**Abberior Instruments STED**

Fluorescence inverted confocal microscope and STED super-resolution nanoscope with high detection efficiency of far-red photons. The system is equipped with four excitation lasers, depletion laser 775 nm and four single photon counting detectors. For more detailed microscope characteristics please see the section “Microscope”.

Basic introduction to STED microscopy can be found here: <http://www.abberior.com/knowledge/microscopy-tutorials/sted/>

**Application**

* Two-color 2D and 3D super-resolution images obtained by STED technique with a pulsed depletion laser 775 nm and pulsed excitation 561 nm and 640 nm
* STED RESCue mode available (STED imaging mode that significantly reduces the light dose sent onto the sample without compromising the resolution)
* 2D and 3D STED images available also with water immersion objective
* Multicolor confocal scanning system with variable pinhole size
* Possible FLIM or FCS acquisition
* Live cell imaging available

**Microscope**

Inverted confocal microscope Nikon Eclipse Ti-E equipped with a piezo Z-stage, motorized XY stage, Perfect Focus System, transmitted light lamp (100 W) and following units:

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| Software | **Imspector** with EasyCommander module |
| Epifluorescence | **CoolLED pE-4000**  (excitation range: 405±20 nm, 480±20 nm, 560±20 nm, 640±20 nm) |
| Excitation lasers | **cw - 405 nm, pulsed - 485 nm, 561 nm, 640 nm** |
| STED laser | **775 nm** (40 MHz pulsed laser, for 2D and 3D STED) |
| Donut formation | Programmable **Spatial Light Modulator** – 775 nm |
| Scanner | **Abberior QUAD scanner**: line frequency up to 2 kHz |
| Objectives | Nikon CFI **Plan Apo Lambda 60x Oil, NA 1.40**, WD 0.13 mm  Nikon CFI **Plan Apo 60x WI, NA 1.27**, WD 0.17 mm, Correction Collar 0.15-0.19, DIC  Nikon CFI **Plan Apo Lambda 10x, NA 0.45**, WD 4 mm, DIC |
| Emission filters | 422-467 nm  500-550 nm  605-625 nm or 580-630 nm  650-720 nm |
| Pinholes | 16 pinholes from 25 μm to 2 mm |
| Detectors | 4x **Photon Counting Module** (Excelitas Technologies)   * + Two time resolved detectors optimized for FLIM   + High detection sensitivity for red light (>62% for 680nm)   **PMT** for transmitted and reflected light (Hamamatsu Photonics)  Monochrome **widefield camera DMK 23U274** (Imaging Source)   * CCD sensor, 1600×1200 (1.9 MP), up to 20 fps, pixel size 4.4 µm x 4.4 µm |
| Acquisition modes | Multi-color acquisition method via interleave mode  (i) pulse interleaved and/or (ii) pixel interleaved and/or (iii) line-interleaved |
| FLIM | TCSPC card: SPC-150 (Becker & Hickl)  Acqusition   * FLIM module in Imspector * SPCM and SPCImage softwares |