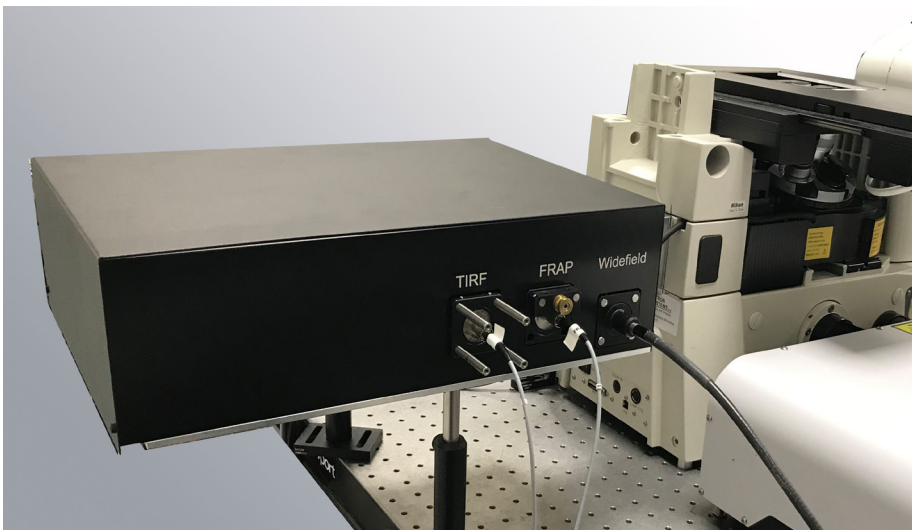


## ORBITAL-500 Superior Flexibility Ring-TIRF, 2D FRAP and Direct Laser Coupling

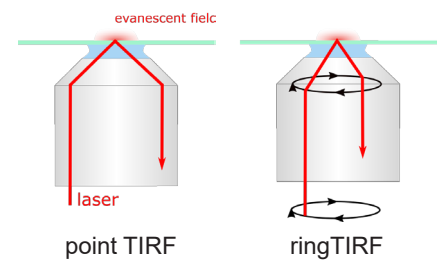
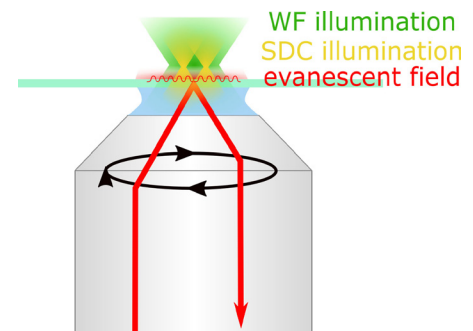
The ORBITAL-500 system is even more flexible. It can combine in one chassis Ring-TIRF, 2D FRAP and one internal laser, which directly beams into the Ring-TIRF scanner. The Direct Laser Coupling (DLC) preserves the maximal laser power for imaging applications like STORM, where high power is needed for switching off e.g. CY5/Alexa Fluor 647. In addition the motorized beam expander option with 2x, 4x and 6x facilitates this process by enhancing the laser power density in the sample.

## ORBITAL-500

Ring-TIRF  
 FRAP  
 DLC-Laser  
 Illumination

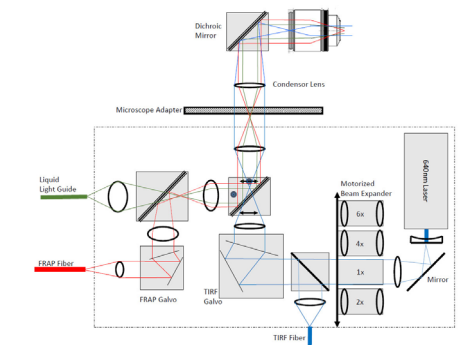


ORBITAL-500 on Nikon Eclipse Ti microscope

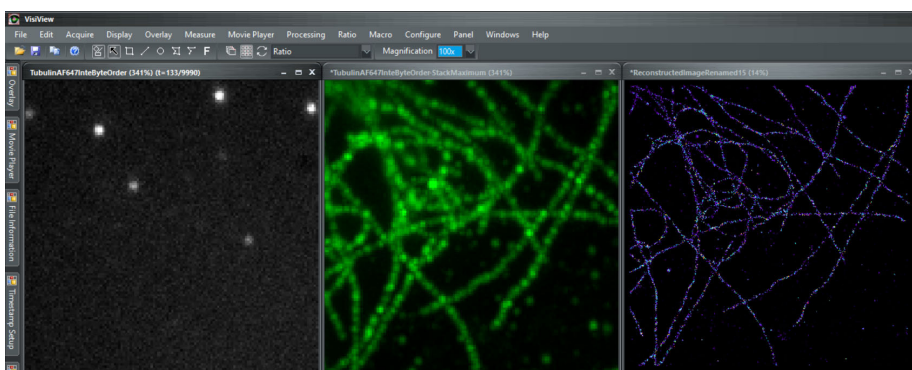


## SMLM - Single Molecule Localisation Microscopy

The single molecule localisation microscopy is a Super Resolution technique that enables a lateral resolution of up to 20 nm. Through stochastic activation of single fluorophores and the precise localization of its position, a super resolved image can be reconstructed. The sparse activation of single molecules is actively controlled by VisiView through regulation of the activation laser power. The recorded signals are analysed in real-time to identify single molecules and to display a reconstructed super resolution image.



ORBITAL-500 Overview



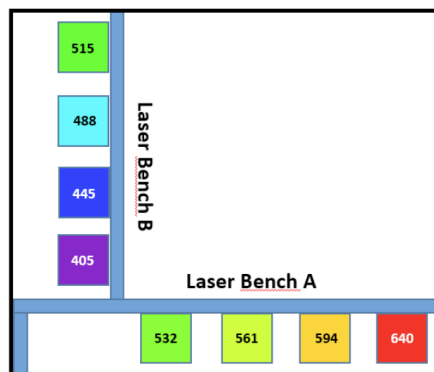
VisiView STORM option e.g. with QuickPALM reconstruction of AF647 labelled tubulin.  
 Fiji realtime import VisiView stack data

# ORBITAL Technology

## VS-LMS Laser Merge System

### VS-LMS Flexible Multiple Laser Engine with highly stable Laser Outputs and Motorized Alignment Option

The new generation of Visitron Systems VS-LMS Laser Merge Systems is now available in a very compact design. The unique optics couples up to eight diodes or solid state lasers to a single collinear lasers beam. This beam can be channeled into three different outputs for simultaneous laser applications like Confocal / FRAP or TIRF.



Wavelength* (nm)	Maximum Power Rating* (mW)
405	300
445	100
488	200
515	100
532	150
561	200
594	150
640	200

### Features and Benefits:

- » multi-line laser source including up to 8 lasers
- » flexible selection of diode and solid state laser modules
- » free illumination selection of any laser combination or pattern
- » FC-coupling design with focus correction
- » thermally managed system
- » port Switcher with up to 3 output ports e.g. VisiScope Confocal, VisiFRAP and VisiTIRF

### VS-AOM Acousto-Optical Modulator - High Speed Optical Shutter

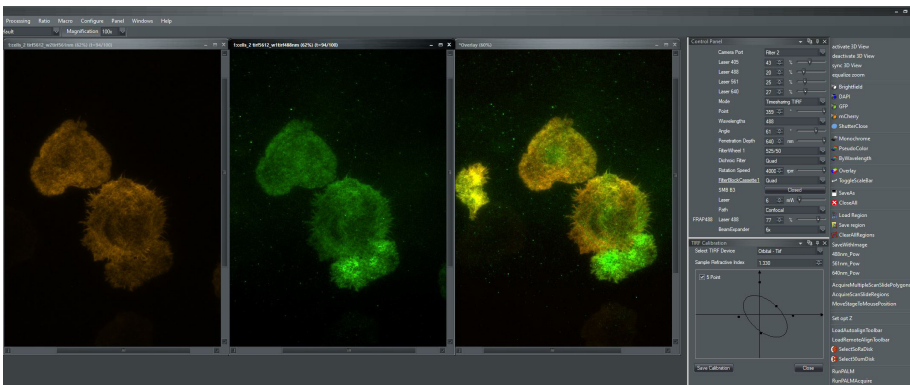
An acousto-optic-modulator (AOM) is a device which can be used for controlling the power, frequency or spatial direction of a laser beam with an electrical drive signal. It is based on the acousto-optic effect, i.e. the modification of the refractive index by the oscillating mechanical pressure of a sound wave. The AOM is used in the VS-LMS for high speed switching and intensity control of solid state lasers.

## VisiView ORBITAL Support and Features

VisiView® is a high performance imaging software for Bio-Medical applications. The software is designed as an integrated imaging software which includes comprehensive microscope control, control of a vast number of peripheral devices, image acquisition and analysis. Its multitasking ability supports realtime image handling and up to 6D multidimensional acquisition. The ORBITAL setup, calibration and control is seamlessly integrated into the VisiView capture mode. In the TIRF illumination configuration, the user can setup five different TIRF modes. Which are Center Beam, Widefield/HILO, RingTIRF, Timesharing TIRF and Point TIRF.

# ORBITAL Technology

# VisiView® Software Support



## ORBITAL Setup Modes

The Center Beam Mode moves the laser to the center of the calibrated ellipse. This is a good control for your TIRF alignment and proper beam focusing. It can be used for basic widefield laser illumination.

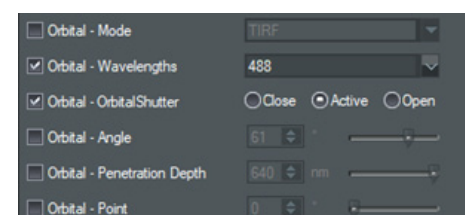
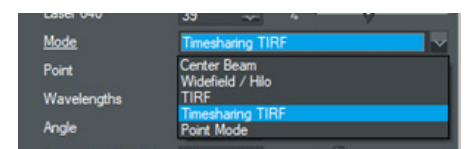
In Widefield/HILO mode the laser spins on a calibrated ellipse but at low incident angle. This offers a more homogenous laser WF illumination than the center beam Mode, which is strongly affected by interference.

The Point Mode simulates a classical TIRF system but the angle and the direction of the laser can be freely adjusted using the angle slider and the point slider component of the ORBITAL device.

In TIRF mode the laser spins on a calibrated ellipse beyond the critical angle which is needed for total reflection. The TIRF mode removes the out-of focus blur and increases signal to noise drastically. In TIRF mode you can directly and precisely control the penetration depth of the evanescent field with a separate slider.

## ORBITAL Synchronisation

If using an ORBITAL, not only the elliptical laser deflection is externally controlled by the ORBITAL controller, but also the laser shuttering, which allows precise synchronisation of laser and angle. Further the stable image quality is assured by synchronizing the laser rotation speed with camera exposure. If exposure times are too low, the system switches to arc scanning instead of scanning full ellipses.



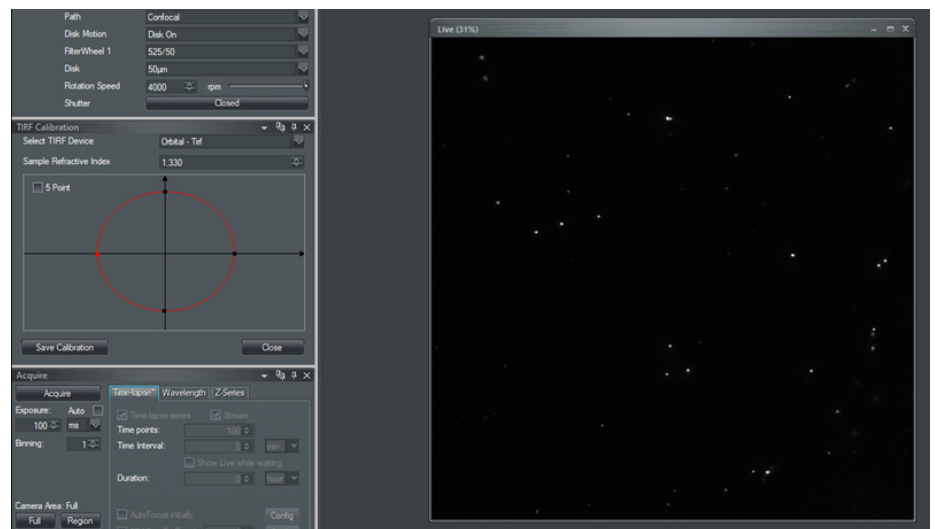
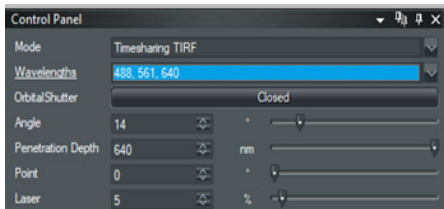
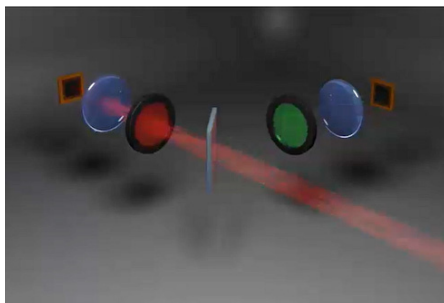
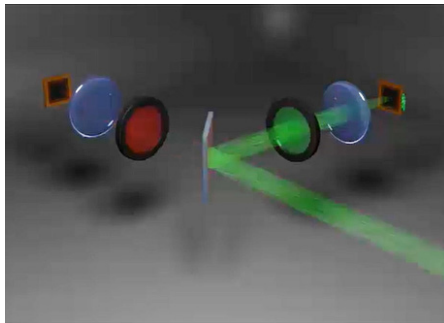
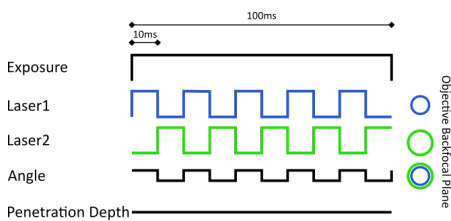
# ORBITAL Technology

## VisiView® Software Support

### VisiView ORBITAL Support and Features

ORBITAL calibration:

By default the calibration is specific for the magnification and illumination you have chosen and will be switched automatically. For the internal calculations, it is required that a magnification with a suitable objective is selected. Make sure you have a TIRF objective and that the magnification is configured with the appropriate refractive index and numerical aperture.



VisiView ORBITAL TIRF angle calibration by 4 or 5 points with 200nm TetraSpec beads

### ORBITAL Timesharing Mode

Usually simultaneous multi-wavelength TIRF imaging suffers from a variation of the penetration depth which is like optical refraction wavelength dependent. This means in order to create equal penetration depths distinct angles are required. But this is not possible if the lasers are switched on at the same time.

The Time-Sharing Mode circumvents this limitation and makes simultaneous multi-wavelength TIRF imaging at equal penetration depth possible.

If set to Time-Sharing Mode the ORBITAL will switch and change the active laser and the incident angle after each elliptical sweep. The time for each elliptical sweep is basically calculated by:

$$A \div \text{floor } A/10\text{ms where as } A = (\text{Exposure time}) / (\text{number of Wavelengths})$$

Thus the ellipses for all wavelengths are done exactly with same exposure time and at complete trajectories but different incident angles. The required angles will be calculated from the selected wavelengths automatically.