Name of Equipment: Confocal Microscope

Technical Specifications: Fully motorized inverted confocal laser scanning microscope with live cell imaging system:

The self-sufficient, optimally functioning confocal microscope system should be of latest model and modular technology suitable for biological applications. The confocal system should be capable of high sensitivity detection mechanism meeting various applications of biological samples including multichannel fluorescence imaging with Z-stack, Live Cell imaging, time lapse applications including colocalization, FRAP, FRET, FLIP, FCS, Auto collation, Photo activation/conversion with modular and upgradable option for two laser photon, FLIM and TIRF application in future. The system should be offered with the following configuration:

Microscope:

1. Fully motorized inverted microscope with left / right-side port and capable of Bright field, Fluorescence and DIC imaging with touch screen TFT/Tab display for controlling motorized components of the of the microscope.
2. Motorized Z-focus drive with minimum z-step resolution of 15 nm or better.
3. Tilting binocular head.
4. 10X eyepieces with a minimum 22 FN or better.
5. 6 position motorized FL filter wheel & 6 position motorized nosepiece.
6. Computer controlled continuous correctionof Z drift with Laser/LED based system having wavelength above 790nm.
7. Motorized X-Y scanning specimen stage with universal sample holder for slides and 35/60mm petridish, 24/96 well plate for multiple position time lapse imaging and stitching and linear encoders for high reproducibility in positioning.
8. A complete automated digital environmental control stage type incubation system (incubator) for long term live cell time lapse imaging with temperature, CO2 and Humidity sensing and control. The system should be able to use 100% CO2 gas supply and to provide 5% pre-heated CO2 gas to the chamber.
9. 12V /100W halogen illumination for transmitted light or high power LED. Fluorescence light source with average life of more than 22,000 Hours.
10. High resolution Semi Apochromat objectives 2X-2.5X/ 4X- 5X with high NA, Plan Apochromat Confocal Grade 10x/0.45 or better, 20x/0.70 or better, 40x/0.85 or better, 60/63x/1.40 oil & 100X/1.4 oil (wide angle for TIRF application) with DIC prisms for all objectives.
11. Fluorescent bandpass filters for DAPI, FITC/GFP, TRITC/Rhodamine and Cy5/Alexa 647

Confocal system:

High sensitivity confocal laser scanning with built in/separate spectral detectors for efficient fluorescence signal collection. The detectors should be capable of working in intensity and spectral mode imaging.

Scanner should be with the following configuration.

1. High speed scanner with min. 180 deg scan rotation with total scan flexibilities of Line, free hand curved line, XY, XYZ, XYZT, XYZλ and XYZTλ combinations.

2. Scan resolution with spectral detectors should be at least 4K x 4K for all channels or higher.
3. The scan field diagonal should be at least 18-20 mm F.O.V. or more.
4. Scan Zoom range 1:40x or more and should be adjustable in steps of 0.1X.
5. Spectral system should be capable of acquiring minimum of 4 or higher frames per second @512x512 pixel resolution without line skipping or interlacing with scan rotation, ROI, stimulation and simultaneously for 4 channels. It should be able to perform fast dynamic live cell time lapse imaging with a high speed of 110 fps or better @ 512x16 resolution or more. Digitization capability of 8/12/16 bit should be available with the system.
6. High speed scanner which can offer at least 25 fps at 512x512 full frame without ROI and maximum speed up to 250 fps at 512x16.
7. Computer controlled continuously variable confocal single pinhole (aperture size and position) with software control for higher brightness and without affecting sectioning performance.
8. The spectral dispersion of the emission light should be based on either reflection or transmission grating with enhanced/improved spectral signal collection device or with prism based spectral dispersion with high efficiency.

Lasers & Laser Combiner:

1. An AOTF or similar technology controlled laser combiner
2. A longer lifetime stable and easy to switch on and switch off solid state/Diode lasers with powers ≥20mW or better.

   A longer lifetime stable and easy to switch on and switch off solid state/Diode lasers with powers ≥20mW or better

   Laser lines - 405, 488, 515/514, 552/561, 638/641/640
3. All lasers should be connected to the scan head through fiber optic cable and controlled through AOTF for fast laser switching and attenuation in pixel precise synchronization with the laser scanner for ROI scan for FRET, FRAP, Photoactivation /conversion experiments and image analysis.
4. The system should be able to perform multicolor imaging in long duration live cell imaging.
5. The Laser line/s chosen should be reflected on the sample in any combination as per experimental demand.
6. All the visible and UV laser lines should be computer controlled for fast laser switching and attenuation in synchronization with the scanner.

Detectors configuration:

The system should come with either hybrid PMT/GaAsp/HYD based detectors.

An additional transmitted light detector should be offered for bright field and DIC imaging.

PMT and GaAsP Detector configuration:

1. Five tunable detectors / array detector for five fluorophore imaging simultaneously and with at least four detectors should be high sensitive GaAsP/Hybrid detectors with more than 40% QE.
2. The system should be capable of recording emission spectra with minimum spectral resolution of 5nm or better.

sCMOS camera with following specifications should be provided for live cell imaging.

1. Camera with 4mp (6.5 µm) or higher resolution.
2. highest speed of 82 fps

   Speed at lest of 82 fps or better

3. Quantum efficiency: 82% or higher; Readout mode: Rolling shutter; Read out noise: <1 e-; dark current 0.6 e^-/p/s.; pixel well depth: 30000; Linearity: 99.8%.

   Quantum efficiency 82% or higher; Readout mode: Rolling shutter; Read out noise: <1 e-; dark current 0.6 e^-/p/s.; pixel well depth: 30000 or better ; Linearity: 99.8%.
4. Camera should have rolling shutter with global clear.
5. Camera should have 12bit and 16bit mode.

Acquisition and Analysis Software

1. Latest computer control Software capable of controlling motorized functions of microscope, scan head control, laser control including AOTF and Image acquisition & processing etc.
2. The image acquisition and analysis software should have all standard confocal features such as microscope control, Confocal system control, basic modules for ROI imaging bleaching FRET/FRAP/Photoactivation modules for acquisition and analysis. XYZ time lapse, spectral profiling in all the channels and multi-channel sequential and simultaneous image acquisition. Intensity profile over Z time, online spectral fingerprinting/ unmixing/deconvolution, High Dynamic range imaging, 2 D image deconvolution of confocal data sets, Measurements, batch processing,
3. The image analysis software should contain following modules: Basic and advanced functions, quantitative measurement analysis for large and complex objects, lineage tracking of motion/cell division, cell tracking/morphology/segmentation analysis and batch processing capability. Should have Line, curved line, frame, Z-stack, Time series imaging capabilities along with tile and mark and find functions.
4. A dedicated 3D & 4D rendering software, 3D imaging and reconstruction, Co-localization, real time stitching should be provided.
5. Multi point and mosaic imaging facility to be offered.
6. Additional offline software with all the analysis modules as above should be offered.
7. Confocal resolution improvement software that could be integrated in online system for improving XY resolution upto 120- 140 nm and up to 400nm in Z. It should be supported with publication.

Computer Workstation (2 numbers)

1. State of art factory built high power branded Workstation with Windows 10 Professional (64 bit) operating system. - Intel 10-Core Xeon E5-2650 V3 (or higher version) - 32 GB RAM - NVIDiA Quadro K2200 4GB graphic board - 256 GB SATA SSD - 512 GB SATA SSD - 3 TByte SATA hard disc drive - 9.5mm Slim SuperMulti DVD Writer - Ethernet Controller - 2 x USB 2.0 - 8 x USB 3.0 - 3 x IEEE 1394 Firewire B - Keyboard and mouse - Ultra-HD 31” LED Monitor (AH-IPS), GB-r-LED backlight for extended color space, 4096 x 2160 pixels (4K).
2. High Power branded Workstation with Windows 10 Professional (64 bit) operating system. - Intel Xeon Quad core E5-1620 V3 3.5 GHz 10MB - 16 GByte RAM - NVIDIA Quadro K620 1GB high performance GPU - 512 GB SATA SSD - 3 TByte SATA hard disc drive - 9.5mm Slim SuperMulti DVDRW 1st ODD - USB 3.0/2.0, IEEE 1394 A/B - Keyboard and mouse Ultra-HD 31” LED Monitor (AH-IPS), GB-r-LED backlight for extended color space, 4096 x 2160 pixels (4K).

UPS

1. An appropriate UPS (6KVA or above) to give a backup of the entire system for at least 30 minutes.
2. Additional 1KVA UPS to support the offline system for 30 minutes.

Anti- Vibration Table:
should be included with the system from the same manufacturer of Confocal or imported one with active Air Compressor, bread board tabletop with M-6 threading for the complete microscope system. Also, a computer table and rack for lasers to be provided preferably from the confocal manufacturer.
5 years of warranty and AMC for next 5 years after expiry of warranty period.

Provision of multiple training sessions or training personnel by the supplier for handling, maintenance, and training of staff (atleast for two years).