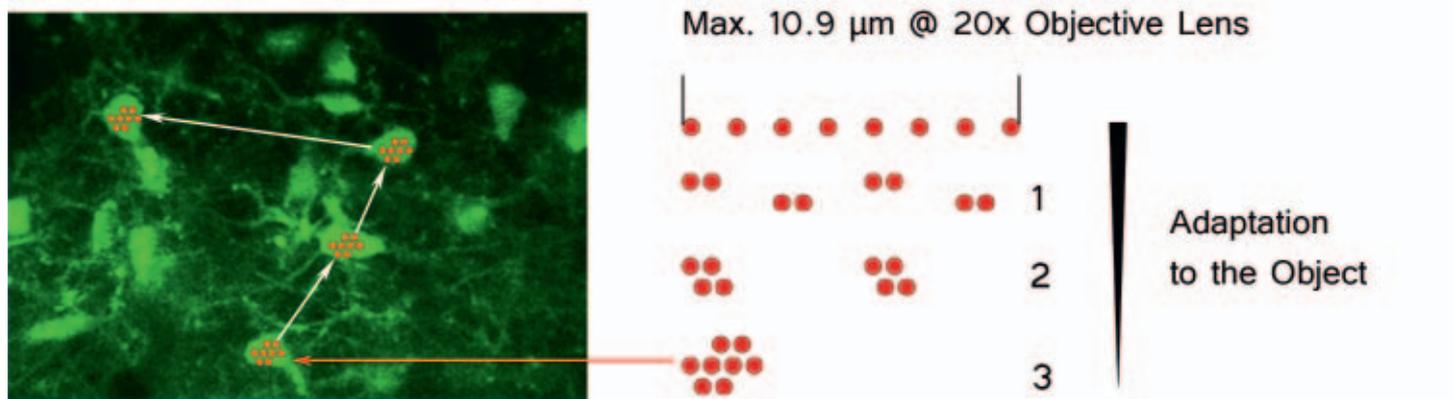


Cloud Scanner

>> *Flexible multi point scanning for brighter images and selective excitation below single cell size* <<

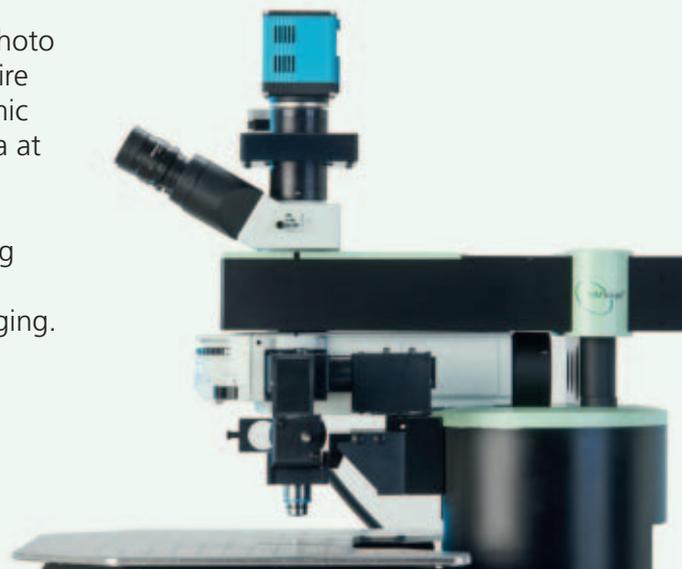


- Higher frame rates
- Brighter images
- Less photo damage
- Fast selective treatment of cell compartments

The Cloud Scanner creates up to 8 multi-focal laser beamlets that have temporal and spatial separation. All laser beamlets can be independently arranged by the user to point 1) a single focus, 2) a line of foci, or 3) even to cover a whole cell body. The beam arrangement is software controlled and can be optimized during the measurement.

Microscopy of dynamic events always depends on signal to noise ratio and therefore on the fluorescence intensity. In turn the fluorescence intensity depends on the fluorophore, the excitation power and the excited area. If it comes to in vivo Ca^{2+} imaging of somata for example the maximum excitation power is limited because of photo bleaching and toxicity. Exciting the cell body by scanning its entire area increases the fluorescence yield but drops down the dynamic capabilities. The Cloud Scanner excites this necessary larger area at once without limiting the temporal resolution.

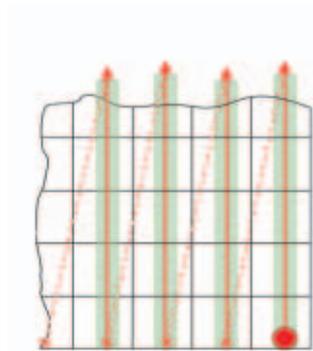
Cloud scanning can be applied to all scanning patterns including raster scan, line scan and point measurements. Increasing the fluorescence signal leads to better results and allows faster imaging.



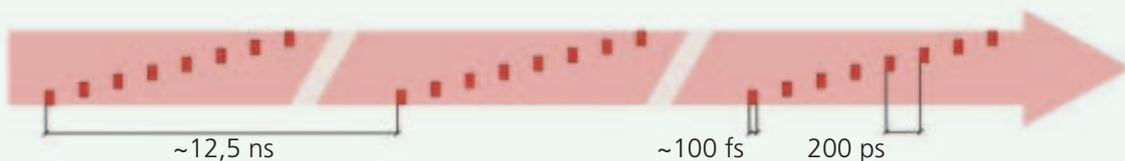
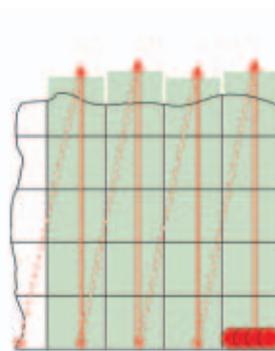
Adaption to the Pixel Size

For most applications intravital microscopy has to provide large field of views in combination with high – but not diffraction limited – resolution. Imaging speed is more important than ultimate resolution. Therefore, low magnification objective lenses and a limited pixel resolution have to be chosen. High NA-objective lenses deliver best 2-photon performance due to their excitation and collection efficiency. One of the most common imaging modalities combines a 20x, NA 1.0 objective lens, 500 by 500 microns FOV and 512 by 512 or even less pixel resolution. The focus size is actually smaller than the pixel size. In other words, only part of the pixel volume will be excited. The Cloud Scanner accounts for this situation allowing the user to define a line of eight foci shorter than the pixel width.

Single beam scanning pathway - a fraction of the pixel area is excited



Cloud scanning pathway - the whole pixel area is excited



Time delay between laser pulses while using a 80 MHz Ti:Sa laser.

200 ps delays between two foci eliminate cross talk and allow to increase the laser power without damaging the sample.

Scanning and data acquisition method remain identical to a standard single beam laser scanning microscope – but it gains brightness. Since speed depends on brightness, the maximum frame rate can be increased substantially.

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