**Major Equipment:**

Microscopy and imaging systems are available for use in the AIM facilities. In room A522 of the Medical School Research Wing there are two inverted laser scanning Confocal microscope with non-linear optics for advanced imaging, one upright brightfield and epifluorescence systems for stereology, and one upright brightfield and epifluorescence for routine checkup and imaging. In addition, users have access to other basic upright and dissecting stereomicroscope for sample viewing or preparation and two dedicated offline analysis workstations.

**Microscopy systems**

**Confocal Station 1.**The core has a Zeiss Axio Observer Z1 inverted LSM 710 NLO laser scanning confocal imaging workstation that provides a wide variety of imaging technologies including multiphoton excitation and spectral analysis. The system has a Violet HeNe (405 nm); Argon Multiline laser (458, 488, 514 nm); Green DPSS laser (561 nm); Orange HeNe (594 nm); Red HeNe laser (633 nm); and Coherent Chameleon Vision II Ti:sapphire laser (680-1080 nm) for multiphoton spectroscopy as well as an X-Cite metal halide lamp for epifluorescence. The system has a 34 channel QUASAR multispectral scanning module, a transmitted-light PMT detector, and 2-non-descanned detectors. For epifluorescence work, the system has a Zeiss HRm CCD camera, which provides high resolution monochrome image capture, low light fluorescence imaging, and time-lapse imaging. The microscope is equipped with a motorized stage suitable for generating high resolutions mosaic images from large preparations and a fast peizo motorizer for rapid Z-axis acquisition. The objectives include a EC Plan NEO 5X 0.3 NA; EC Plan NEO 10X 0.3 NA; Plan-APO 20X 0.8 NA; LD A-plan 40x; Plan APO 63X 1.4 NA Oil; Plan APO 100X 1.4 NA Oil; C-Plan APO 10X 0.8 NA W; C-Plan APO 63X 1.2 NA W.

Cell electrophysiology. The system has a patch voltage-clamp subsystem (HEKA EPC10 double) and a Patchstar double manipulator.

Vessel studies. There is a Danish Myo Technology pressure (CP120) and wire (CW120) myographs, with an AD Instruments powerlab data logging system and a Dell Dimension workstation.

Live-cell imaging. There is a PECON heating and environmental control system and an 8 channel Automate Scientific programmable perfusion system with Teflon valves.

Multiphoton imaging (“upright” configuration and live animal studies). The Zeiss 710 has an LSM Tech Objective Inverter, stage platform, a Harvard apparatus stereotaxic unit for mice and rats, and equipment for whole-animal anesthesia and ventilation. The inverter system has an Apochromatically corrected DIC 40X W Plan-Apochromat with an NA of 1.0 - an immersion objective designed for electrophysiology recordings on an upright microscopy workstation.

**Confocal Station 2.** Olympus FV1000 Laser Scanning Confocal Microscope with multiple laser lines and objectives. The system has a Violet (405 nm); Argon Multiline laser (458, 488, 515 nm); Green laser (561 nm); and Red laser (633 nm); as well as a mercury arc-lamp for epifluorescence. The system has two PMT detectors with adjustable spectral detection (CHS1 and CHS2) and two additional PMTs with fixed wavelength detection (CH3 - BA585-615 or BA585-685 or BA 655-755; CH4 - BA 655-755) as well as a transmitted-light PMT detector (TD1). The dichroic mirrors are BS20/80; DM405/488; DM405/488/561/633; DM458/515; DM405-440/515; DM405/488/559-561. The objectives are UPLFLN 4X NA 0.13; UPLSAPO 10X NA 0.40; UPLSAPO 20X NA 0.75; UPLAPO 40X OI NA 1.0; PLAPON 60X O NA 1.42; UPLFLN 40X O 1.30.

**Stereologer Station 3.** The core has a Zeiss Axio Imager A1 upright microscope with multiple objectives (4X, 10X, 20X, 40X, 100X), and an HBO lamp source. For fluorescence, the microscope is equipped with Zeiss filter set 17 (excitation: BP 485/20nm, emission: BP 515-565nm), filter set 20 (excitation: BP 546/12nm, emission: BP 575-640nm), filter set 49 (excitation: G 365nm, emission: BP 445/50nm), and filter set 50 (excitation: BP 640/30nm, emission: BP 690/50nm). Additionally, the microscope is equipped with a motorized X-Y-Z stage (Applied Scientific) Three Axis Stage Controller for use with closed loop D.C. Servo motor stages, and Z-axis drive. The microscope is fitted with a Firewire 800Mbps Digital Color Camera System. 2448(H) x 2048(V) display and 3.45 x 3.45 (um) pixel size and 5.0MP pixels for superior resolution and *Stereologer* software for state-of-the-art unbiased cell/object counting, branching, and volume metrics. FIJI (ImageJ-based) image acquisition software with extensive plug-ins is available for analysis and post-hoc image processing.

**Brightfield & Epifluorescence Station 4.** The core also has a Nikon Optiphot brightfield/ epifluorescence microscope with multiple objectives (10x, 20x, 40x, 100x Oil), and a HBO 100W lamp source. For fluorescence, the microscope is equipped with Nikon filter set DM400 (UV-2a), DM430 (V-1A), DM510 (BA 580), and DM580 (G-1A, BA 580). The microscope only has the option to do double stain imaging. For image capturing, the microscope is fitted with a Spot Pursuit 4MP digital camera that is used together with Spot 5.1 Advanced digital imaging software. Image Pro Plus 6.3 software for image analysis of fluorescent-labeled samples is also available.

**Stereomicroscope Station 5.** The core facility has a Zeiss stereomicroscope and halogen light source for tissue and imaging chamber preparation.

**Offline analysis systems.** The core facility has three offline analysis systems. One is equipped with a Dell Dimension Computer and fully functional copy of Zeiss Zen software, and Image J. In addition, the facility has a second copy of Image Pro plus 6.3 on a dedicated analysis computer.

**Core Services:**

• Training of faculty, students, and personnel on various microscope systems and image analysis software.

• Research applications for image acquisition and photomicroscopy.

• Research applications for Confocal LSM image analysis with Zeiss Zen software.

• Other image analysis capabilities with MediaCybernetics Image Pro Plus 6.3, and NIH Image J.

• Referral for sample preparation, staining, and immunohistochemistry processing.