

## Annexure I

### Technical Specifications for an inverted Laser Scanning Confocal Fluorescence Microscope

#### 1.0 Bidder Eligibility Criteria-I

Sl. No	Bidder Eligibility Criteria-I	Complied / Not Complied	Reference Page No.	Remarks, If any
1	The bidder/OEM should have supplied and installed same LCR Meter model quoted at least 2 similar items to IITs, NITs, IISERs, CSIR Labs or other Govt. R&D organizations in the last 10 years worldwide, PO copies or installation certificates along with contact details of end user need to be submitted as the proof of supply. IIT Madras reserves its right to verify the claims submitted by the bidder and the feedback from the previous customers will be part of technical evaluation.			

#### 2.0 Technical Specifications II

S.NO	SPECIFICATION	COMPLIED/ NOT COMPLIED	REFERENCE PAGE NO
<b>MICROSCOPE:</b>			
1	The instrument should be a fully motorized and computer-controlled inverted fluorescence microscope for BF/FL and DIC applications with a remote touch screen TFT/LCD/ Tab display for full control of all motorized microscope functions. It should have the capability for upgradation to include specialised applications such as Optical tweezer, FLIM FCS.		
2	High-precision built-in Z-focus drive with a step resolution of 10 nm or better:		
3	High Precision Motorised Scanning Stage with sufficient working distance to image samples in multi-well plate and 35/60mm petri-dishes etc		
4	Long working distance universal motorised condenser (N.A.0.55/W.D.27mm) with 7 position turrets for DIC Prisms and phase rings.		

5	An ergonomic tilting binocular observation tube with a pair of 10X eyepieces of FN22 or more		
6	The microscope should be equipped with Fluorescence illuminations with a Bright LED/130 W Hg Lamp lamp with a built-in attenuator and shutter for the external fluorescence light source, fibre adapter to an optical port of scan unit, motorized switching between LSM light path and fluorescence illumination. The light source should cover all the wavelengths (DAPI, FITC, TRITC, Cy3) and Cy5 for observation/imaging.		
7	<p><b>Objectives:</b></p> <p>i. Long working distance Plan Apochromat objective 1.25/ 4/5 X objective for imaging large specimen in confocal mode</p> <p>ii. Plan Apochromatic 10X Dry objective</p> <p>iii. Plan Semi Apo chromatic long working distance 20X 0.7NA objective with Phase and correction collar for imaging samples on plastic bottom dishes and slides with or without cover glass</p> <p>iv. High-Resolution Plan Apochromatic 40X 0.95 NA Dry objective with a correction collar to adjust the depth of imaging of thick tissues</p> <p>v. High-resolution Plan Apochromatic 60X NA 1.42 or better Oil immersion objective</p> <p>vi. High-resolution Plan Apochromatic 100X NA 1.45 or better Oil immersion objective</p>		
8	Motorized fluorescence turret with 8 positions for a filter cube and in-built shutter and individual bandpass filters for fluorescence filters for DAPI, FITC/GFP, and TRITC/Rhodamine/Cy3 and Cy5 should be quoted.		
9	Motorized 6-position DIC nose piece		
10	Camera Port: Camera port (prepared) on the side port should have a motorized side port for camera attachment and motorized beam path selection between eye observation and confocal imaging.		
11	IR/LED-based drift compensator (Hardware Module) to		

	maintain the same focus throughout long-hour experiments (more 24 Hrs). The drift compensator should work with both plastic and cover glass bottom dishes with one-touch auto focus, continuous autofocus and one-shot autofocus modes for various biophysical applications.		
<b>CONFOCAL IMAGING SPECIFICATION</b>			
1	The state-of-the-art confocal system should have a set of resonant scanners for precise high-resolution imaging with different scan resolution formats from at least 4x4 or better. A high-speed resonant scanner should be offered with a minimum FOV of 18 mm or more. @ 1X Zoom,		
2	The system should be equipped with a dedicated high speed resonant scanner with scanning video rate speed of upto 25 FPS at 512x512 at one X Zoom without compromising the full FOV of 15-18mm.		
3	Laser point scanning and built-in confocal detection unit with two true Spectral High sensitive GaAsP/HyD detectors and two photomultiplier tube (PMT) based detectors. These PMT based detectors should be upgradable to dedicated NIR detectors as a 5 <sup>th</sup> detector with spectral sensitivity 18-20% @850nm		
4	Spectral detectors should be with independent voltage and gain controls and each detector must be able to work in both, intensity mode and spectral mode. All the fluorescence detectors must be filter-free spectral type and should be in-built inside the scan head for better sensitivity.		
5	The minimum detectable spectral bandwidth of each detector should be at least 2 nm or better throughout the visible spectral range of 400-750 nm or better, and the maximum spectral bandwidth at one go must be at least 100 nm or higher for each detector. The linear spectral dispersion should be through Diffraction grating or prism with the ability to select any desired bandwidth from both sides of the spectrum with a minimum bandwidth of 2nm throughout the spectral range of 400-750nm to avoid any autofluorescence/breakthrough.		
6	The detectable bandwidth should be adjustable at every 2 nm or better increments from 400-750nm. A shorter		

	bandwidth adjustment would be needed for samples having a lot of autofluorescence and background fluorescence.		
7	The scanner should be able to scan in various areas such as rectangles, clips, polygons, lines etc., and a scanning zoom of 1X- 40Xtimes or better with ROI Scan should be achieved. Scan rotation of 180 degrees or better should be available.		
8	Confocal scan FOV should be at least 18 mm or better at Zoom 1.		
9	Scanning Imaging Modes: (i) Line, curved line, frame, Z-stack, Time series, tile imaging capabilities; (ii) Real ROI bleach for FRAP, Photo-activation/conversion experiments. (iii) Spectral un-mixing with fingerprinting for separation of overlapping excitation/emission spectra of fluorophores. (iv) Standard geometry Measurements like length, areas, angles, etc including intensity measurements. FRET and FRAP image causation and analysis		
<b>CAMERA</b>			
1	Camera: Monochrome scientific sCMOS based camera with cooled sCMOS sensor.		
2	No. of pixels: 2048 x 2048 or higher		
3	Pixel size : 8 x 8 $\mu\text{m}^2$ or lower.		
4	Speed: 30 frames per second or higher at full resolution		
5	Quantum efficiency: 80 % or higher		
6	Cooling capability : Should able to cool -10 °C at ambient temperature of 25°C		
<b>LASER</b>			
1	The system should have at least the following laser lines: 405/408nm, 488nm, 561nm, and 635/638/640 nm or equivalent. All the lasers should have a minimum power of 20mW or above. It should be upgraded to 3 visible lasers (445, 514 and 594nm controlled by same AOTF. Alternatively, a white light laser may be quoted.		
2	All lasers including 405 nm should be connected to the scan head through a fibre optic cable, controlled by an acoustic-optic tuneable filter AOTF; for precise switching and swift selection of the desired laser lines and		

	attenuation in pixel-precise synchronization with the laser scanner for Real ROI scan for FRAP.		
3	The system should be future upgradable to perform Excited state dynamics measurements for SINGLE MOLECULE DETECTION experiments with Fast FLIM image acquisition for rapid kinetic analysis such as FLIM-FRET.		
<b>WORK-STATION AND SOFTWARE</b>			
1	The vendor should provide a high-end workstation with Windows 10/11 Professional (64 bits) operating system, Intel Xeon processor (8 core system or better), with 96 GB working memory, and Nvidia Quadro graphics card with at least 8 GB graphic card memory. Should come with a fast 1TB SSD system drive and an additional 2 TB normal HDD drive for the temporary storage of data. Should come with a suitable network adapter and an HP 9.5mm Slim DVD writer. Keyboard, mouse and mouse pad and 2 Thunderbolt ports. The workstation should come with a high brilliance monitor of display size 37.5”, and a resolution of 4K-Wide 3840 x 1600 @ 60 Hz.		
2	The imaging software should control all the motorized functions of the confocal microscope. It should have the capability of multi-dimensional acquisition namely XY, XT, XZ, YZ, XZT, XYT, XYZ, XYZT.		
3	Image processing of 3D data having features like Transparent, Maximum Intensity and Depth Coding, shadow projection, clipping, Orthogonal Sectioning and Annotation tool to add comments to 3D volume. 3D construction, 4D ( XYZt) rendering.		
4	The software should have analysis functions such as intensity measurement over time, over depth and over lambda. Advance measurements like real-time ratio imaging and co-localization should be possible. The software should have an online ratio, FRET, FRAP acquisition, FRET and FRAP acquisition and analysis, and a spectral unmixing module.		
5	Confocal system control software for control of scan process and image processing. - Control of motorized hardware. - Multidimensional image acquisition. - Software-aided hardware configuration based on fluorophores used. - Processing, quantification and easy		

	export of data. - Extension of functionality with additional licenses. - Online software and hardware manual. – should include hardware dongle and installation CD.		
6	Suitable active anti-vibration table with compressed air damping, for the complete microscope system		
7	The vendor must have a company-certified engineer based in Tamil Nadu for instant post-sale support and the lowest downtime.		
<b>MISCELLANEOUS/ ADDITIONAL TERMS AND CONDITIONS</b>			
1	System and accessories should work with 220v @50 HZ.		
2	Power requirements for the laser to be specified.		
3	The principals/local agents are responsible for the complete installation, testing, and integration of the system.		
4	Operational and application training should be provided on-site.  Operating manuals should be provided along with the system.		
5	The quote should clearly specify the warranty or guarantee period for the machinery/equipment. The warranty should be for three years from the date of commissioning of the equipment.		
6	AMC- 2 years optional warranty. (Will not be considered for price bid evaluation)		