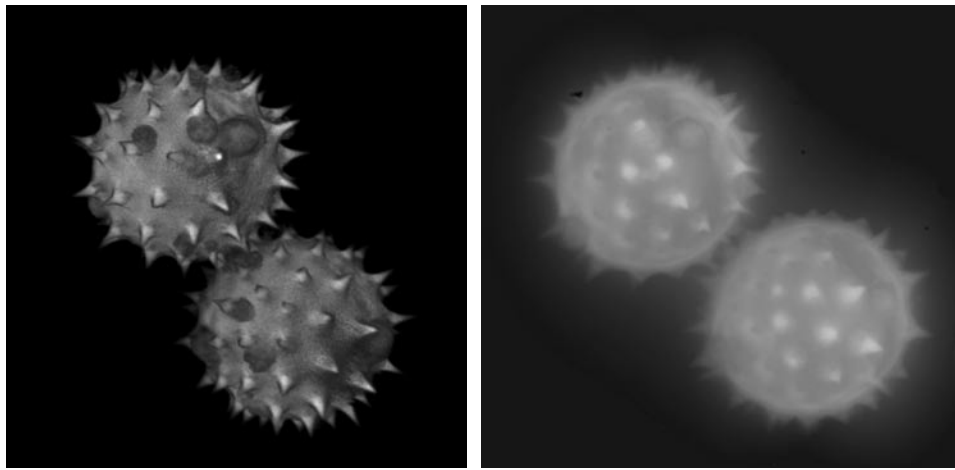
For multidimensional image acquisition with VivaTome, the Mark&Find, Multichannel Fluorescence, Z-Stack, Time Lapse and MosaiX modules can be used in any combination to adapt the system accurately and economically to the task at hand.

Fast Acquisition

With Fast Acquisition, you can greatly increase the speed of your Z-stack, multichannel, and time lapse functions. The images are stored directly on the hard disk using a data streaming process. Components such as the camera, light source or piezo focusing unit can be utilized at maximum speed. Fast Acquisition ensures that the interaction of the various components is timed precisely.

Fast MosaiX							
Dual Camera	HDR Imaging	Widefield Multichannel Unmixing					
Digital High Speed Recorder	Fast Acquisition	Imaging Plus	ASSAYbuilder	ELISPOT			
Panorama	Mark&Find	ApoTome	3D Measurement	TMA	SFM		
MosaiX	Time Lapse	3D Deconvolution	QuantiFISH	Ratio	Tracking		
Extended Focus	Z-Stack	2D Deconvolution	AutoMeasure	AutoMeasure Plus	Colocalization		VBA
Autofocus	Multichannel-Fluorescence	Inside4D	Interactive Measurement	Online Measurement	Physiology	Asset Archive	Commander
Image Acquisition		Image Processing	Image Analysis			Documentation	Configuration

Modules relevant for VivaTome are shown in orange.



3D rendering of pollen grains, 100x/1.4 Plan-APOCHROMAT, left rendering of sectioned image stack, right rendering of widefield image stacks.

Extended Focus

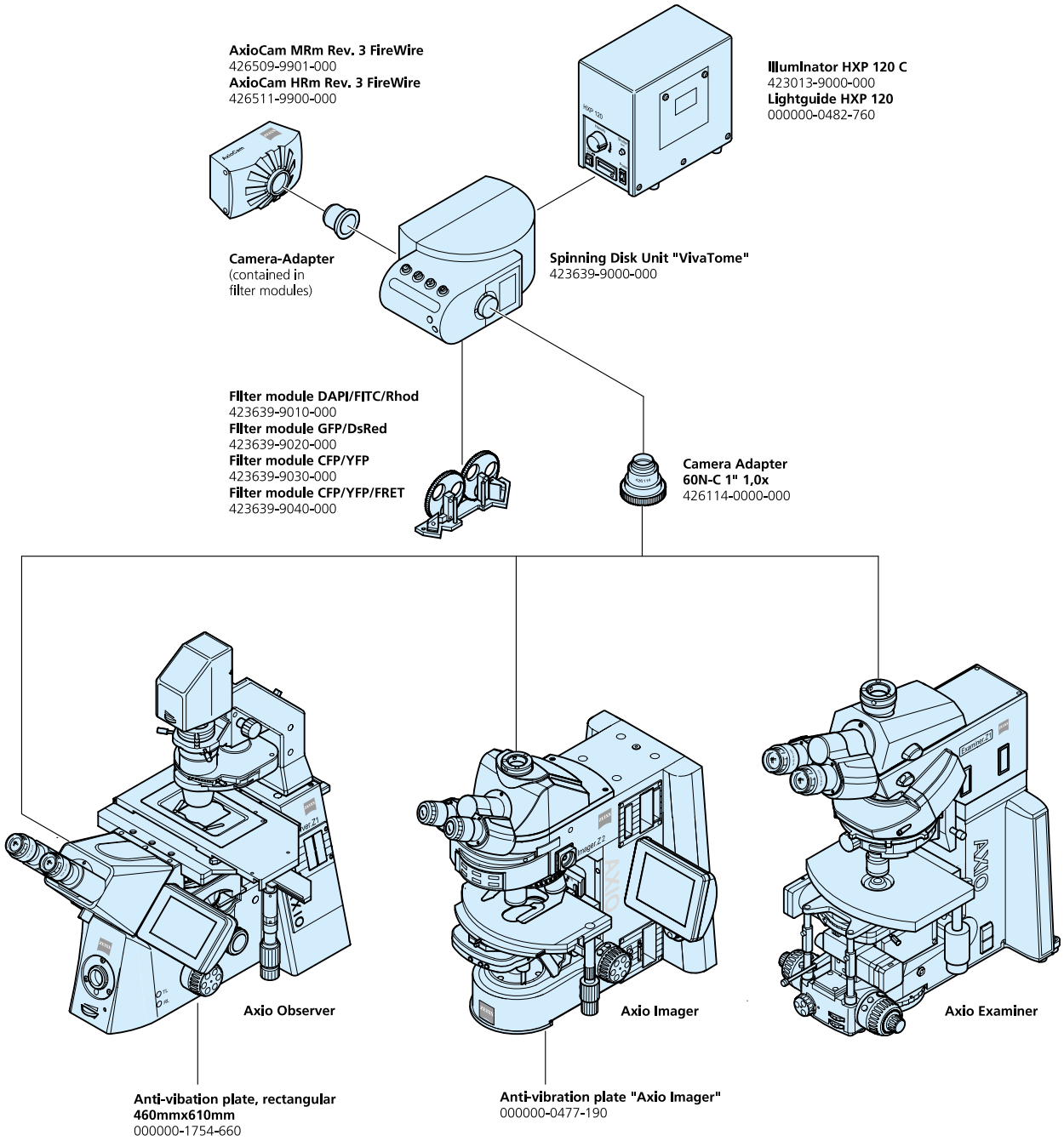
The Extended Focus module allows three-dimensional image stacks to be displayed in a two-dimensional plane. The sharpest details are extracted and combined from each plane. The resulting image has a significantly greater depth of field when compared to the direct image capture of a single plane.

Inside4D

The Inside4D rendering module aids in the visualization and presentation of three-dimensional data sets. After capturing the optical sections, you can immediately create a 3D rendering of the data set. This is an outstanding tool for animating your three-dimensional images to make them more effective in Internet or PowerPoint™ presentations.

Measuring and analyzing

With the image analysis modules of AxioVision, you can apply a variety of methods to your samples. There are modules for measuring intensity or area as well as colocalization. You can measure volumes, surfaces or distances in three-dimensional image stacks easily and with a wide range of functionality.





Carl Zeiss MicroImaging GmbH

07740 Jena, Germany

BioSciences | Göttingen Location

Phone : +49 5515060 660

Telefax: +49 551 5060 464

E-Mail : micro@zeiss.de

www.zeiss.de/vivatome