

Imaging Shared Resource Reference Guide 28 September, 2016

Location: Suite 287 (West Building)

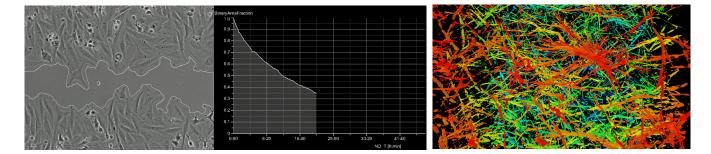
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OVERVIEW

The Imaging Shared Resource is a core facility with the primary goal of providing exceptional microscopy and imaging services, as well as individual access to a variety of state-of-the-art imaging resources for Wistar users and members of the local research community. The imaging systems have been designed to be extremely flexible in order to reflect a broad range of challenging scientific questions and specimens. These systems allow researchers to determine how the temporal and spatial organization of regulatory events within cells, tissues and organisms impact both normal and pathological processes.

CONFOCAL EQUIPMENT

Current major equipment includes both confocal and widefield systems, including our Leica TCS SP8 X white light confocal system with stage-top environmental control (including hypoxia), our TCS SP8 MP intravital 2-photon microscope built on a fixed-stage upright system, as well as a Leica TCS SP5 II laser scanning confocal microscope with 9 laser lines, AOBS and an environmental chamber. All confocals include resonant scanning and sensitive HyD detectors and provide excellent spectral separation.

WIDEFIELD AND OTHER SYSTEMS

Widefield systems include Nikon 80i and E600 upright and TE2000 inverted fluorescence microscopes equipped for color and monochrome image capture, Nikon SMZ1500 and 800 stereomicroscopes, and a customized live-cell time lapse system based on a Nikon TE300 inverted microscope running Nikon Elements AR software. This automated system is also used to capture large stitched images of full slides for analysis of large samples at high magnifications. The Facility also has a Perkin Elmer IVIS 200 small animal, whole body luminescence and fluorescence imager, special low magnification (photomacrography) systems, as well as a variety of traditional photographic cameras, lenses and lighting equipment. In addition, the Facility collaborates with the Flow Cytometry Facility to provide service with an Amnis ImageStream imaging flow cytometer, and with the Molecular Screening Facility to support a Perkin-Elmer Operetta high-content screening and imaging system.

TRAINING, ASSISTANCE & SERVICES

Users of the Imaging Facility may be trained for unassisted use of all core assets, or they may take advantage of the expertise of the facility staff to request assisted service with the staff performing the imaging and providing analysis instead. The Imaging staff also provides assistance to researchers with other aspects of their experimental design: ideal approaches to specimen documentation are often unique to the experiment, and the staff can help determine the most effective imaging protocols to answer a particular question.

Imaging Facility staff are also available to help investigators get the most out of their own microscopes. Training and basic maintenance (including cleaning, alignment and changing bulbs) are available on request. Customized image analysis and Photoshop training, creative imaging for journal covers, and guidance on digital imaging ethics help to round out the services available from the Facility.

Current Equipment Configurations:

Widefield Microscopy - Conventional microscopy utilizing standard techniques in brightfield, darkfield, fluorescence, phase contrast and differential interference contrast is available on a variety of instruments in the main Core Facility suite 287. Upright, inverted and stereomicroscopes capable of low to high magnification documentation are available with individual image capture workstations networked to the Wistar servers.

Nikon 80i Upright Microscope

Location: Suite 287 West

Objectives:

2X, 4X, 10X, 20X, 40X, 60X 100X Oil

Illumination options:

Brightfield, Darkfield, Fluorescence

Fluorescence Filter Cubes:

DAPI, GFP/FITC, TRITC/DS Red, Texas Red and CY5 filters **Image Capture**:

• QImaging Aqua highly sensitive monochrome digital camera (1360 x 1036) with ImagePro Plus software for best fluorescence imaging. The camera works with the same interface as the QImaging cameras on the other microscopes.



• Nikon Ri1 1-chip color camera (1280 x 1024 to 4076 x 3116) with Nikon Elements D software.

Workstation

Custom PC Core i7 with 1TB hard drive, USB2, Firewire, MultiCD/DVD reader/writer running ImagePro Plus ver 7.0 and Nikon Elements D.

Nikon E600 Upright Microscope:

Location: Suite 287 West

Objectives:

4X, 10X, 20X, 40X and 100X oil (2X and 60X available) **Illumination options**:

Brightfield, Darkfield, Fluorescence, DIC, Polarization

Fluorescence Filter Cubes:

DAPI, CFP, GFP/FITC, YFP, TRITC, Texas Red, CY5, Triple Cube (DAPI-FITC-RFP available for viewing only)

Image Capture:

- QImaging Evolution QEi highly sensitive monochrome digital camera (1360 x 1036) with ImagePro Plus software for best fluorescence imaging. The camera works with the same interface as the QImaging cameras on the other microscopes.
- Nikon Ri1 1-chip color camera (1280 x 1024 to 4076 x 3116) with Nikon Elements D software

Workstation

Dell Dimension 9200 Core 2 Duo with 500GB hard drive, USB2, Multi-card reader, MultiCD/DVD reader/writer running ImagePro Plus ver 7.0 and Nikon Elements D



Zeiss Axioskop 2 Upright Microscope (backup for Brightfield and DIC):

Location: Suite 287 West

Objectives:

2.5X, 5X, 10X, 20X, 40X

Illumination options: Brightfield, DIC, Fluorescence

Fluorescence Filter Cubes: DAPI, GFP/FITC, TRITC

Image Capture:

Multiple options including Nikon Ri1, Micropublisher 5 and QImaging Evolution QEi as available

Workstation: Multiple back-up computers as available (PCs)



Nikon TE2000 Inverted Microscope

Location: Suite 287 West

Objectives:

4X, 10X, 20X, 40X (all Long Working Distance) 2X (non-phase), 60X (non-phase, close working distance) and 100X oil (non-phase) available

Illumination options:

Brightfield, Phase Contrast, Oblique, Fluorescence

Fluorescence Filter Cubes:

DAPI, CFP, FITC, TRITC, Texas Red, CY5

Image Capture:

- QImaging Blue EXi highly sensitive monochrome digital camera (1360 x 1036) with ImagePro Plus software for best fluorescence imaging. The camera works with the same interface as the QImaging cameras on the other microscopes.
- Nikon Ri1 1-chip color camera (1280 x 1024 to 4076 x 3116) with Nikon Elements D software

Workstation

HP Pro Core i5 with 500GB hard drive, USB2, Firewire, MultiCD/DVD reader/writer running ImagePro Plus ver 7.0 and Nikon Elements D.



Nikon TE300 Inverted Microscope (Live-Cell Time-Lapse System):

Location: Suite 287 West

This widefield time-lapse system provides 2D imaging in brightfield, phase contrast and multiple fluorescence channels for long-term experiments captured at multiple sites. It is commonly used for 24-48 hour scratch assays performed in 6-well plates, but experiments lasting as long as 5 days to follow generational cycling of cell cultures have also been done. In addition, we can take advantage of the system automation to capture large fields of view of culture plates or histology slides at high magnifications using automated stitching algorithms.



Custom macros can be developed to automatically analyze the results of these experiments as well. Use of the TE300 is carried out in close association with Fred Keeney.

Objectives:

4X, 10X, 20X, 40X (all Long Working Distance) 2X (non-phase), 60X (non-phase, close working distance) and 100X oil (non-phase) are available

Illumination options:

Brightfield, Darkfield, Phase, Oblique, Fluorescence

Fluorescence Filter Cubes:

DAPI, FITC, TRITC, CFP, YFP, Triple Cube (DAPI-FITC-RFP available for viewing only)

Fluorescence Filter Wheel:

DAPI, Pacific Blue, CFP, GFP, TRITC, CY5

Motorized XY Stage

Multiple inserts for well plates, 35mm dishes, standard glass slides. Positions can be saved for return sessions. **Z-axis controller** with encoders for fixed or automatic focusing is included.

Environmental Chamber

Temperature and CO2 control surrounding entire microscope

Image Capture:

- Q-Imaging Retiga EX digital camera, highly sensitivity monochrome camera (1360 x 1036 pixel resolution)
- Nikon Ri1 1-chip color camera (1280 x 1024 to 4076 x 3116) with Nikon Elements AR

Workstation

HP Z420 Workstation with XEON ES-1620 processors @ 3.70 GHz, 1TB and 3TB SCSI hard drives, 32GB RAM, NVIDIA Quadro K600, USB 2 and 3 and Firewire, MultiCD/DVD reader/writer, and Dual 19" Flat Screens running Nikon Elements AR

Additional

Automatic fluorescence and brightfield shutter

Time-Lapse Capabilities

5D imaging (X, Y, time, wavelength, positions)

Nikon SMZ800 and 1500 Stereo Microscopes:

Location: Suite 287 West

Low magnification acquisition is generally done with stereomicroscopes because the long working distance provides room for both specimens and multiple lighting options. Our systems can be used with transmitted, reflected or fluorescence light and a variety of cameras can be mounted, depending on need. Larger subjects also create challenges with shallow focus, but software with extended focus options is available on our workstations.

Magnification Range:

- 1X objective with 1X 6.3X variable zoom (SMZ800)
- 1.5X objective with 0.75 11.25X variable zoom (SMZ1500)

Illumination options:

Brightfield, Darkfield, Polarization, Oblique Illumination, Episcopic Lighting, Fiber optic light guides, Fluorescence (SMZ1500)

Fluorescence:

- GFP, RFP filter cubes in SMZ 1500
- GFP possible with Illumatool 9900 system

Image Capture:

Nikon Ri1 1-chip color camera (1280 x 1024 to 4076 x 3116) with Nikon Elements D software

Workstation

Dell Dimension 9150 Pentium D with 500GB hard drive, USB2, Firewire, Multi-card reader, MultiCD/DVD reader/writer running ImagePro Plus ver 7.0 and Nikon Elements D



Confocal Microscopy – Two Leica laser scanning systems, including an inverted TCS SP5 II, and an inverted TCS SP8 X white-light confocal are available for high-resolution single cell observations, thick specimen analysis, co-localization studies, 3D timelapse and live, fast imaging. Resonant scanning, sensitive HyD detectors and spectral detection are standard, making these systems ideal for live, weak specimens. FRET and FRAP acquisition and analysis is also built into each system. Analysis of the captured images can be done with the Leica analysis package or with other 3rd party options available in the Core. Primary assistance with confocal systems is provided by James Hayden.

Leica TCS SP8 X WLL Scanning Confocal Microscope:

Location: Suite 287 West

This instrument is ideally constructed to support a wide variety of live-cell imaging projects. The white-light laser, combined with AOBS and HyD detectors, provides optimum excitation and detection efficiency for most fluorescent proteins. The resonant scanner captures rapid biological events and reduces specimen exposure and phototoxicity. The Automatic Focus Control ensures the selected focus plane does not drift, and the LightGate feature of the white light laser eliminates reflections from the support glass – perfect for imaging focal adhesions. The Stage-top incubation chamber supports long-term standard and hypoxia experiments.

Microscope:

Leica DMi8 CS Inverted system with AFC (Automatic Focus Control) to maintain Z positioning

Objectives:

10X dry, 20X oil, 40X oil, 63X oil (40X water, 63X glycerine, and 100X oil available)

Illumination options:

Brightfield, Fluorescence, DIC

Fluorescence Filter Cubes (for visible inspection):

DAPI, CFP, FITC, YFP, Rhod long pass,

Fluorescence Excitation Wavelengths:

405nm and a continuous white-light laser tunable from 470 - 670nm in 1nm increments with up to 8 lines used simultaneously. Also allows for gated imaging to eliminate interference from reflected light.

AOBS (Acousto-optical Beam Splitter)

Tandem (Resonant) Scanner

Up to 8,000 lines per second

Image Capture:

- (3) Leica PMT detectors
- (2) HyD detectors
- TLD Transmitted Light Detector

Environmental Chamber:

Tokai-Hit Stage Top Incubation System with GM-8000 Hypoxia Gas Mixer



Leica TCS SP5 II Scanning Confocal Microscope:

Location: Suite 287 West

Microscope:

Leica DMI6000 Inverted system

Objectives:

10X dry, 20X long working distance, 20X oil, 40X oil, 40X water, 63X oil, 63X glycerine, 100X oil

Illumination options:

Brightfield, Fluorescence, DIC

Fluorescence Filter Cubes (for visible inspection): DAPI, FITC, TRITC (all long pass)

Fluorescence Excitation Wavelengths:

405nm, 458nm, 476nm, 488nm, 496nm, 514nm, 561nm, 594nm, 633nm

AOBS (Acousto-optical Beam Splitter)

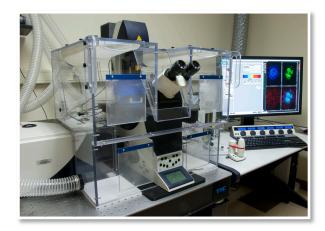
Tandem (Resonant) Scanner

Up to 8,000 lines/sec

Image Capture:

- (3) Leica PMT detectors
- (2) HyD detectors
- TLD Transmitted Light Detector

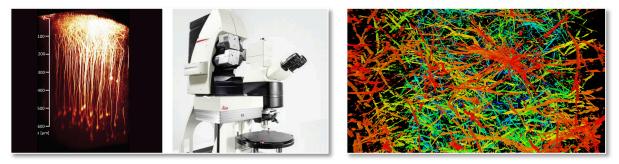
Environmental Chamber with temperature and CO2 control



2 Photon Microscopy – 2 photon microscopy continues where traditional confocal leaves off. With laser excitations in the longer wavelengths, 2P allows deeper imaging with less phototoxicity. This system is designed for *in vivo* studies, such as imaging directly into inguinal lymph nodes, or through cranial windows while the subject animal is anesthetized. It is also used with explanted specimens such as lymph nodes and brain slices, kept viable in heated media. Thick, fixed specimens, such as spheroids and skin reconstructs also benefit from the increased penetration and imaging capabilities of this system.

Leica TCS SP8 MP Spectral 2-Photon Intravital Microscope:

Location: Room 281



Microscope:

Fixed stage, upright system based on Leica DM6000

Objectives:

25X/1.0 Water dipping with motorized correction collar, (10X/.40 dry available)

Illumination options:

Brightfield, Fluorescence

Fluorescence Filter Cubes (for visible inspection):

DAPI, FITC, TRITC, CFP /YFP

Laser:

- Laser Blue 488nm
- Coherent Chameleon XR Ti:Sapphire laser, 710-1040nm wavelengths (variable)

Tandem (Resonant) Scanner

Up to 8,000 lines/sec

Image Capture:

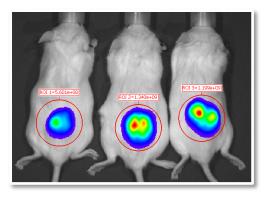
- 1 HyD-RLD 2 channel NDD detector
- 1 RLD 2 channel PMT detector
- 1 TLD 1 channel PMT detector

Time Lapse Capabilities:

5D imaging (X, Y, Z, time, wavelength)

IVIS 200 – Whole Body Imager – Location: Room HR4, Animal Facility

The IVIS 200 whole body imaging system is a quantitative instrument that measures luminescence and fluorescence emissions in a variety of samples. It is primarily used for detection and quantitation of tumor growth in longitudinal studies of live mice, which are maintained inside the barrier facility in room HR4 of the Animal Facility, where the instrument resides*. The IVIS is capable of imaging 1-5 mice at a time and includes anesthesia management inside the dark box. In addition to the standard luminescence imaging most commonly used, the system is capable of approximating structured 3D measurements (non-



quantitative) to determine sample depth in the tissue, as well as a range of fluorescent wavelengths to standardize and quantify GFP, RFP and Cy5.5 fluorescence. Primary assistance with the IVIS is provided by Laila Pöché.

* Animals being used with the IVIS imager must be housed in Rm HR4 with the instrument

Magnification Range / Field of View:

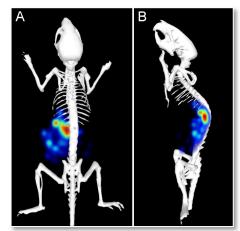
3.9 x 3.9cm, 6.5 x 6.5cm, 13 x 13cm, 19.5 x 19.5cm, 26 x26cm (close-up of half a mouse – 5 mice at once)

Objective:

f/1 – f/8 1.5x, 2.5x, 5x, 7.5x, 10X

Imaging Modes:

- White light Photograph
- Luminescence
- Fluorescence
 - o 400 900nm



Analysis:

Allalysis:	
Planar Spectral Image	Determines the average depth and total photon flux of a luminescent point source in a user specified region of interest. Analyzes a sequence of luminescent images acquired using different emission filters.
Display multiple fluorescent or luminescent reporters	Uses the Image Overlay function to display multiple luminescent or fluorescent images on one photographic image.
Subtract tissue auto fluorescence using blue shifted background filters	Uses the image math feature to subtract a background image from the primary image.
Point Source Fitting	Estimates the optical properties of tissue, the location and power of a point source, or the fluorescent yield of fluorophores.
Spectral unmixing	Extracts the signal of one or more fluorophores from the tissue autofluorescence. Distinguishes the spectral signatures of different fluorescent or luminescent reporters when more than one reporter is used in the same animal model.

Macro Imaging Systems



Traditional Photographic Imaging - Location: Suite 287 West

The macro imaging system is comprised of elements that replace traditional 35mm photography equipment. The heart of the system is the Nikon D200 DSLR that can capture up to 12 megapixels. With the associated macro hardware such as bellows and lighting equipment, and the standard and macro lenses we have, this system can be used for any subject from a group shot of 100 people, to a 10-day old mouse embryo. Flexible lighting options with a Bencher copy stand and external lights allow us to photograph anything that may have been photographed conventionally in the past. We regularly photograph gels and Western blots, Petri dishes, transwell membranes and well plates to quantify cell colony growth patterns, excised mouse lungs, lymph nodes and other small wet specimens, as well as live mice and even portraits of people.

Imaging of large, GFP-tagged tissues is available with the Illumatool illuminator. We have used this to image specimens like mouse lungs with GFP-tagged metastases and whole GFP mouse embryos, pups and adults, as well as whole adult mouse legs and sectioned sheep hearts.

Magnification Range:

Infinity – 20X (depending on lenses and bellows extension)

Lenses:

- 60mm Micro-Nikkor, 105mm Micro-Nikkor, f1.2 50mm Nikkor
- Luminar lenses 25mm, 40mm, 63mm and 100mm (on loan)
- 18-135mm Nikkor Zoom
- a variety of reversed lenses including 28mm, 50mm and 90mm (on loan)

Illumination:

- Epi and diascopic fiber optics
- Brightfield transillumination (flat panel viewer)
- Illumatool fiber optic fluorescence illuminator for large GFP specimens like whole mice
- Bencher Copy Stand with integrated tungsten lights
- Calument Travelite 750 studio strobes

Image Capture:

• Single-shot color imaging with Nikon D200 – up to 4000 x 3000 RGB pixel resolution

Microscopy Workstations

Location: Suite 287 West

In addition to the image capture options available in the facility, we also have several standalone workstations where additional processing can be done without tying up the instruments. Advanced Image Analysis, including 2D time-lapse movies, Co-localization studies, FRAP analysis and other customized procedures can be performed on client experiments, or we can work with users to teach them how to do it themselves.

Images analysis with Nikon AR, Leica LAS X and ImagePro ver. 7.0 allows users to quantify measurements in automatic, semi-automatic or fully manual modes. The 3D modules provide processing of image stacks from the 2 Photon microscope and confocal systems. Cells in live cell movies can be tracked, areas and perimeters can be measured and intensity measurements (with the correct acquisition techniques) can be determined. Preparation of publication figures can also be done here.

Analysis Workstations

Leica Workstation:

High performance HPZ6840 Workstation with 2X 4-Core Xeon E5-267 V3 3.5GHz processor, NVIDEA Quadro K4200 4GB GPU, 16GB RAM, 128GB SATA SSD, 256GB SATA SSD, 2x 1TB SSD RAID 0, 5x 3TB SATA hard disc drive RAID5 and 2TB SATA hard disk drives, Firewire B and USB3 interfaces, Blu-ray Writer.

3D Workstation:

Dell Precision T3600 workstation with XEON(R) CPU E5-2665 0, AMD FirePro 5900 (FireGL V), 16GB RAM, 1TB SCSI Disk Drive, CD and DVD read/write capability, USB2 and 3 interfaces.

Imaging Workstation:

Dell Pentium Core 2 Duo workstation with 2-4GB RAM, 120-400GB hard drives, CD and DVD read/write capability, USB2, Firewire.

Software

- Nikon Elements AR
- ImagePro Plus 7.0 and ImagePro 3D
- Leica LAS X software with 3D module
- Huygens Deconvolution Software (bundled with Leica LAS X)
- NIH Image J and FIJI (FIJI is just Image J with all the macros built in)
- Live Image software for IVIS
- Helicon Focus
- Volocity 4.3
- Adobe Creative Suite 5
 - Photoshop
 - o InDesign
 - o GoLive
 - o Illustrator
- Microsoft Office Suite

Printers

HP MFP M477 Color Laser Printer (draft quality)

Facility Services

All services of the Imaging Shared Resource are coordinated through the main laboratory in suite 287, although some equipment is placed in remote locations. The Imaging Facility staff can provide all listed services or researchers may be authorized to work with the equipment unassisted. Interested users must be trained on any equipment they wish to use before unassisted operation of the equipment is permitted.

We provide:

- **TRAINING** on all Facility equipment and software
- **STAFF IMAGING** with Facility resources (we do the work for you)
- **ON LOCATION** microscope servicing and assistance in your laboratory
- **CONSULTATION** on experimental protocols, imaging applications and aesthetic considerations (i.e. for journal covers)

Additional Services

Live-Cell Time-Lapse Microscopy – Using the Nikon TE300 inverted microscope in suite 287 with its Nikon Elements AR workstation, we can run multiple time-lapse studies of cells grown in culture. With the incubation chamber, XY stage and Z axis controller, we can track multiple areas in separate wells at the same time in multiple fluorescent channels. Stable imaging for up to 72 hours can be accommodated on this system with longer sequences possible. Cells may be grown in 6 well plates or 35mm dishes standard, but we can accommodate other vessels as needed.

Live-cell imaging is also available with the SP5 and SP8 confocal systems. These systems are higher resolution and allow true 3D (Z-stack) capture in multiple channels, and multiple locations (6D imaging). Experiments can be run using our new **hypoxia** chamber system, which allows 3-gas control of environmental conditions (CO^2 , O^2 and N^2)

Whole Sample Imaging

Using the TE300 live cell system, with XY stage, we can also scan large areas of entire slides and provide high-resolution files using the automated system. This is primarily for brightfield histology slides, but fluorescence imaging is also possible. Up to 4 slides can be set at a time for unattended imaging, automated image stitching and semi-automated analysis. Other sizes of sample containers can be accommodated as well for customized imaging.

Image Post Production and Analysis - Image capture is just the first part of acquiring high quality images. Whether taken with Core equipment or other laboratory microscopes, the next step in optimizing images involves image processing and analysis. The facility staff is available to train users both on the intricacies of the image capture software as well as popular third party image processing programs such as Nikon Elements AR, Leica LAS X, ImagePro Plus and 3D, NIH Image J and FIJI and even Adobe Photoshop. We have the ability to **design custom software macros** to help objectively quantify your results and process large data sets. We can run the analysis on your images or show you how to do it yourself.

Macro, Specimen, Small Animal and other Specialty Image Capture - We provide additional expertise in a variety of specialized scientific imaging situations such as photomacrography, small

animal and gross specimen photography, gel and blot documentation, ultraviolet imaging as well as fluorescence in larger subjects, like whole mice. Additionally, creative imaging and graphic design for journal covers and public relations uses can provide proven exposure to the research and to the Institute as a whole. These specialized techniques can be provided as a direct service or for training to those interested in long-term applications. Special set-ups to capture 1-shot images of large histology sections for analysis are available. Our equipment and services are flexible enough to meet almost any need.

Group photography of your lab is also available at your convenience.

Confocal Systems (Wistar Rate) (8 hrs x \$35/hr) + (X hrs x \$17.50/hr)

Training, Individual Assistance &	Wistar	Academic	Commercial
Imaging Support	\$50/hr	\$75/hr	\$150/hr
Staff Service Fee			

Widefield Training – generally 2 hours each for brightfield or fluorescence Confocal Training – generally 3 sessions at 3hrs/each = 9 hrs total IVIS Training – generally 2 sessions at 2hrs/each = 4 hrs total

Fee Schedule as of August 1, 2015

Nikon 80i Upright Microscope

Nikon E600 Upright Microscope

Widefield Microscopy Systems:

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cal and Laser-based Systems:	Wistar	Academic	Commercia
Leica TCS SP5 II Confocal	\$35/hr	\$52.50/hr	\$105/hr
Microscope Leica TCS SP8 X Confocal Micro	scone		
Leica SP8 MP 2-Photon Intravital	-		
Animal Imaging	Wistar	Academic	Commercia
Perkin Elmer IVIS 200	\$90/hr	\$135	\$270
Il SLR-based Systems Nikon D200 Macro System	Wistar	Academic	Commercia
Nikon D200 Studio System	\$20/hr	\$30/hr	\$60/hr
e Analysis Workstations			
High performance HPZ6840	Wistar	Academic	Commercia
Workstation	\$20/hr	\$30/hr	\$60/hr
Dell Precision T3600 Workstation Dell Imaging Workstation	l		
Cell Time Lapse Imaging	8 hrs @ 100% + X hrs @ 50%		

Wistar

\$20/hr

Academic

\$30/hr

Commercial

\$60/hr

Facility Policies

Training Policy

Anyone wanting to use facility equipment unassisted <u>must</u> sign up for training on that instrument before they will be allowed to use it by themselves. Training by someone else in your lab is not acceptable to qualify for unassisted use. Please contact James Hayden (<u>jhayden@wistar.org</u>) or Fred Keeney (<u>fkeeney@wistar.org</u>) to set up training sessions. Training can be done in groups of 2 or 3 as needed.

Sign-up Policy

All Facility equipment can be reserved on the Wistar Core Facility Calendar, providing the user has been authorized on a specified instrument. Once booked, the user is responsible for the time reserved and will be billed starting at the time reserved and ending at the time signed out. If the user cannot make the reserved appointment, it is their responsibility to delete the entry from the calendar to open the time slot for other users. If the time is not deleted, the original user is billed for the reserved time. Entries can be deleted anytime up to 1 minute before the start of the reservation – after that, an invoice is automatically generated starting at the reserved time and continuing until the end of the reservation or until the user logs in to the system and signs out.

Microscopy Server

In most cases, images are acquired and saved directly to folders on the Microscopy Server. Full access to the files (read and write capabilities) is available from any Facility workstation. Limited access (read only) is available over the Wistar network to personal lab computers. The server is backed up and archived on a regular basis through the Bioinformatics system.

As always, our standard policy in regard to all generated data is that <u>it is the ultimate</u> <u>responsibility of the user to back up their information</u>. Although we are endeavoring to maintain a continuous backup of all generated data, we cannot be responsible for the loss of files if the user does not personally back them up as well. Files on the microscopy server are backed and archived daily. As a rule of thumb, we will be able to back-up one year's worth of data on the server at a time, so you can expect that your files will stay on the server about a year before they are removed to make room for new files. The deleted files will continue to be available through the archive. It is important that everyone maintain folders by year, month and day to facilitate finding and deleting older files chronologically.

File Naming for the Microscopy Server

You can expect that files will remain on the server until about May of the following year. At that point, all files from the previous year need to be removed to keep room available for ongoing work. To make this easier, each user should have a "Year" folder inside their folders. Inside that folder, each session should be placed in a folder with the date according to the following format: YearMonthDay, i.e. 2015July12 This will make it much easier to find and purge files as needed at the end of each year.