

Bangalore Microscopy Course: 2017

National Centre for Biological Sciences and Centre for Cellular And
Molecular Platforms, Bangalore, September 17-24, 2017

Course Organizers

Ron Vale, Nico Stuurman: University of California, San Francisco, USA

Satyajit Mayor, H. Krishnamurthy: National Centre for Biological Sciences, India

Rahul Roy: Indian Institute of Science, India

Manoj V Mathew: C-CAMP, India

Sangeeta Nath: inStem, India

Faculty:

Christian Tischer, EMBL, Germany

Erik Manders, University of Amsterdam, Netherlands

Clare M. Waterman, NIH/NHLBI, USA

Jennifer Ross, UMass Amherst, USA

Nathan Claxton, Nikon Instruments Inc., USA

Rahul Roy, Indian Institute of Science, India

Satyajit Mayor, National Centre for Biological Sciences, India

Stephen Ross, Nikon Instruments Inc., USA

Sudipta Maiti, Tata Institute for Fundamental Research, India

Takahiro Fujiwara, Kyoto University, Japan

Support and Attendance by:

Carl Zeiss, Confocal.nl, DSS Imagetech, Hamamatsu, Icon Analytical, Leica Microsystems,
Luxendo, Nikon, Olympus, Okolabs, Photometrics, Towa Optics, Tokai Hit, Scientific
Volume Imaging

Course Schedule

Sunday, Sept. 17: Opening Lecture, Poster Session

- 12:30-01:30pm** **Lunch**
Location: 1st Floor, Main Canteen
- 01:30-02:30pm** **Participants Registration**
Location: Southern Laboratories Complex (SLC) Reception
- 02:30-04:00pm** **Opening Remarks:** Satyajit Mayor, NCBS, India
Research Lecture 1: Jan Ellenberg, EMBL, Heidelberg
Location: Happus, LH1 (Eastern Laboratory Complex)
- 04:00-04:15pm** **Coffee Break**
- 04:15-05:30pm** **Didactic Lecture 1:** Basics of Microscopy, Köhler illumination and Resolution, Diffraction. - Rahul Roy, IISc, India
Location: Happus, LH1 (Eastern Laboratory Complex)
- 05:30-08:00pm** **Poster session** by the students attending the course & Social
Location: Southern Laboratories Complex (SLC) Atrium
- 08:00-09:00pm** **Dinner**
Location: 1st Floor, Main Canteen

Monday, Sept. 18: **Basics of microscopy**

- 08:30-01:15pm** **Laboratory 1:** Microscopy light path, Köhler illumination on rails and Introduction to Transmitted Light Microscopy
Samples: Resolution and Distortion Test Target, Bead Slides, Diatoms, Pond Water
Lab leader: Jennifer Ross
Lab Instructors: Satyajit Mayor, Christian Tischer, Rahul Roy, Stephen Ross, Erik Manders, Clare M. Waterman, Nathan Claxton, Takahiro Fujiwara
Lab Support: Chandrima and Rimple
Location: Raspuri (New Teaching Lab)
- 11:00-11:15am** **Coffee Break**
- 01:15-02:15pm** **Lunch**
Location: 1st Floor, Main Canteen
- 02:15-03:15pm** **Didactic Lecture 2:** Contrast enhancement -Phase Contrast, Dark Field, Polarized Light, DIC - Stephen Ross, Nikon Inc., USA
Location: Raspuri (New Teaching Lab)

- 03:15-04:15pm** **Didactic Lecture 3:** Principles of Fluorescence and Fluorescence microscopy- Clare M. Waterman, NIH/NHLBI, USA
Location: Raspuri (New Teaching Lab)
- 04:15-04:30pm** **Coffee Break**
- 04:30-06:30pm** **Laboratory 2:** Phase Contrast, DIC and Dark Field Microscopy
Lab leader: Stephen Ross
Lab instructors: Clare M. Waterman, Christian Tischer, Erik Manders, Jennifer Ross, Rahul Roy, Satyajit Mayor, Nathan Claxton, Takahiro Fujiwara
Lab Support: Amrutha and Rimple
Location: Raspuri (New Teaching Lab)
- 06:30-07:30pm** **Didactic Lecture 4:** Fluorescent dyes, Fluorescent Proteins and Selection of Fluorescent Probes - Rahul Roy, IISc, India
Location: Raspuri (New Teaching Lab)
- 07:30-08:30pm** **Dinner**
Location: 1st Floor, Main Canteen
- 08:30-11:00pm** **Free time on microscopes**

Tuesday, Sept. 19: Fluorescence Microscopy

- 08:30-10:30am** **Laboratory 3:** Fluorescence Lab A: Examine the fluorescence light path and dichroic mirrors/filters
Lab leader: Rahul Roy
Lab instructors: Christian Tischer, Stephen Ross, Takahiro Fujiwara, Jennifer Ross, Erik Manders, Satyajit Mayor, Clare M. Waterman, Nathan Claxton
Lab Support: Chandrima and Amrutha
Location: Raspuri (New Teaching Lab)
- 10:30- 10:45am** **Coffee Break**
- 10:45-11:45pm** **Didactic Lecture 5:** Practical Aspects of Objective Lens Design, covering lenses and aberrations- Stephen Ross, Nikon Instruments Inc., USA
Location: Raspuri (New Teaching Lab)
- 11:45-12:45pm** **Didactic Lecture 6:** Speckle Microscopy, Clare M. Waterman, NIH/NHLBI, USA
Location: Raspuri (New Teaching Lab)
- 12:45-01:45pm** **Lunch,** 1st Floor, Main Canteen

- 01:45-03:45pm** **Laboratory 4:** Fluorescence Lab B: Time Lapse Imaging and Z projections of PSF
Lab leader: Erik Manders
Lab instructors: Christian Tischer, Jennifer Ross, Rahul Roy, Stephen Ross, Takahiro Fujiwara, Satyajit Mayor, Clare M. Waterman, Nathan Claxton
Lab Support: Chandrima and Rimple
Location: Raspuri (New Teaching Lab)
- 03:45-04:45pm** **Didactic Lecture 7:** Image Analysis- Christian Tischer, European Molecular Biology Laboratory, Germany
Location: Happus (LH1)
- 04:45-05:00pm** **Coffee Break**
- 05:00-07:00pm** **Laboratory 5:** Image Processing/Analysis Workshop
Lab Leader: Christian Tischer
Lab Support: Mugdha and Lakshmi
Location: Raspuri (New Teaching Lab)
- 07:00-08:00pm** **Free time on microscopes**
- 06:30-09:00pm** **Dinner** (For Faculty- Down Town Bangalore)
- 08:00-09:00pm** **Dinner** (For Participants, Others - 1st Floor, Main Canteen)
- 09:00-11:00pm** **Free time on microscopes**

Wednesday, Sept. 20: Optical Sectioning Techniques

- 08:30-09:30am** **Didactic Lecture 8:** TIRF Microscopy- Nathan Claxton, Nikon Inc., USA
Location: Malgova
- 09:30-10:30am** **Research Lecture 2:** Jennifer Ross, UMass Amherst, USA
Location: Malgova
- 10:30-10:45am** **Coffee Break**
- Session 1**
10:45-12:45pm **Laboratory 6:** Optical Sectioning Techniques (Point Scanning and Spinning Disk) 2 x 2 hours modules
- Session 2**
02:45-04:45pm **Lab Leader:** H. Krishnamurthy
Lab Support: Amrutha, Chandrima, Divya, Ranjana, Rashmi, Rimple, Manisha, Ratnasri K, Saurabh Umrao, Sunaina Banerjee

- 1. Equipment:** Olympus FV3000 point scanning microscope
Lab Instructor: Olympus Application Specialist
Location: CIFF, Southern Laboratories Complex (SLC)
- 2. Equipment:** Leica SP5 point scanning microscope
Lab Instructor: Leica Application Specialist
Location: CIFF, Southern Laboratories Complex (SLC)
- 3. Equipment:** Andor Spinning Disk microscope
Lab Instructor: Chaitra
Location: Mayor Lab, Southern Laboratories Complex (SLC)
- 4. Equipment:** Zeiss LSM 780 point scanning microscope
Lab Leader: Sudipta Maiti
Lab Instructor: Thomas
Location: CIFF, Southern Laboratories Complex (SLC)
- 5. Equipment:** Zeiss LSM 880 point scanning microscope
Lab Instructor: Zeiss Application Specialist
Location: CIFF, Southern Laboratories Complex (SLC)
- 6. Equipment:** Perkin Elmer Spinning Disk System
Lab Instructor: Joseph
Location: CIFF, Southern Laboratories Complex (SLC)

- 12:45-01:45pm** Lunch, 1st Floor, Main Canteen
- 01:45-02:45pm** **Didactic Lecture 9:** Optical Sectioning Techniques- Confocal, Two Photon Excited Fluorescence, etc.- Sudipta Maiti, TIFR, India
Location: Raspuri
- 04:45-05:00pm** Coffee Break
- 05:00-06:00pm** **Research Lecture 3:** Clare M. Waterman, NIH/NHLBI, USA
Location: Happus (LH1)
- 06:00-7:00pm** Dinner, 1st Floor, Main Canteen
- 07:00-10:00pm** Cultural and Social Event

Thursday, Sept. 21: Special Topics

- 08:30-09:30am** **Didactic Lecture 10:** Fluorescence Microscopy based methods to study protein dynamics in live cells: from FRAP to FCS - Sudipta Maiti, TIFR, India
Location: Raspuri (New Teaching Lab)
- 09:30-10:30am** **Didactic Lecture 11:** Stimulated Emission Depletion Microscopy (STED), Rahul Roy, IISc, India
Location: Raspuri (New Teaching Lab)

10:30-10:45am

Coffee Break

Session 1

10:45-12:15pm

Laboratory 7: Specialized techniques 2 x 1.5 hours modules, distributed by student signup and selection. There will be 2 rotations of 1.5 hours each.

Session 2

01:15-02:45pm

Lab Support: Amit, Amrutha, Chandrima, Divya, Jesvin, Kanika, Manisha, Ranjana, Rimple, Rashmi, Ratnasri K, Saurabh Umrao, Sunaina Banerjee, Sankarshan

Technique 1: Integrating microscope systems and Configuring Automation Software for Microscope

Lab Instructor: Stephen Ross, Nathan Claxton

Equipment: Nikon Ti Microscope

Location: Raspuri (New Teaching Lab)

Technique 2: Homo-Försters Resonance Energy Transfer (Homo-FRET)

Lab Leader: Satyajit Mayor

Lab Instructor: Joey

Equipment: Nikon TIRF microscope with dual camera

Location: Mayor Lab, Southern Laboratories Complex (SLC)

Technique 3: Fluorescence Correlation Spectroscopy (FCS)

Lab Instructor: Sudipta Maiti

Lab Instructor: Thomas

Equipment: Zeiss LSM 780 NLO

Location: CIFF, Southern Laboratories Complex (SLC)

Technique 4: Laser Micro Dissection and Catapulting

Lab Instructor: Ritusree and Zeiss Application Specialist

Equipment: PALM Microbeam Laser microdissection and catapulting-Carl ZEISS

Location: CIFF, Southern Laboratories Complex (SLC)

Technique 5: TIRF Microscopy

Lab Instructor: Ananya/Nishan

Equipment: Olympus cellTIRF-4Line Microscope

Location: 2nd Floor, Southern Laboratories Complex (SLC)

Technique 6: SIM Super Resolution Microscopy

Lab Instructor: Nikon Application Specialist

Equipment: Nikon N-SIM

Location: CIFF, Southern Laboratories Complex (SLC)

Technique 7: Light Sheet Microscopy

Lab Instructor: Luxendo Application Specialist

Equipment: Luxendo – Invi-SPIM

Location: CIFF, Southern Laboratories Complex (SLC)

Technique 8: Re-Scan Super Resolution Confocal Microscopy

Lab Instructor: Erik Manders

Equipment: Confocal.nl Re-Scan Confocal Microscope

Location: CIFF, Southern Laboratories Complex (SLC)

Technique 9: Fluorescence Recovery After Photobleaching (FRAP)

Lab Instructor: Joseph Jose Thottacherry

Equipment: Perkin Elmer Spinning Disk System

Location: CIFF, Southern Laboratories Complex (SLC)

Technique 10: Airyscanning Super Resolution Confocal Microscopy

Lab Instructor: ZEISS Application Specialist

Equipment: ZEISS LSM 880 Airy scanning microscope

Location: CIFF, Southern Laboratories Complex (SLC)

Technique 11: Automated Platform for Live Cell Imaging

Lab Instructor: ZEISS Application Specialist

Equipment: ZEISS Celldiscoverer 7

Location: CIFF, Southern Laboratories Complex (SLC)

12:15- 01:15pm Lunch, 1st Floor, Main Canteen

02:45-03:00pm Coffee Break

03:00-05:00pm Technical Presentations by the members of the industry

Location: Happus (LH1)

(03:00-03:15pm) **Talk 1:** Light sheet microscopy on the edge: high speed volume imaging for delicate samples, Jürgen Mayer, **Luxendo**

(03:15-03:30pm) **Talk 2:** New Stage Top Incubator STX series - Happiness for Cells, Success for Researchers, Shinichiro Endo, **Tokai Hit**

(03:30-03:45pm) **Talk 3:** ZEISS Celldiscoverer 7, Your Automated Platform for Live Cell Imaging, Dr. Xianke Shi, **Carl Zeiss**

(03:45-04:00pm) **Talk 4:** Huygens Deconvolution, Visualization and Analysis to find the truth behind your blurry and noisy microscopic image, Drs. Gitta Hamel, **Huygens**

(04:00-04:15pm) **Talk 5:** Infrared Photo-induced Force Microscopy (IR PiFM), Dr. Sung Park, **Icon Analytical**

(04:15-04:30pm) **Talk 6:** OLYMPUS MPE-RS: Live in-situ brain mapping and optogenetics Multiphoton Microscopes with dedicated MPE Objectives, Ganesh Kadasoor, Ph.D., **Olympus**

(04:30-04:45pm) **Talk 7:** Flexibility of Super Resolution imaging with Nikon platform, Dr. Nabanita Chatterjee, **Nikon**

- (04:45-05:00pm)** **Talk 8:** New DMi8 S – Cutting Edge Live Cell Imaging Systems, Dr. Peter Laskey, **Leica Microsystems**
- 05:00-05:15pm** **Coffee Break**
- 05:15-06:15pm** **Didactic Lecture 12:** Single Molecule Imaging - Takahiro Fujiwara, Kyoto University, Japan
Location: Raspuri (New Teaching Lab)
- 06:15-07:30pm** **Graduation, Group Photo**
- 07:30-08:30pm** **Dinner**, 1st Floor, Main Canteen
- 08:30-10:00pm** **Free time on microscopes**
Laboratory 4 (Supplementary): Camera characterization

Friday, Sept. 22: Research Lectures, Specialized Techniques

- 08:30-09:30am** **Didactic Lecture 13:** FRET Microscopy- Satyajit Mayor, NCBS, India.
Location: Happus (LH1)
- 09:30- 10:30am** **Research Lecture 4:** Sudipta Maiti, TIFR, Mumbai
Location: Happus (LH1)
- 10:30-10:45am** **Coffee Break**
- Session 1**
10:45-12:15pm **Laboratory 8:** Specialized techniques, 2 x 1.5 hours modules, distributed by student signup and selection. There will be 2 rotations of 1.5 hours each.
- Session 2**
02:15-03:45pm **Lab Support:** Amit, Amrutha, Chandrima, Divya, Jesvin, Kanika, Manisha, Ranjana, Rimple, Rashmi, Ratnasri K, Saurabh Umrao, Sunaina Banerjee, Sankarshan
- Technique 1:** Integrating microscope systems and Configuring Automation Software for Microscope
Lab Instructor: Stephen Ross, Nathan Claxton
Equipment: Nikon Ti Microscope
Location: Raspuri (New Teaching Lab)
- Technique 2:** Homo-Försters Resonance Energy Transfer (Homo-FRET)
Lab Leader: Satyajit Mayor
Lab Instructor: Joey
Equipment: Nikon TIRF microscope with dual camera
Location: Mayor Lab, Southern Laboratories Complex (SLC)
- Technique 3:** Fluorescence Correlation Spectroscopy (FCS)
Lab Leader: Sudipta Maiti
Lab Instructor: Thomas
Equipment: Zeiss LSM 780 NLO
Location: CIFF, Southern Laboratories Complex (SLC)

Lab Instructor: Ritusree and Zeiss Application Specialist
Equipment: PALM Microbeam Laser microdissection and catapulting-Carl ZEISS
Location: CIFF, Southern Laboratories Complex (SLC)

Technique 5: TIRF Microscopy
Lab Instructor: Ananya/Nishan
Equipment: Olympus cellTIRF-4Line Microscope
Location: CIFF, Southern Laboratories Complex (SLC)

Technique 6: SIM Super Resolution Microscopy
Lab Instructor: Nikon Application Specialist
Equipment: Nikon N-SIM
Location: CIFF, Southern Laboratories Complex (SLC)

Technique 7: Light Sheet Microscopy
Lab Instructor: Luxendo Application Specialist
Equipment: Luxendo – Invi-SPIM
Location: CIFF, Southern Laboratories Complex (SLC)

Technique 8: Re-Scan Super Resolution Confocal Microscopy
Lab Instructor: Erik Manders
Equipment: Confocal.nl Re-Scan Confocal Microscope
Location: CIFF, Southern Laboratories Complex (SLC)

Technique 9: Fluorescence Recovery After Photobleaching (FRAP)
Lab Instructor: Joseph Jose Thottacherry
Equipment: Perkin Elmer Spinning Disk System
Location: CIFF, Southern Laboratories Complex (SLC)

Technique 10: Airyscanning Super Resolution Confocal Microscopy
Lab Instructor: ZEISS Application Specialist
Equipment: ZEISS LSM 880 with Airy scan Module
Location: CIFF, Southern Laboratories Complex (SLC)

Technique 11: Automated Platform for Live Cell Imaging
Lab Instructor: ZEISS Application Specialist
Equipment: ZEISS Celldiscoverer 7
Location: CIFF, Southern Laboratories Complex (SLC)

12:15-2:15pm **Discussion over Lunch:** Small group discussions of students with faculty over lunch about their Research/Microscopy interests

03:45-04:00pm **Coffee Break**

04:00-05:00pm **Didactic Lecture 14:** Structured Illumination Microscopy (SIM) and it's relation to Re-scan Confocal Microscopy (RCM), Image Scanning Microscopy (ISM) and Confocal Microscopy, Erik Manders, University of Amsterdam, Netherlands

- Location: Happus (LH1)**
- 05:00-06:00pm** **Research Lecture 5:** Satyajit Mayor, NCBS, India
Location: Happus (LH1)
- 6:00-10:30pm** **Workshop Dinner and Social (Downtown Bangalore)**

Saturday, Sept. 23: Projects

- 09:00-10:00am** **Research Lecture 6:** Takahiro Fujiwara, Kyoto University, Japan
Location: Happus (LH1)
- 10:00-11:00am** **Research Lecture 7:** Rahul Roy, IISc, India
Location: Happus (LH1)
- 11:00-11:15am** **Coffee Break**
- 11:15-01:00pm** **Projects**
- 01:00-02:00pm** **Lunch**
- 02:00-04:00pm** **Projects**
- 04:00-04:15pm** **Coffee Break**
- 04:15-07:15pm** **Projects**
- 07:15-8:15pm** **Dinner, 1st Floor, Main Canteen**

Sunday, Sept. 24: Projects

- 09:00-11:00am** **Projects**
- 11:00-11:15am** **Coffee Break**
- 11:15-01:00pm** **Projects**
- 01:00-02:00pm** **Lunch**
- 02:00-04:00pm** **Projects**
- 04:00-04:15pm** **Coffee Break**
- 04:15-06:15pm** **Project Presentation by Participants**

Projects

Project-1: Imaging Brain Slices-Widedfield vs. Two-Photon Microscopy

Guide: Kambadur Ananthamurthy and Sriram Narayanan

Equipment: Beja Fry Multiphoton Microscope

Location: CIFF Southern Laboratories Complex (SLC) and Lab 1

Project-2: Characterization of STED imaging and STED-FCS

Guide: Manoj

Lab Support: Amit/Nishan

Equipment: Abberior STED

Location: CIFF Southern Laboratories Complex (SLC)

Project-3: Image Processing

Guide: Christian Tischer

Lab Support: Mugdha and Lakshmi

Equipment: Perkin Elmer Spinning Disk and Olympus FV3000

Location: CIFF Southern Laboratories Complex (SLC)

Project-4: Diffusion law and its experimental executions

Guide: Thomas

Lab Support: Chandrima Patra

Equipment: Zeiss 780 NLO

Location: CIFF Southern Laboratories Complex (SLC)

Project-5: Ultrafast single molecule TIRF microscope to follow dynamics of fluorescence conjugated Halo-GPI on membrane surface.

Guide: Sangeeta

Lab Support: Ananya Sadhu

Equipment: Nikon Ultrafast TIRF Microscope

Location: 1st Floor Clean Room, Southern Laboratories Complex (SLC)

Project-6: Fluorescent Non-canonical Amino Acid Tagging (FUNCAT)

Guide: Jesvin

Lab Support: Kanika

Equipment: Olympus FV3000

Location: CIFF Southern Laboratories Complex (SLC)

Project-7: Optical Tweezers

Guide: Chittaspandini Kulkarni

Lab Support: Sankarshan

Equipment: Thorlabs Optical Tweezers Kit

Location: 1st Floor Clean Room, Southern Laboratories Complex (SLC)

Project-8: Polarization sensitive Emission anisotropy based Total Internal reflection microscopy to study molecular ordering of integrins in focal adhesions

Guide: Joseph K Mathew

Lab Support: Sarayu

Equipment: Nikon TIRF with Dual Cameras

Location: R4, Mayor Lab, 1st Floor, SLC