Overview and Goals

The Optical Biology Core (OBC) comprises three facilities on the UC Irvine campus: 1) a self-use OBC facility equipped with fluorescence microscopes and image analysis software, 2) the Non-Linear Optical Microscopy lab (NLOM), a collaborative facility dedicated to the use of lasers and other optics in Biology and Medicine, and 3) a flow cytometry facility in Hewitt Hall equipped with three multiparameter flow cytometers. The OBC is a Shared Resource funded in part by the Chao Family NCI-Comprehensive Cancer Center Support Grant (P30 CA062203) from the National Cancer Institute.

1) OBC Self-Use Facility:

The facility, located on the main UCI campus, is equipped with four confocal microscopes. Among them is the recently installed Zeiss LSM 980 Airyscan with 2-photon capabilities, a crucial feature for deep tissue imaging, Fluorescence Lifetime Imaging (FLIM), and Second Harmonic Generation (SHG). Both the LSM 980 and Zeiss LSM 780 are equipped with single photon lasers covering the spectrum from DAPI (405nm) to far-red (633nm) and a 32-channel spectral detector. Additionally, the facility houses a Zeiss LSM 900 with Airyscan, offering 4 laser lines, and Leica SP8 confocal microscopes, each with 6 laser lines from UV to far-red, equipped with 2 HyD and 2 PMT detectors. The Airyscan detectors on the LSM 980 and LSM 900 significantly enhance resolution, signal-to-noise ratio, imaging speed, and multicolor capabilities. They support 3D and super-resolution imaging, reduce photo-damage, and are compatible with various sample types. The user-friendly software and the possibility of upgrading existing equipment make them invaluable tools for high-quality fluorescence imaging in various research domains.

The facility also features a Zeiss Z1 Lightsheet (Single Plane Illumination Microscope) designed for imaging both live samples and cleared tissues. The Z1 is equipped with 4 laser lines (405nm, 488nm, 561nm, and 633nm) and a custom chamber for organically cleared samples. Additionally, a super-resolution Lattice SIM Elyra 7 with Single Molecule Localization (SMLM) capability is available, offering an impressive 60nm resolution with SMLM achieving resolutions as fine as 10nm. Moreover, the system supports Total Internal Reflective Fluorescence (TIRF) imaging, which is particularly valuable for investigating events and processes occurring at or near the cell membrane.

Users have access to multiple workstations for data analysis, featuring software such as Imaris for advanced 3D/4D analysis, Imaris Stitcher, Arivis Vision 4D, and Huygens Deconvolution. Importantly, these instruments are available for use 24/7. The technologies and services provided in this facility are:

- Confocal microscopy
- 2-photon microscopy
- Airyscan confocal microscopy
- Live Cell Imaging
- Single Plane Illumination Microscopy (SPIM)
- Lattice SIM and STORM localization microscopy
- TIRF Imaging
- Fluorescence Lifetime Imaging Microscopy (FLIM) / FRET via FLIM
- Single particle tracking
- Image Correlation Spectroscopy (ICS) / Raster Image Correlation Spectroscopy (RICS)
- Mapping of molecular aggregates using Number & Brightness (N&B) analysis
- Extensive training and workshops to enable users to deploy the full capabilities of the systems.
- Further information regarding all services is on the OBC page on the Cancer Center (CFCCC) website.

2) The NLOM Lab:

is located in the Beckman Laser Institute and Medical Clinic (BLIMC). It is equipped with an extensive array of commercial and home-built microscopes for confocal and nonlinear optical microscopy as well as for conventional optical microscopy. Besides the imaging platforms provided as shared resources, the NLOM Lab provides collaborative opportunities for using the multiphoton microscopy (MPM)-based technologies developed in the lab for highly optimized human skin imaging with the goal of non-invasively diagnosing skin cancers and other skin diseases and understanding skin biology and functionality. Users are supported by extensive shop facilities that allow construction and modification of imaging platforms (https://sites.uci.edu/nlom/).
The imaging platforms provided as shared resources in this facility are:

- Leica SP8 Falcon + coherent anti-Stokes Raman Scattering (CARS) for confocal and nonlinear optical microscopy. Imaging modalities include confocal and two-photon excited fluorescence (TPEF), second harmonic generation (SHG), CARS and fluorescence lifetime microscopy (recently acquired through a 1.6 mil high-end shared instrumentation grant).
- Zeiss LSM 510 for conventional optical microscopy as well as confocal and nonlinear optical microscopy (imaging modalities include confocal, TPEF and SHG microscopy)

3) The Flow Cytometry Facility: The Institute for Immunology (IFI) runs an open-use Flow Core Facility providing the latest technology and professional technical assistance for flow cytometric analysis, flow-imaging, and sorting. The facility operates a suite of four multi-parameter flow cytometers that are well equipped for fluorescence-activated cell sorting (FACS) and emerging flow cytometry assays. The facility manager, Dr. Michael Hou, PhD, has more than 10 years of experience in the use of numerous flow cytometry platforms and continues to develop new applications with the advancement of instruments and emerging applications. Dr. Hou is available for one-on-one consultation for experimental design, instruction on use of the instruments, and data analysis.

The technologies provided in this facility are:

- **Amnis® ImageStream®x Mark II Imaging Flow Cytometer** combines the phenotyping abilities of flow cytometry with the detailed imagery and functional insights of microscopy.
  - Excitation lasers at 405nm, 488nm and 642nm allows for 12 high-resolution images of every event in the flow cell, including bright field, dark field and 10 fluorescent channels.
  - Multiple magnifications with 20X/40X/60X objectives (motorized and autofocus)
  - Speed: up to 5000 cells/sec (ideal for rare cell analysis)
  - Applications are for quantitative and qualitative measurements (i.e. cell signaling and intensity, internalization, cell cycle, morphology, cell-cell interaction, co-localization, autophagy, etc)
  - Data are analyzed using IDEAS® software loaded on a separate workstation.
- **ACEA NovoCyte Quanteon Flow Cytometer** features 4 lasers and 25 Silicon photomultipliers with 7.2 logs of dynamic range plus forward and side scatter parameters. The photodetectors and signal processing allow for exceptional sensitivity and resolution, including particles at 100 nm. It has a flexible optical configuration for user-directed choice of mirrors and filters. The fluidic system is exceptionally stable and includes automation for high throughput analysis of multiple plate formats (24/48/96/384) in addition to support for traditional FACS 5 ml tubes. Startup, shutdown, and other fluidics maintenance procedures are largely automated.
- **ACEA NovoCyte 3000 Flow Cytometer** is equipped with three lasers (405nm, 488nm and 640nm) with multiple bandpass filters for analysis of up to 13 parameters. The NovoCyte 3000 is also equipped with the NovoSampler Pro, an autosampler option compatible with standard 5ml sample tubes or 24/96 well plates for high throughput analysis.
- **BD FACSAria Fusion** is equipped for state-of-the-art cell sorting with biosafety expertise, thereby allowing analysis of cells exposed to BSL2 virus, bacteria or fungi. The FACSAria Fusion uses 4 lasers allowing for a total of 11 simultaneous colors, and the Temperature Control system allowing for cooling of the sort chamber and collection device.
- Further information regarding all services offered can be found at the OBC page on the CFCCC website or [https://sites.uci.edu/ififlowcore/](https://sites.uci.edu/ififlowcore/).