Confocal Laser Scanning Microscope (CLSM)

Model Name: Leica TCS-SP8

Specifications

- AOTF controlled Five laser lines 488, 514, & 633nm
- Five fully adjustable PMT detectors (i.e. can be set to capture any given range of emission from 400 to 800 nm),
- Transmitted light detector with DIC option
- Two filter cube based avalanche photo diode (single photon sensitivity) detectors which can be used with green and far-red fluorophores for imaging or FCS
- 10x, 20x, 40x, & 100x (oil immersion) objective lenses
- Resonant scanner (8kHz) for very high speed image acquisition
- Uses Leica Specific Software (LASX)

Applications:

Confocal Laser Scanning Microscopy (**CLSM**) is an optical microscopic technique for obtaining the 2-D and 3-D high-resolution optical images, projections as well as 3-D reconstructions of auto fluorescent objects excited with aid of laser-induced light operating at variable visible wavelength. The key feature of confocal microscopy is its ability to acquire in-focus images from selected depths, a process known as optical sectioning. Images are acquired point-by-point and reconstructed with a computer, allowing three-dimensional reconstructions of topologically complex objects. CLSM can be utilised to study microfossil, organic walled spores, pollen, fungi and fossil protists as well as helpful in determining the structure and composition of the fossilised cell wall of higher plants.

Capability:

- High resolution blur-free confocal imaging in fluorescence and reflected light modes;
- Individual continuous cellular tomograms without slicing cells and stereographs which are observed by three dimensional reconstructions;
- High depth of focus extended focus images approaching SEM quality without special object preparation;
- Easy visualization of 3D structures by stereo pairs, red/green anaglyphs, 3D reconstructions, animation;
- Concurrent analysis of surface structures and internal structures (e.g. cavitation, wall structures);
- Multi-channel fluorescence imaging, including blue fluorescence (excitation 405 nm) through far red fluorescence (excitation 633 nm);
- Co-localization analysis;
- Live cell imaging and quantitative analysis over time.
- Spectral imaging;
- Topography analysis, surface roughness data, and Z height profiling;
- Stereology acquisition and analysis;
- Transmitted imaging (bright-field, phase contrast, polarized light, DIC).

Type of Material:

Both, microfossils bearing petrographic thin sections (thickness up to 150 – 200 Micron) and palynological slides and live biological specimens prepared through standard protocols can be studied on the microscope.

Dos or don't for CLSM Laboratory:

- Only authorised/registered users are allowed to access the CLSM laboratory;
- Non authorized users are not allowed to remove objectives of the microscope or attempt any maintenance;
- Users are not permitted to setup, alter and save new configurations of lasers and filters. The pre-programmed configurations are suitable to cover most of the applications in the Institute.
- There is only one on/off switch for the entire system. Do not touch any other on/off switch on the microscope, computer or power supplies.
- When completed your studies and imaging work please switch off the lasers and wait until they are properly shut down before exiting the LMS software;
- Copy your images to CD, and DVD or network drive before you leave. There is no scope of long time storage of images left on the microscope operating system and may be deleted at any time without warning.
- Please bring England Finder Co-ordinates of your specimens to save time.
- Data generated will be provided on CD (Compact Disc) or DVD (Digital Versatile Disc).
- Petrographic thin section slides are to be mounted with XXXX epoxy.
- Keep thickness of the mounted rock slice between 100-200 μm.
- Analysis of palynological material shall be conducted on Permanent slides. Analysis of samples is restricted to generation of spectra in case 3D imaging in case of CLSM.
- Interpretation of spectra and imaging is available in certain cases and it will be chargeable extra.