Laser Scanning Confocal Microscope

The LSCM is primarily used for imaging fluorescent samples. Organelles and proteins in cells or tissues are imaged using native fluorescence or by adding fluorescent tags. The LSCM is based on point scanning approach to image formation whereby the information for each pixel is obtained sequentially instead of the broad area excitation approach used for wide-field imaging. A laser is used for selective excitation of a fluorophore and an optical bandpass filter limits the light reaching the detector. The LSCM is useful for three-dimensional reconstruction of data. Spatial resolution ~ 300 nm is achievable with objectives of numerical aperture of 1.3.

Specifications

Excitation wavelength	multi-line argon ion laser (457, 488, 514 nm) three HeNe lasers (543, 594, 633 nm)
Filters	Bandpass Longpass
Objective lens	10x/0.3NA 20x/0.75NA 40x/1.3NA 63x/0.9NA (dipping objective) 100x/1.3NA
Simultaneous excitation	2 channels 488 nm and 594 nm (recommended)
Acquisition Differential	Time sequence
interference contrast	Using 10x and 100x objective lenses
Advances capabilities	Objective inverter (LSM Technologies) for upright microscopy applications Widefield imaging, CCD camera as detector (broadband excitation and emission) ~405nm excitation (achieved using the ultra-fast laser system and SHG) FCS2 Temperature control chamber (contact Dr. Jim Xiang for use of the FCS2) Two- Photon Excitation Microscopy Fluorescence Lifetime Imaging Microscopy

Settings

User may excite fluorescence of elements within the sample, or add fluorescent tags. Excitation energy is selected by use of one, or two lasers simultaneously. Sequential colour-imaging, and z-sectioning procedures may be used to acquire images. Widefield images may be collected for comparison. This system uses the Verdi / Mira / Pulse Picker / Harmonic Generator suite.