

## Specifications for Upright Confocal

The system should have confocal technology for fixed & Live Cell sample imaging including FRAP, FRET and Photo activation/conversion.

The system should consist of:

1. Fully Motorized Upright Fluorescence Research Microscope for BF/DIC/Fluorescence preferably with touch screen display for controlling the motorized components of the microscope.
2. Programmable and motorized X-Y stage, Universal sample holders for slides, cover slips, 35/60 mm Petri dishes and 6/24/96 well plates.
3. High precision motorized Z-focus drive with step size of 20 nm or less and with PieZo focusing.
4. 12V/100W halogen illumination for transmitted light & 120W metal halide illumination for Fluorescence should be offered.
5. Motorized 6-position DIC nosepiece, Universal Motorized Condenser with modules for DIC, 8-10 (or better) position fluorescence turret for accommodating fluorescent filters for sample visualization and camera based imaging.
6. High-resolution confocal grade objectives of 10x/0.40, 20X/0.70, 40x/1.30 oil, 40x and 60/63X/1.0 long distance water dipping objective with at least 2 mm working distance, 60/63X/1.40 oil immersion, 60/63X silicon objective or an equivalent objective for high-resolution live cell imaging and 100X oil immersion objective (or equivalent high-resolution confocal-grade objectives) should be offered.
7. Automated shift free DIC accessories for all objectives. Band pass fluorescent filters for DAPI, GFP, CFP, YFP, Cy3, Cy5 with automatic identification facility should be possible.
8. Monochrome cooled high-resolution CCD camera (or equivalent camera) with 1.4 million effective net pixel resolution or better controlled by confocal software with multichannel, Z-stack and time series imaging should be quoted
9. Suitable imported anti-vibration Table with compressed air suspension for complete system or better should be quoted.
10. High-sensitivity confocal laser point scanning with built-in high sensitivity spectral detectors. Detection unit should be capable of working in Intensity (conventional intensity) and Spectral mode Imaging. It should be capable of simultaneous detection and separation of minimum four fluorophores or better based on high sensitive detectors with QE of 45% or better.
11. Scanner unit should have laser ports for Vis, UV and IR lasers. The laser scanner should have the capability of fast scan for bleaching/photo-activation & normal scan for Imaging, to conduct experiments like FRAP, FLIP, photo activation, photo-conversion and photo-bleaching. Maximum scan resolution should be at least 6Kx6K or better per channel and should reduce to 16KX16K resolution or better. Scan speed should be 5fps or better @ 512x512 pixels resolution (Specific Modes Intensity and Spectral should be mentioned). The scan head should be able to perform fast dynamic live cell time-lapse imaging with a temporal resolution of 5msec or better.
12. The scan head should include PMT for fluorescence and laser based DIC imaging. The scan field should be 18 mm or better. Scan Zoom range 1X to 40X or better with increments of 1X or better.
13. Visible laser illumination system with AOTF control for the following laser lines:  
Multi Argon 457 or 458/488/514 nm laser with minimum of 35mW laser power or better to optimally excite CFP, super glow GFP, Alexa 488, GFP, FITC, YFP, etc. For simultaneous GFP and YFP imaging the system should be equipped with appropriate emission wavelength filter set. It should have band pass filter 505-530nm and long pass filter 530nm to collect GFP and YFP signals, respectively.  
DPSS 561nm Laser with minimum of 20 mW or more to optimally excite Ds Red, Nile red, MitoTracker Red, mCherry, Texas Red etc.

Red HeNe 633/635 Laser with minimum of 10 mW or better to optimally excite Cy5, Alexa 633, TO-PRO3 etc.

And Blue diode 405nm for DAPI excitation etc

The system should have optimum laser lines and filters for imaging of photoactivatable fluorescent GFP and green to red photoconversion of Dendra-2. (This requires a UV laser apart from 488 nm laser and special filters to excite chromophore and collect emission spectra between 553nm to 573nm)

14. The system should be supplied with micro point ablation laser with appropriate attenuator for targeted killing of a single cell in living tissue or a layer of cells without damaging the neighboring cells. Micro point ablation laser should have both software and pedal operated control.
15. The Computer Workstation should have the newest 64 bit control computer with Intel Xeon Core Processor or better, newest DDR, RAM 12 GB or better: 2 TB storage or better upgradable to 4 TB or better, DVD, SuperMulti SATA +R/RW or better. Gigabit Ethernet, Win 7 OS OR newer version, USB 2.0, Fire wire port, large 30" LCD monitor or better.
16. Confocal Software will be required for
  - a. Complete control of all motorized functions of the microscope, digital camera, scan head and Laser control (including AOTF, image acquisition and processing).
  - b. Saving of all instrument parameters along with the image for repeatable/reproducible imaging
  - c. Frame/line/lambda capturing, Z-Stack, Time series imaging capabilities
  - d. ROI bleach for FRAP experiments and allow for doing photo-activation and photo-conversion experiments and their analysis.
  - e. FRET Imaging & Calcium Imaging.
  - f. Co-localization analysis and volume rendering
  - g. Real time ratio-display & Real Time Spectral Unmixing. It should have the capability to separate bleed through and auto fluorescence.
  - h. 2 D and 3D image deconvolution.
  - i. Diverse measurements and statistical processing
  - j. Software should be capable of recording different Live cell imaging experiments.
17. Third party software Amira should be supplied along with the system. Amira has versatile use in image processing and reconstruction. The software should come with image segmentation, slice alignment and automatic and manual image registration for creating movies for time-lapse data. It should be capable of 3D projection and visualization and at the same time it should be able to extract volume of 3D reconstructed images. <http://www.vsg3d.com/amira/amira>
18. Every part should have a part number quoted (including objectives) that can be matched to the catalog. Please give the page number in the catalog.
19. The bidders should clearly specify the after sales/service/application support capabilities (including warranty and terms of Annual Maintenance contract). Provide all information as regards pre-installation requirements (i.e. room, environment) for system installation.
20. Online UPS system with at least 30 minutes back-up time to support the complete system including lasers and computer should be provided.
21. A list of at least 3 users in India (along with contact details and purchase order) who are using this confocal system should be provided. Also provide a detailed list of users of the system in India/abroad with contact details.