



SCIENCE FOR THE BENEFIT OF HUMANITY

Introduction to The Rockefeller University Resource Centers

**Overview for New Graduate Fellows
September 19, 2016**



The Role of Core Facilities

- Ensuring and encouraging access to equipment, services and products that are beyond the financial or technical means of most individual laboratories
- Supporting research in a cost effective and scientifically effective manner
- Improving competitiveness for researchers and the University
- Providing continuity of staffed expertise
- Training and education in specialized methods and technologies
- Serving as scientific brokers both internally and externally



SCIENCE FOR THE BENEFIT OF HUMANITY

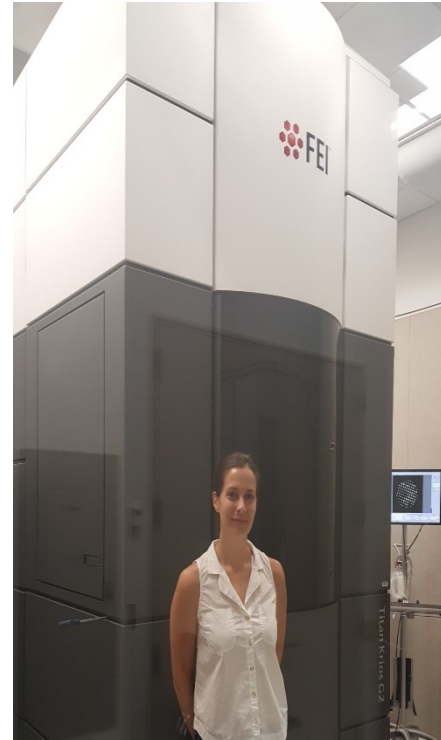
Cryo-Electron Microscopy Resource Center

Microscopy Suite: CRC B13

Phone: 212-327-7282



Mark Ebrahim
Senior Staff Scientist
mebrahim@rockefeller.edu



Johanna Sotiris
Research Support Specialist
jsotiris@rockefeller.edu

Cryo-Electron Microscopy Resource Center

Instruments



FEI Talos Arctica
200kV Transmission Electron
Microscope

1.4 Angstrom Resolution

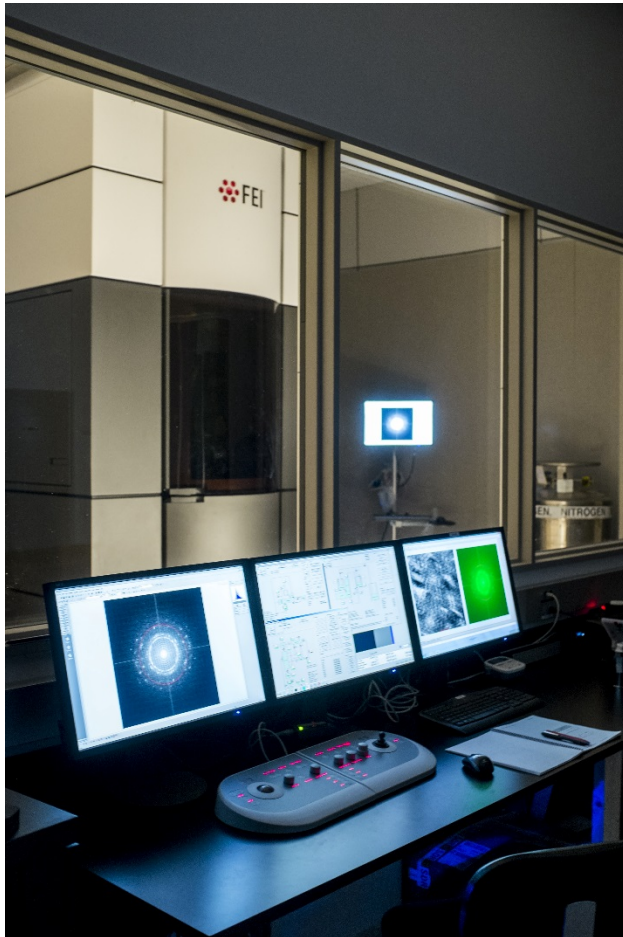
Equipped with Gatan K2
Summit direct electron
detector



SCIENCE FOR THE BENEFIT OF HUMANITY

Cryo-Electron Microscopy Resource Center

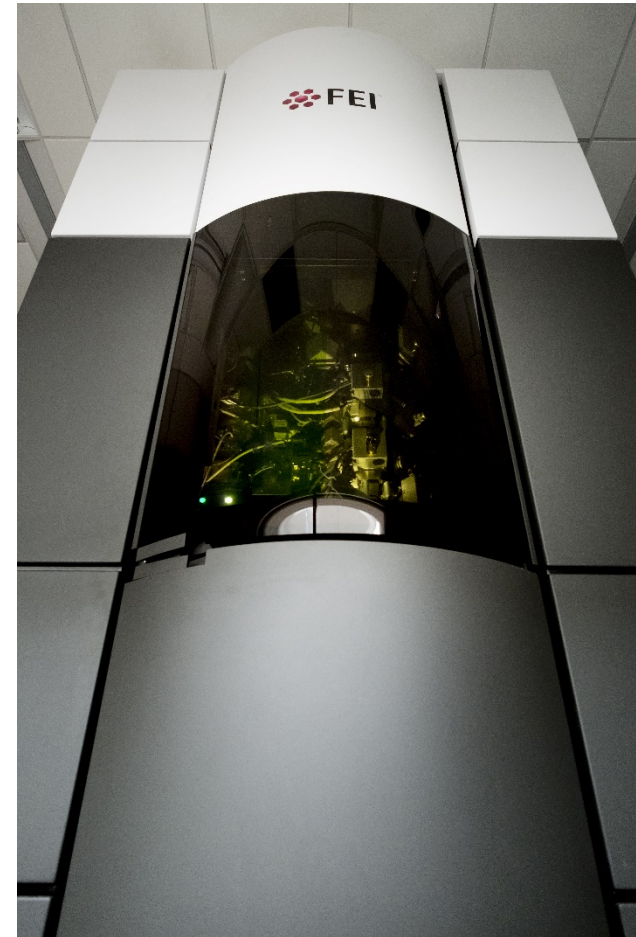
Instruments



FEI Titan Krios
300kV Transmission
Electron Microscope

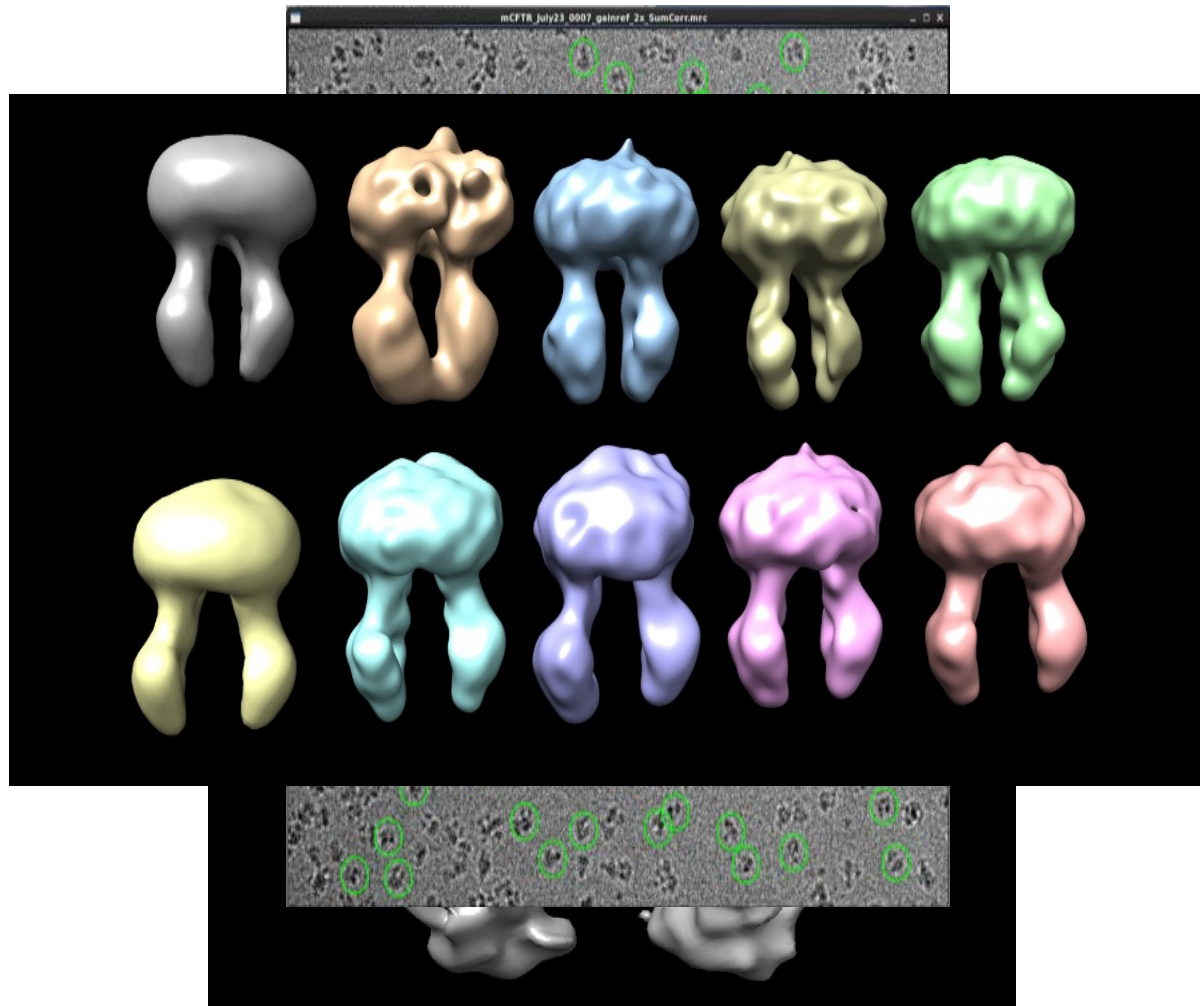
1.0 Angstrom Resolution

Equiped with Gatan K2
Summit direct electron
detector



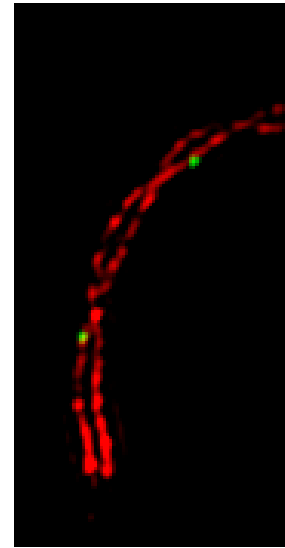
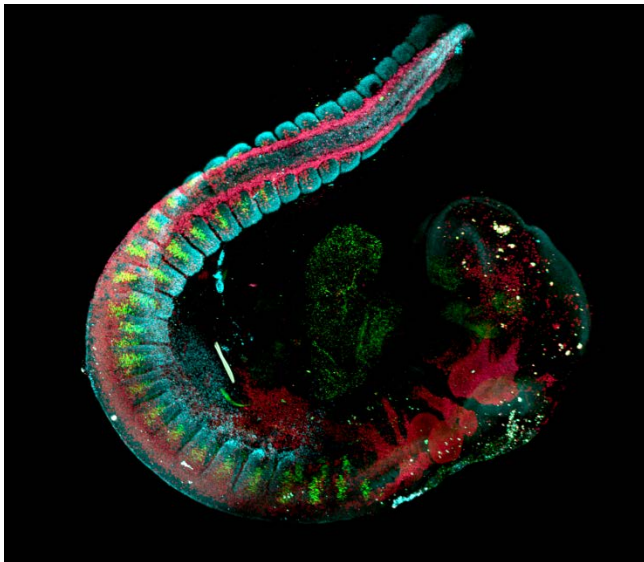
Cryo-Electron Microscopy Resource Center

Applications



Bio-Imaging Resource Center

- Advice on microscopy and specimen preparation
- Training on the microscopes
- Training in image analysis
- Initial consultations and tours
- Possible collaborations



Widefield

Light Sheet

TIRF

Super-resolution

Confocal

Spinning disk confocal

Deconvolution

Multiphoton

FRET

FLIM

FCS

Laser microdissection

Staff of the BIRC

(DWB 201-203)

<http://inside.rockefeller.edu/bioimaging/>



Alison

➤ **Alison North** (Senior Director) - Ph.D. in Cell Biology - any microscope I can get my hands on in between doing the less exciting administrative stuff, plus OMX specialist;

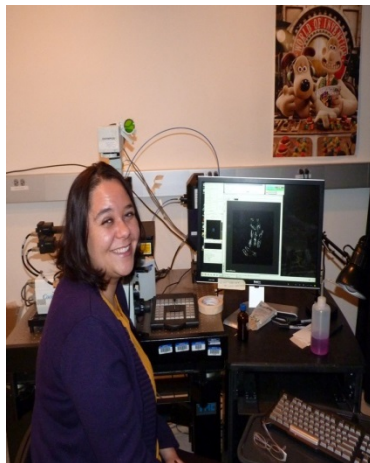
➤ **Kaye Thomas** (Assistant Director) - Ph.D in Immunology – all types of microscopy, STORM specialist;



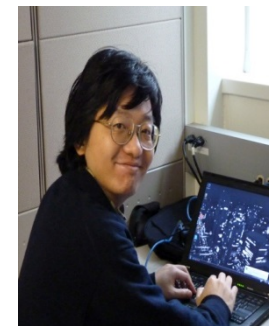
Christina

➤ **Christina Pyrgaki** (Research Support Specialist) - Ph.D in Molecular Biology – all types of microscopy, multiphoton specialist;

➤ **Tao Tong** (Research Support Specialist) - Masters in Computing and Biochemistry/Molecular Biology – systems administration, image analysis, microscopy.



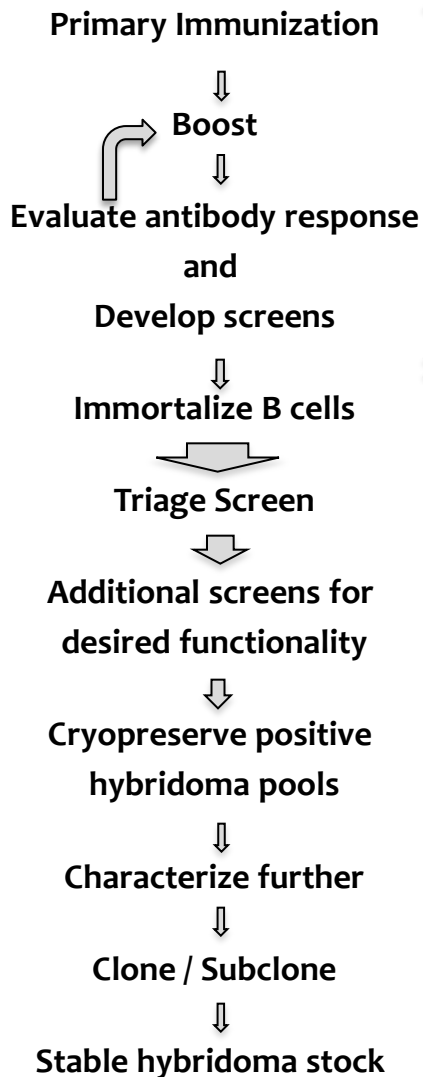
Kaye



Tao

Antibody and Bioresource Core Facility

Custom MAb Development

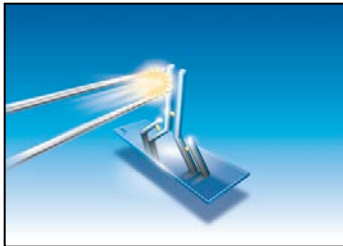
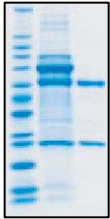
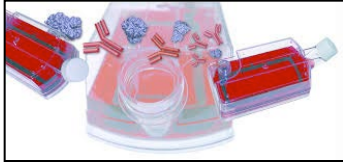


Polyclonal Antibodies
(3 to 12 months)

Create Monoclonal Antibodies
(2 to 3 months)

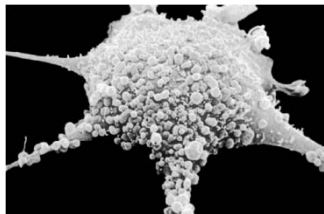
- Comprehensive project design and management
- Generate robust humoral response
 - Immunogens: Cells, protein, peptides, and other haptens
 - Hosts: Mice (WT/KO), hamsters, and rats
- B cell immortalization (hybridomas)
- Maintain and screen thousands of hybridomas by ELISA
- Clone positive hybridoma pools to establish stable lines
- Archive fusions, positive hybridoma pools and clones

Antibody and Bioresource Core Facility



Monoclonal antibodies

- ***In vitro* production**
 - Large scale production in bioreactors
- **Purification**
 - milligrams to grams at > 95% purity
- **Modification – Available for Consultation**
 - Conjugation to such things as Alexa dyes, Pacific Dyes, Qdots, APC, PE, HRP or biotin
 - Fragmentation [FAB, F(Ab')₂ and F(Ab')₂m]



Mycoplasma Testing

Antibody and Bioresource Core Facility



Cell Line Distribution Service

- Alleviates the research lab the work associated with distributing cell lines
- Informational and physical repository for published cell lines
- Hundreds of hybridomas, tumor and lines as well as stem cell lines

Antibody and Bioresource Core Facility

skiabcf@mskcc.org

macfwebext.mskcc.org

Bronk 415

x 7030

415 E 68th Street

ZRC 1553

646-888-2331



Syeda
Rizvi

Lloyd
Bourne

Frances
Weis-Garcia

Laura
Campos

Electron Microscopy Resource Center

Devrim Acehan, Ph.D.
Specialist



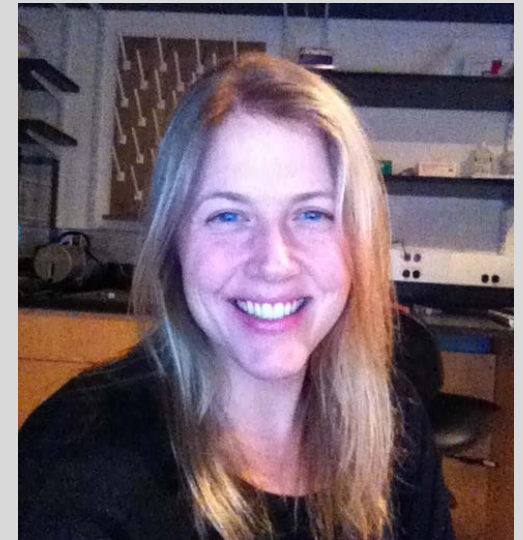
Structural Biologist

Kunihiro Uryu, Ph.D.
Director



Neuropathologist

Nadine Soplop, Ph.D.
Associate

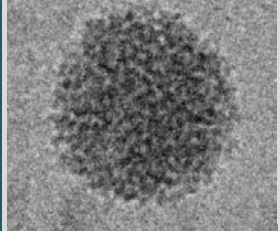


**Drosophila
Embryologist**

EMRC: For Comprehensive Study for Structure and Function

Transmission EM

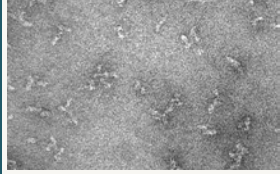
Cryo-EM (HCV)



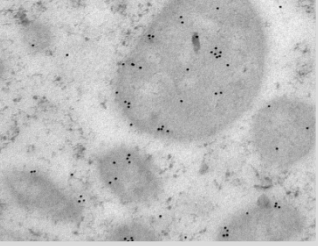
DNA-Protein Complex Spread



Negative stain (Protein Complex)

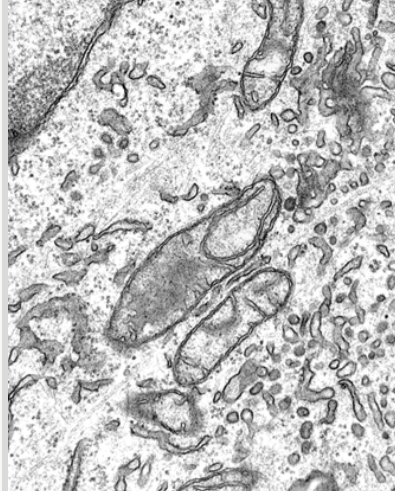


Immuno EM



TEM JEOL JEM1400Plus
Serial-EM and EDS

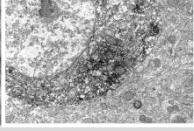
TEM (Thin Section)



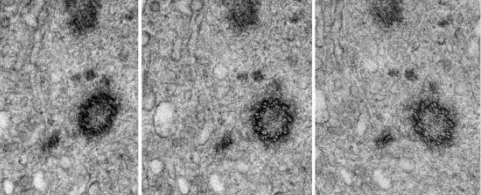
Light Microscopy



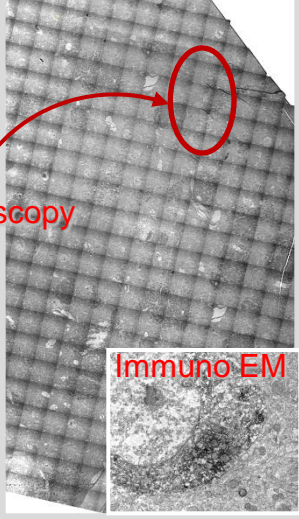
Immuno EM



Tomography

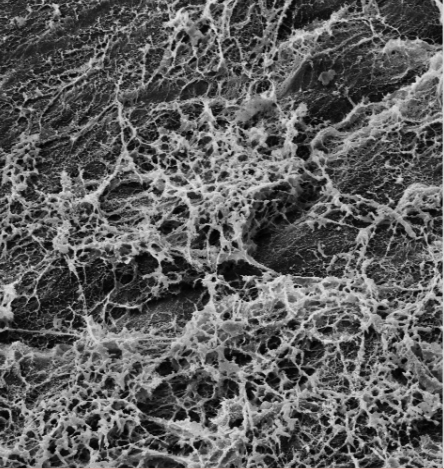


EM Montage

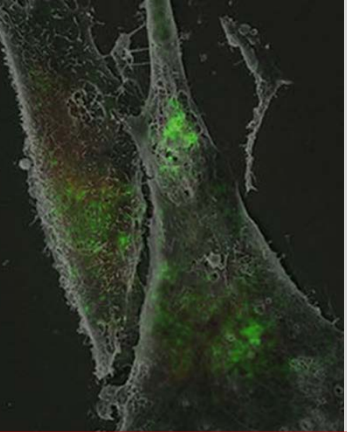


Scanning EM

Artery fibrinolysis

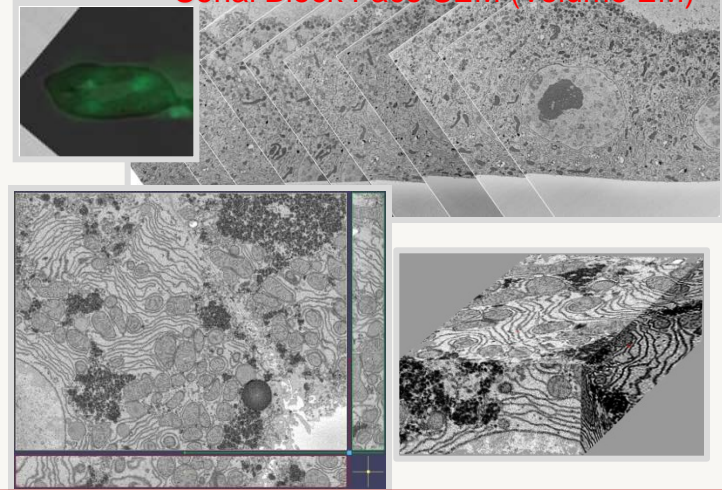


Correlative Light and Electron Microscopy



SEM Carl Zeiss-LEO
Merlin With 3View

Serial Block Face SEM (Volume EM)



Flow Cytometry Resource Center

Svetlana Mazel, Ph.D., RU

Director

DWB 205

<http://inside.rockefeller.edu/fcrc/>



Stanka Semova, M.S.
Senior
Research Support Specialist



Songyan Han, Ph.D.
Research Support Specialist



Selamawit Tadesse, M.S.
Research Support Specialist

FCRC Services

On Equipment

- **Staff-operated equipment at FCRC**
 - Cell sorting on three BD FACSAria (cell sorters)
 - Data acquisition on the ImageStream-X (imaging flow cytometer)
- **All the Flow Cytometry equipment at FCRC**
 - Maintenance
 - Quality control testing
 - Troubleshooting and minor repairs
 - Communication with the vendors for appropriate service
 - Service request
 - Post-service follow-up
 - Data management
- **Equipment and computer/programs upgrades and acquisitions**
 - Proposals
 - Installations with sufficient quality controls

Education and Training

- **“Beyond the Basics” Flow Cytometry class**
 - FCRC Classes in groups of 5-15 people
 - Pre-scheduled approximately one or two months in advance



- **Instrumental training “hands-on”**
 - Personal session scheduled based on request
 - BD LSR-Fortessa
 - BD LSR II
 - HTS
 - BD Accuri®C6
- **Software Training (Introduction or Troubleshooting)**

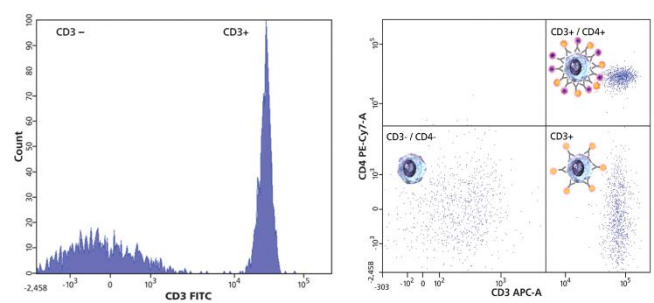
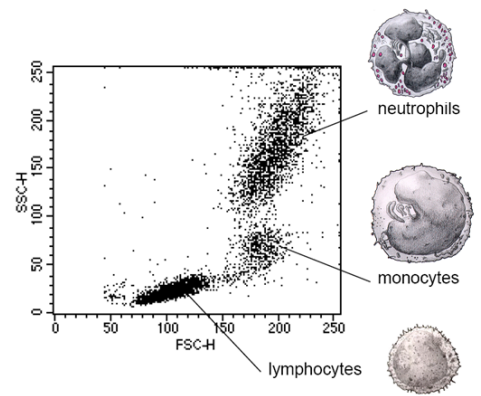
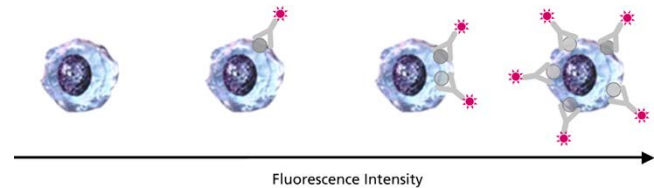
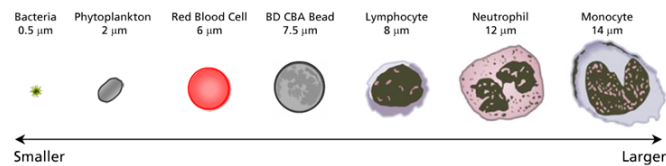
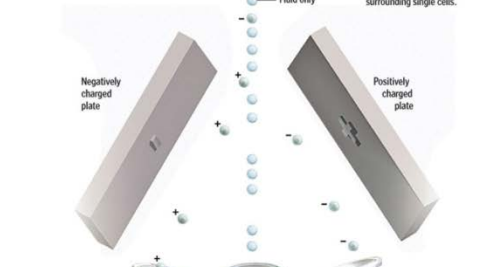
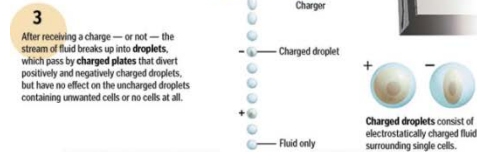
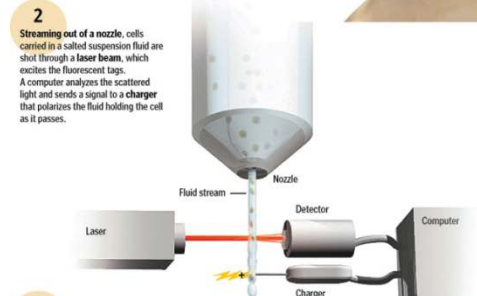


Consultation and Help

- **Consultations**
 - Experimental Design
 - Pre-Sort
 - Pre-ImageStream
 - Pre-Hands-On
- **Instrumental Help**
 - Set up
 - Troubleshooting
- **Troubleshooting on experimental design**
- **Consultation and help with data analysis**
- **Help with data preparation for the publication and scientific presentations**

What Could Be Done at FCRC, the Flow Cytometry Resource Center?

Operation	Extended Business hours, by FCRC Staff				24/7/365, self-operated			
Lasers (colors and wavelength) correlated to # of Detectors	Sorters			Image Analyzer	Advanced Analyzers			Basic Analyzer
	BD FACS Aria-II-1	BD FACS Aria-II-2	BD FACS Aria-II-3	Image Stream-X	BD LSR II-1	BD LSR II-2	BD LSR Fortessa	BD Accuri C6
Blue Laser; 488nm	3	3	3	5	3	3	3	4/3/2
Red Laser; 633-658nm	3	3	3	2	3	3	3	0/1/2
Violet Laser; 405-408nm	3	4	4	5	4	4	5	
Yellow/Green Laser; 561nm	4	4	4	4	4	4	4	
UV Laser; 355nm		2	2		2		3	
Blue/Violet Laser; 455nm			2			2		
Total # of Detectors	13	16	18	10	16	16	18	4



What Could Be Done at FCRC,

Graphical Population Definitions

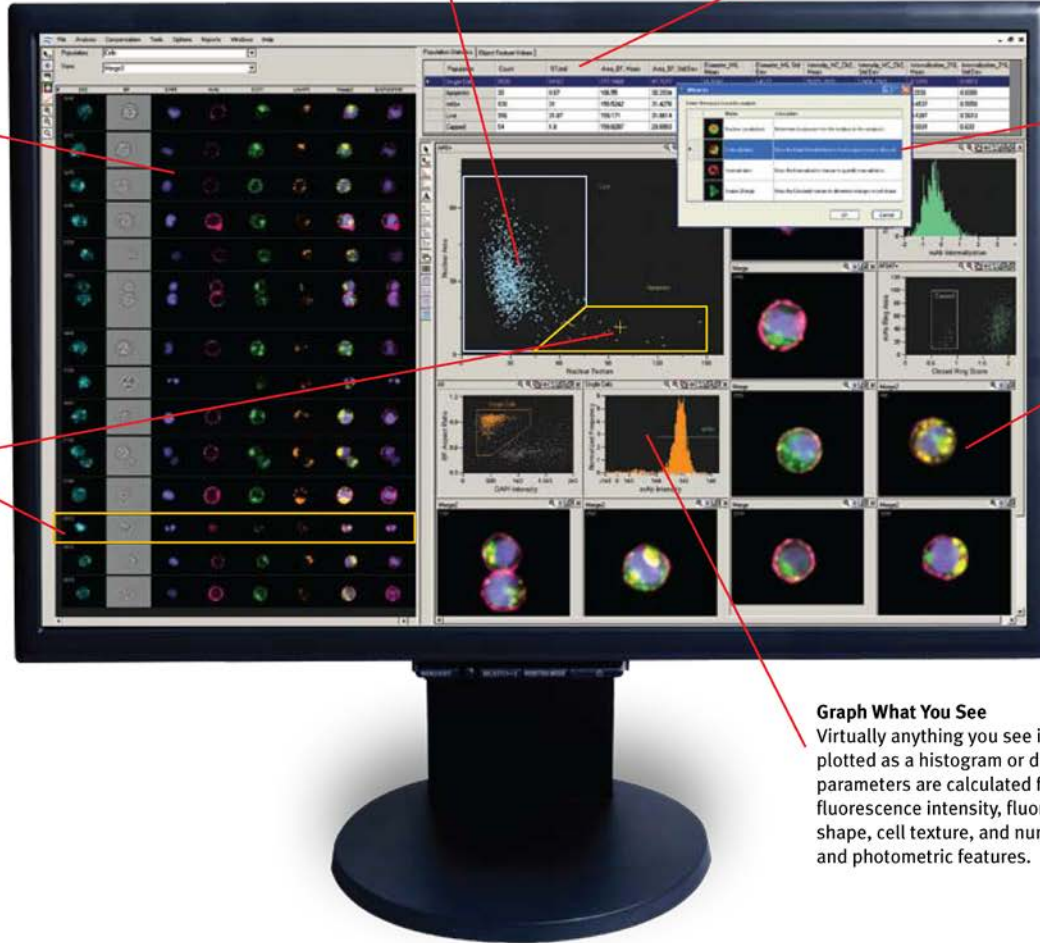
Define populations using familiar graphical tools and combine them with logical functions.

Comprehensive Population Statistics

Characterize your cell populations with a wide range of statistical metrics to reveal differences in cell morphology, phenotype, and function.

Inspect Your Populations

The Image Gallery allows you to see every image of every cell or perform a "virtual cell sort" to inspect and validate the cells within a specific population.



Wizards Simplify Analysis

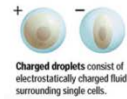
Pre-configured and optimized analysis wizards are provided for many common applications.

Flexible Image Display Tools

Create composite images, pseudo-color representations and a host of other image transformations for reporting and publication.

Graph What You See

Virtually anything you see in the imagery can be plotted as a histogram or dot plot. Hundreds of parameters are calculated for every cell, including fluorescence intensity, fluorescence location, cell shape, cell texture, and numerous other morphologic and photometric features.



(colors are correlated)

Blue Laser;

Red Laser;

Violet Laser;

Yellow/Green Laser;

UV Laser; 3

Blue/Violet

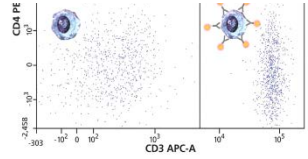
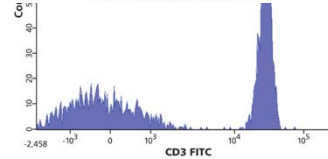
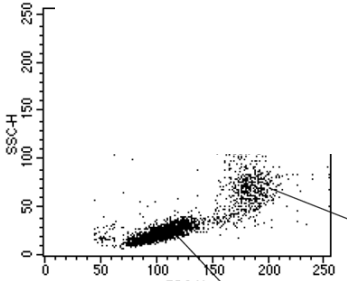
Total

Bacteria 0.5 μm

Phytoplankton 2 μm



Smaller



Services at the Genomics Resource Center

- **Microarray systems**

- Whole genome expression profiling
- Whole genome SNP genotyping
- Custom SNP genotyping (48-1536 plex level)
- Chromosome copy number analysis
- MicroRNA analysis
- DNA methylation



Affymetrix



Agilent



Illumina

- **Next-generation sequencing**

- Whole genome sequencing
- Targeted sequencing
- Transcriptome sequencing (RNA-seq)
- Protein-DNA binding analysis (ChIP-seq)
- Small RNA discovery and quantification
- Methylated DNA



HiSeq 2500



NextSeq 500



MiSeq

- **Realtime PCR systems**

- Individual gene expression analysis
- Pathway analysis
- Individual SNP genotyping



QuantStudio 12K-flex



LightCycler 480



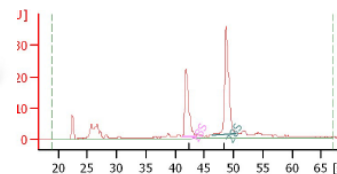
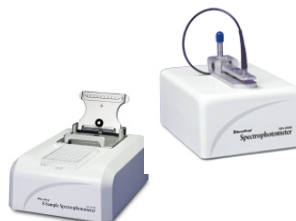
- **Single Cell Analysis**

- Fluidigm C1 Single Cell Auto Prep System



- **Others**

- Covaris Ultrasonicator
- Agilent Bioanalyzer
- Agilent TapeStation
- Qubit fluorometr
- ND 2000 and ND 8000
- PCR machines 96/384-well



Transcriptome Sequencing (RNA-Seq)

- Standard mRNA sequencing
100 ng - 1 ug total RNA
- Low-input mRNA sequencing
100 pg – 10 ng total RNA
- Stranded mRNA sequencing
100 ng - 1 ug total RNA
Identify antisense expression
Enhance alignment and transcript annotation
- Total RNA sequencing (rRNA depletion)
Non-coding RNA (lincRNA)
Small nuclear RNA (snRNA)
Small nucleolar RNA (snoRNA)
- Ribo-profiling
Which mRNA are actively translated
- TRAP-Seq
Cell type specific mRNA expression
- Single Cell mRNA sequencing

Genomics Resource Center

WRB 725 24/7 access

www.rockefeller.edu/genomics

genomics@rockefeller.edu



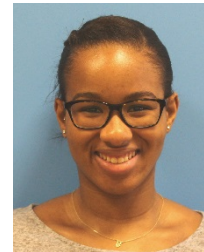
Connie Zhao, Ph.D.
Director



Christine Lai
Research Support Associate



Hong Duan, PhD
Research Support Specialist



Nneka Nnatubeugo
Research Support Assistant



Bin Zhang
Research Support Specialist



Sophie Huang
Bioinformatics Specialist



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High Throughput and Spectroscopy Resource Center DWB 216

<http://inside.rockefeller.edu/htsrc>

Fraser Glickman, Ph.D

Director

Carolina Adura, Ph.D

*Research Support
Specialist*

Jeanne Giganti, MS

*Research Support
Specialist*

Lavoisier Ramos-Espiritu,
Ph.D.

*Research Support
Specialist*

Brittany Dhital, MS

Research Support Assistant

- Small Molecule Libraries
- High Throughput Assay Technologies
- Spectroscopy
- High Throughput liquid dispensing/Lab automation
- Chem Informatics
- Drug Discovery Consultation and Support

Compound/Cheminformatics

Your Recent Screening Assays [Create a new screening assay](#)

[See all screening assays](#)

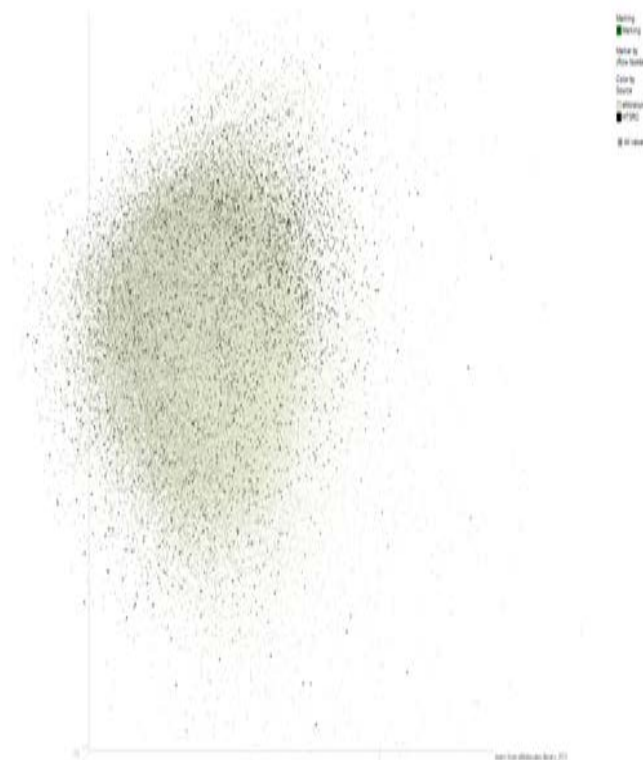
Screening assay	Category	Molecules
HTSRC.CellGlo.HepG2.Pilotstudy Pilot study with plate LOPAC at 10 µM. Luminescence CellTiter Glo reading HepG2 cells.	Cell - Lum	1286
Vartanian.TLR2.PilotScreen Pilot Screen of the LOPAC library (317-320) at 10µM. Duplicate screens over two separate days.	Cell - Luminescence	1286
Shaham.LCin1.PilotScreen Pilot Screen of LOPAC library at 40µM.	Cell - Fluorescence Imaging	2279
Strickland.Fluor-Pol.PrimaryScreen	Biochemical-FP	92597
MacKinnon.hBK.PilotScreen (inhibition) Pilot Screen of the LOPAC and PAINS libraries in duplicate at 10µM.	Biochemical-Fluorescence	2279

Your Recent Molecules [Create a new molecule](#)

[See all molecules](#)

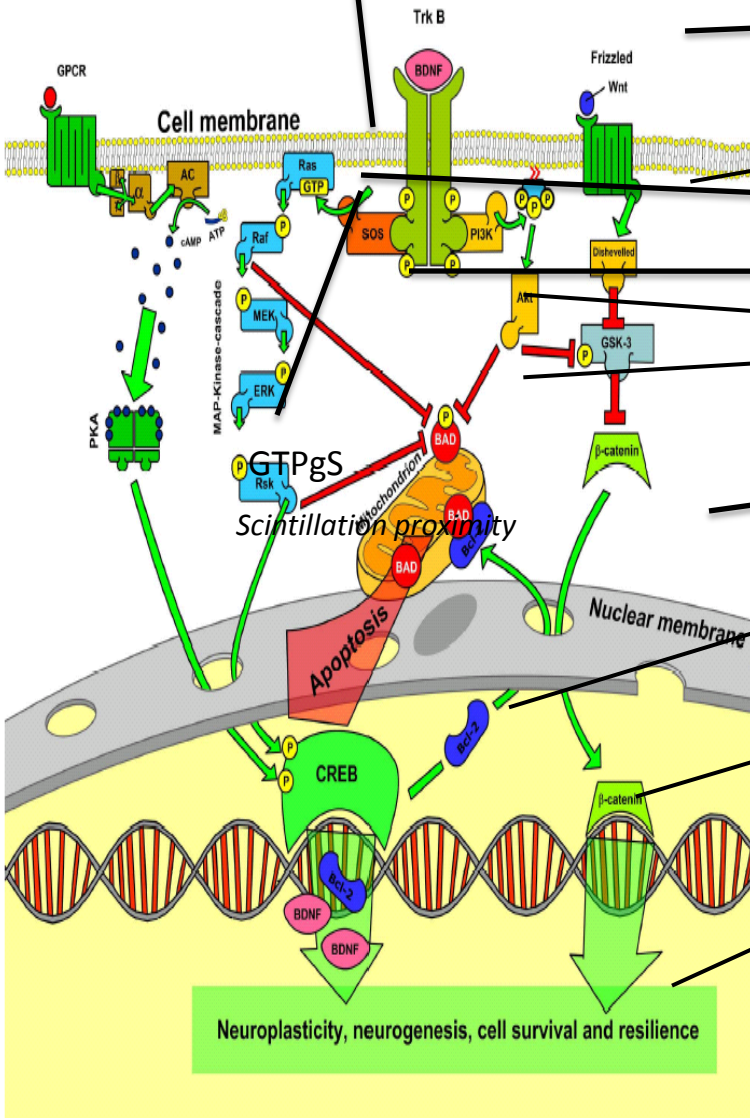
RU-0111505	RU-0000616	RU-0000830
RU-0182354	RU-0182428	RU-0182408

10 Scatter Plot



Membrane potential

HTS Assay Technologies



Receptor binding
Scintillation proximity

Receptor Activation
*ELISA, AlphaScreen
TR-FRET*

cAMP Calcium
phospho protein detection
*Alpha, FLIPR
ELISA
AlphaScreen
TR-FRET*

Protein-protein interactions

Nuclear translocation
high throughput microscopy

Reporter Gene Assays
Luciferase constructs

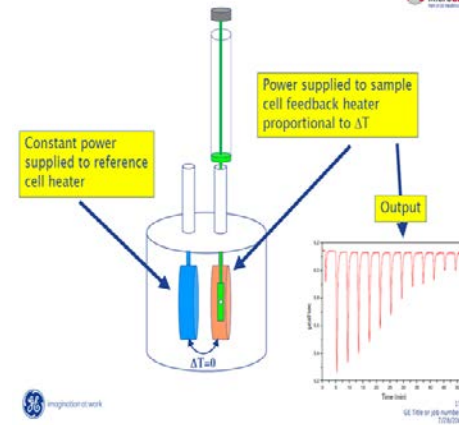
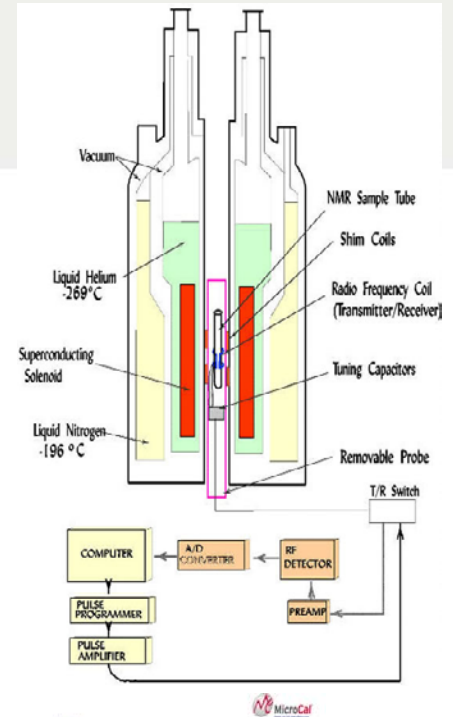
morphological changes
high throughput microscopy

Cell Survival
*MTT
Alamar Blue
Cell-titerGlo (ATP detection)*

Neuroplasticity, neurogenesis, cell survival and resilience

Analytical Techniques

- Nuclear Magnetic Resonance Spectroscopy
- Surface Plasmon Resonance
- Microscale Thermophoresis
- Circular Dichroism
- Fluorescence Polarisation
- Fluorescence Resonance Energy Transfer
- Radioligand binding
- Luminescent Oxygen Channeling (AlphaLISA)
- Infrared Scanning (LICOR)
- High Throughput microscopy
- Isothermal Calorimetry



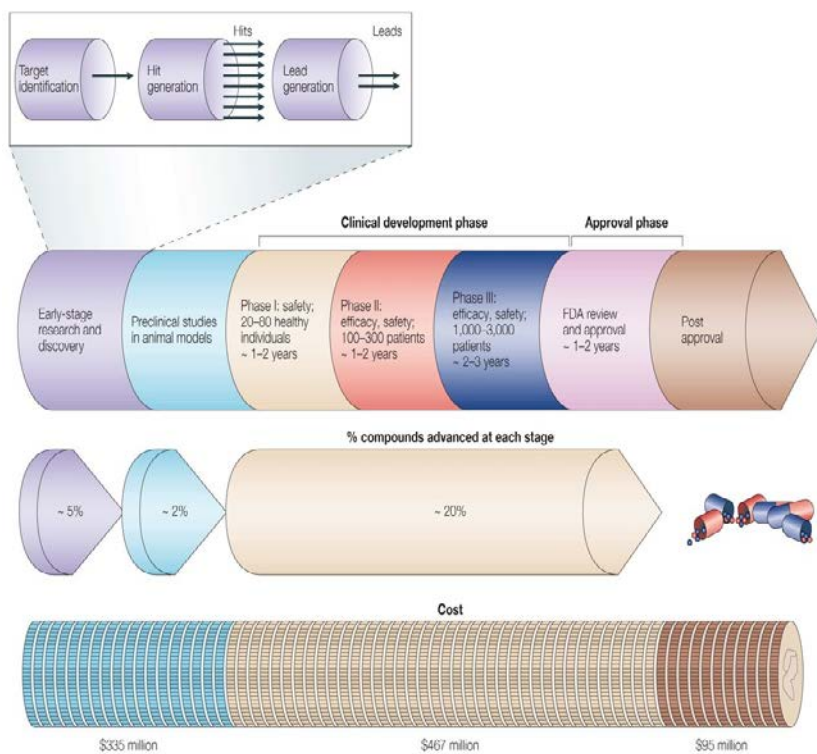
Tecan Evo 150 multi-functional liquid handler



- Microtiter plate-to-plate transfers in both 96- and 384-well plate formats
- 1uL to 200uL
- 8-channel liquid handling arm parallel pipetting from 1uL up to 1000uL.
- high-throughput plate stackers for up to 100 plates
- barcode reader
- disposable tip washer
- replicate plates, cherry pick compounds, serially dilute across a plate, and with the center's assistance, build a custom application for one's needs

Drug Discovery

- How to convert a molecular target into a screening assay
- Design of Secondary assays for Selectivity and specificity analysis
- Development of Structure Activity Relationships
- External Pharmacology/Chemistry Resources





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Proteomics Resource Center

Who we are...





SCIENCE FOR THE BENEFIT OF HUMANITY

Proteomics Resource Center



Joe

Henry

Sue

Brian

Milica

Henrik



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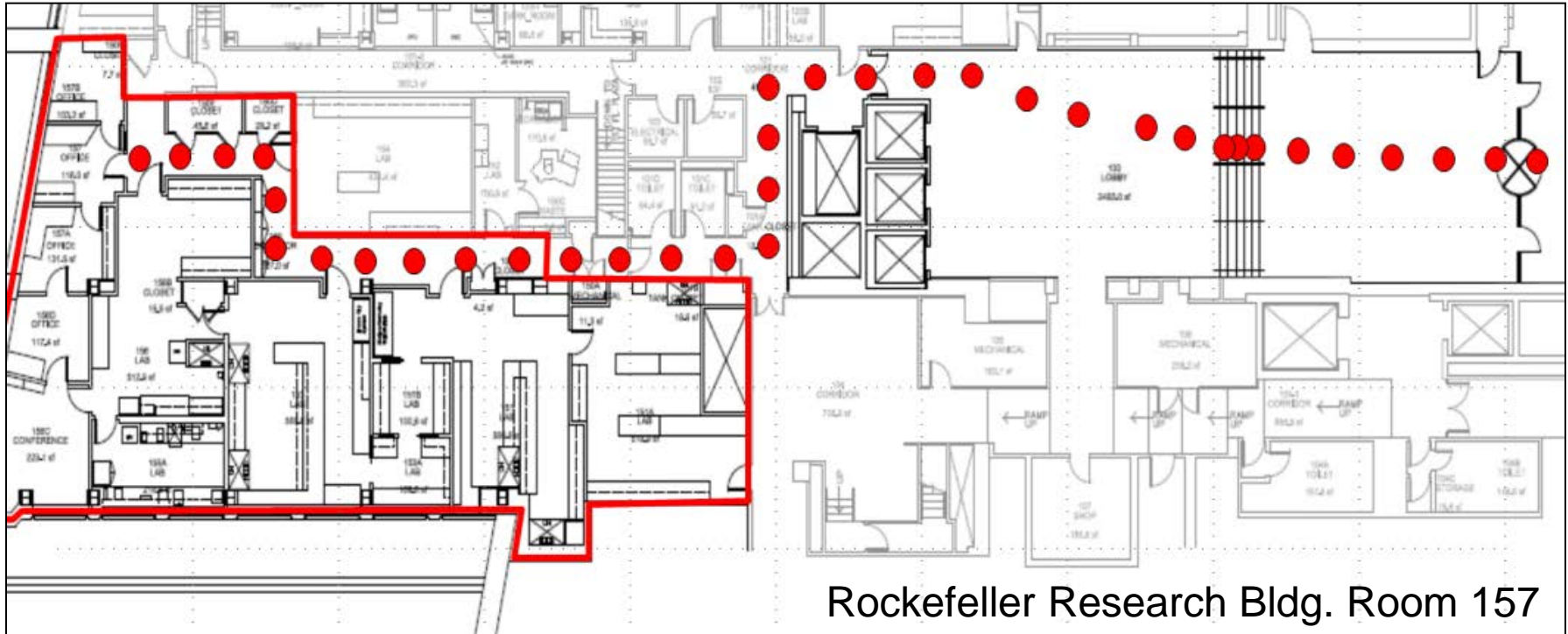
Proteomics Resource Center

What we do...

- **Proteomics – identification and quantitation of proteins, peptides and small molecules by mass spectrometry**
- **Peptide synthesis**
- **Analytical chemistry**
- **User instrumentation**
- **Help planning experiments...**

Proteomics Resource Center

Where to find us...



Rockefeller Research Bldg. Room 157

<http://inside.rockefeller.edu/proteomics/>

Comparative Bioscience Center



COMPARATIVE BIOSCIENCE CENTER



Veterinary Services

Laboratory of
Comparative Pathology

Training & Videos

Transgenic Laboratory

IACUC

Gene Targeting
Laboratory

Services



Welcome to the Comparative Bioscience Center

The Comparative Bioscience Center (CBC) provides a comprehensive and humane program of laboratory animal care in support of Rockefeller University's *in vivo* research. We continually strive to refine and promote quality husbandry and health care and we are dedicated to maintaining a collaborative, user-friendly environment that actively promotes and facilitates translational research dedicated to the betterment of science and the animal care and use program.

COMPARATIVE BIOSCIENCE CENTER

VETERINARY
SERVICES

COMPARATIVE
PATHOLOGY LAB

TRAINING
& VIDEOS

TRANSGENIC
LABORATORY

IACUC

GENE TARGETING
LABORATORY

ANNOUNCEMENTS

New Non-Compliant Cage Card Policies and Guidelines
[Read more](#)

QUICK LINKS

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[Forms & Policies](#)
[Services](#)
[Rates](#)

cbc.rockefeller.edu

- AAALAC accredited
- Providing comprehensive support for *in vivo* studies
- CBC Research Resources: IVIS Spectrum, Vevo 2100 Micro-Ultrasound, RS-2000 Biological Irradiator

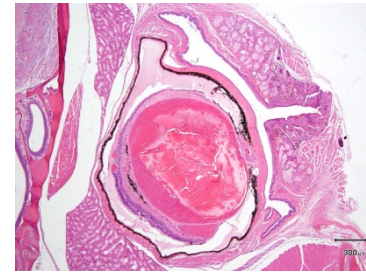
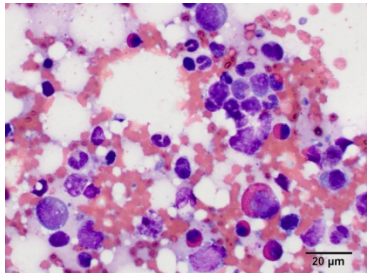


SCIENCE FOR THE BEAUTY OF HUMANITY

Laboratory of Comparative Pathology (LCP)

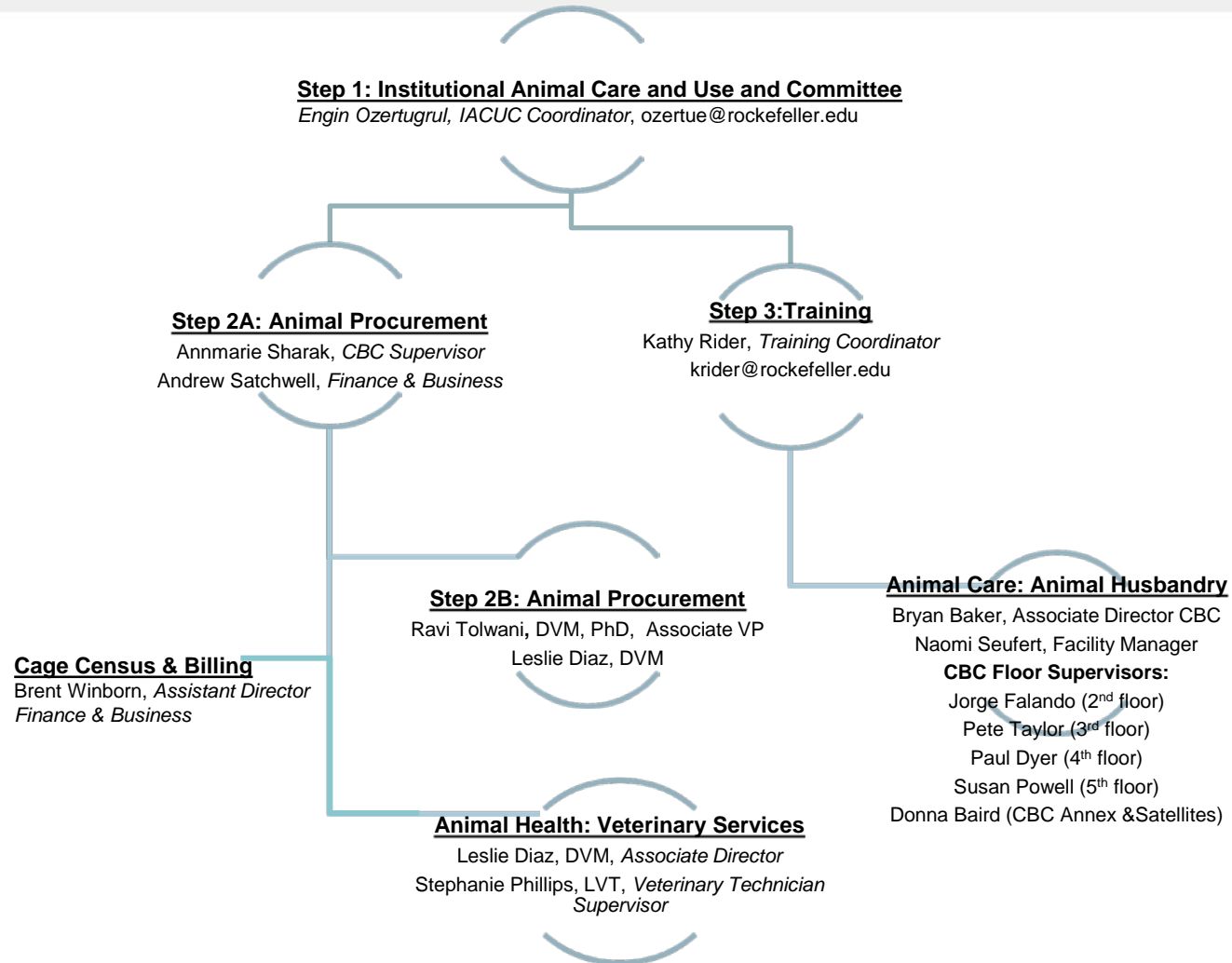
Tri-Institutional Resource: MSKCC, WCMC, RU

- Necropsy, histopathology, clinical pathology
 - Including immunohistochemistry, immunofluorescence, frozen sections, blood profiles, bone marrow, etc.
- Genetically engineered animal (GEA) phenotyping core
- Mission:
 - Support investigative staff using a variety of animal models
 - Research pathology
 - Animal health programs (sentinel, quarantine, veterinary services)
 - Digital photography, slide scanning, aid with manuscript preparation
 - Training
 - Investigative staff
 - Blood collection, perfusion, necropsy, bone marrow/tissue collection, etc.
 - Training is free of charge





Comparative Bioscience Center





Laboratory of Comparative Pathology (LCP)

Tri-Institutional Resource: MSKCC, WCMC, RU

Comparative Pathologists

Julie White DVM, Dipl. ACVP



**Michelle Lepherd, BVSc, PhD
Dipl ACVP**

Sebastien Monette, VMD, MVsc, Dipl. ACVP



**Tri-Institutional Laboratory of
Comparative Pathology &
Genetically Engineered Mouse
Phenotyping Service**

**Memorial Sloan-Kettering Cancer Center
Zuckerman Lab: 940**

646-888-2422

WCMC lab: C-708, 710

212-746-3399

Email : lcp@mskcc.org

<https://cbc.rockefeller.edu/comparative>



SCIENCE FOR THE BENEFIT OF HUMANITY

Gene Targeting Resource Center

Tri-Institutional Resource: MSK, WCMC, RU

Chingwen Yang, Ph.D.

Director

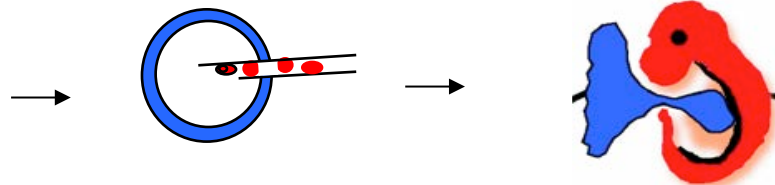
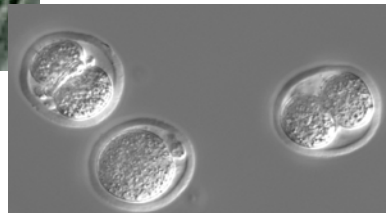
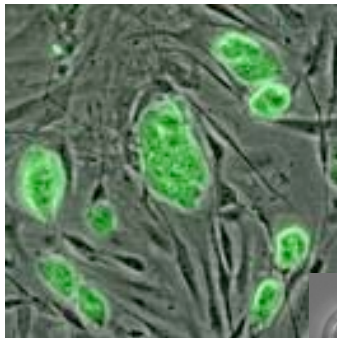
DWB 703

Tel: 212-327-8649

yangc@rockefeller.edu

<http://rockefeller.edu/geneTargeting>

- Gene targeting in mouse ES cells
- ES cells management
- Derivation of mouse ES cell lines
- ES mouse production
- QC of transgenic vector DNA
- TALEN/CRISPR



Gene Targeting Resource Center



Pradip Kar, M.S.

Ekaterina Zafranskaia, M. D.

Dorjee Shola, Ph. D.

Jing Gao, M. D.

Chia-Yun Han, M.S.

Transgenic Services Laboratory

Director

Rada Norinsky

CBC 542-546

Phone: 212-327-7783

rada.norinsky@rockefeller.edu

Great Team of

Transgenic Services Laboratory

CBC 543-546

Phone 212-327-7738

Jahnney Torres, Roxana Cubias,
Xiang-Qing Li, Paula Emhardt,
William Ramirez

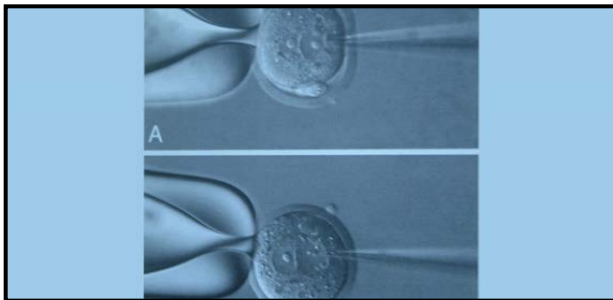


Transgenic Services Laboratory

Services

- ◆ Transgenic/Chimeric Mice Production
- ◆ IVF Services/Assisted Reproduction
- ◆ Strains Cryopreservation
- ◆ Strains Rederivation
- ◆ Strain Rescue

Transgenic Mice Production



Chimeras Production



Strains Cryopreservation



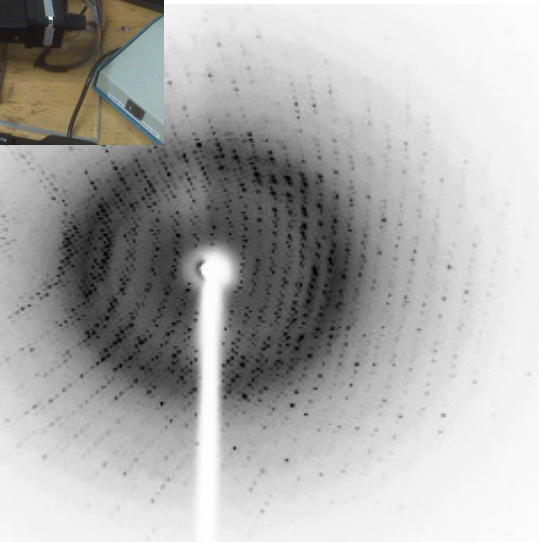
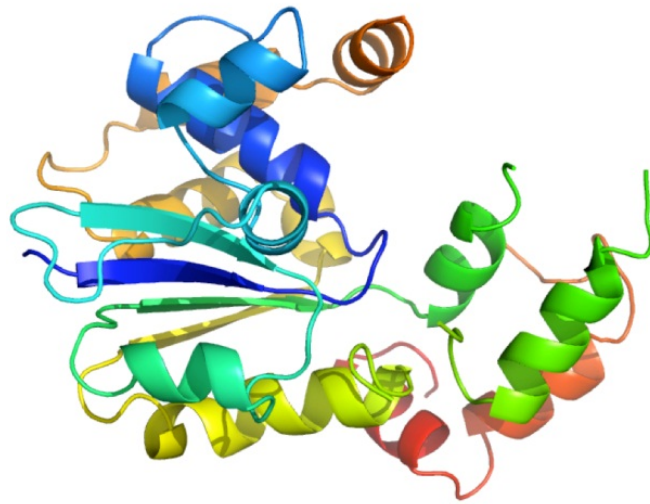
IVF Services/Assisted Reproduction



Structural Biology Resource Center

Do X-ray
crystallography
with us

From crystal growth
to structure determination



Structural Biology Resource Center

Or.... make and purify proteins with us



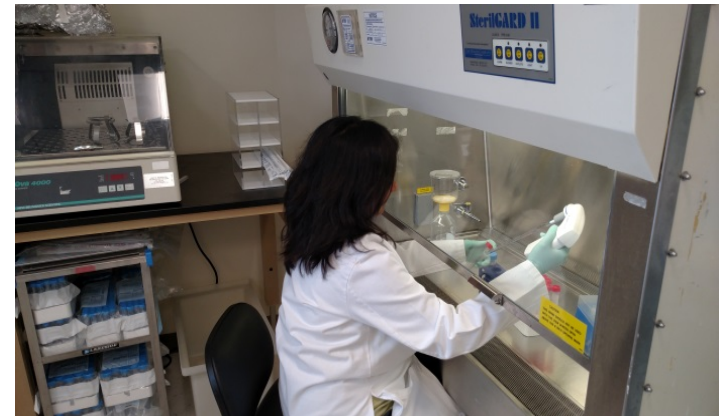
DO YOU WANT PURIFIED PROTEIN?

The Structural Biology Resource Center
has a new protein expression and
purification training center.

- We will provide the tools and knowledge for satisfactory results.
- You will develop the expertise to continue working independently.

THE SBRC NOW HAS :

Chromatography
Biosafety Cabinet
Centrifuges
Incubators
Insect Cell Lines



Structural Biology Resource Center



Äkta Purifier FPLC (Chromatography)



Phastsystem
2 and 3D gel
electrophoresis



Cryoshipper



Plate microfuge

Deena Oren, Ph.D.

Manager

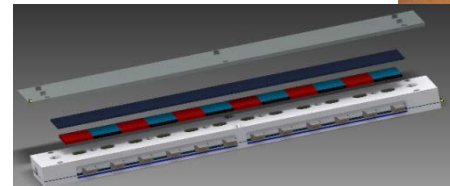
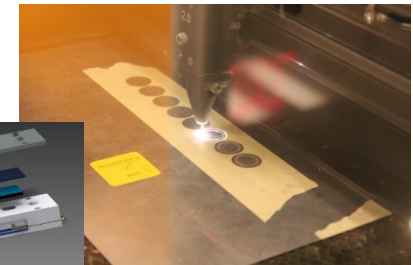
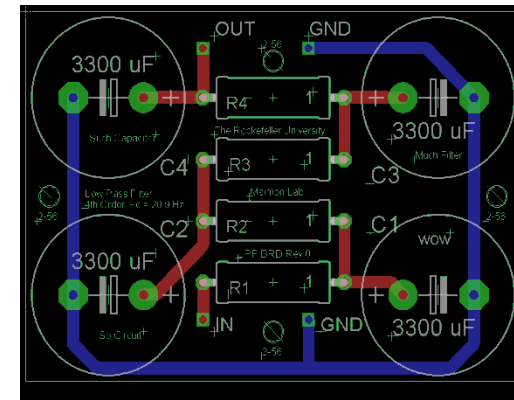
RRB Suite 140

inside.rockefeller.edu/sbrc

Precision Fabrication Facility (PFF)

- [How Can You Improve Your Science?](#)

- Training to use various design and fabrication tools
- Design consultations and assistance
- Stock materials (plastic and metal)
- Workspace and other tools
- Equipment
 - 3D and 2D modeling software
 - 3D Printer (16 micron)
 - Laser Cutter
 - CNC Mill
 - Knee Mill
 - Lathe
 - Electronics and Microprocessors





SCIENCE FOR THE BENEFIT OF HUMANITY

Precision Fabrication Resource Center (PFRC)

Location: CRC SC02

Phone: x7056

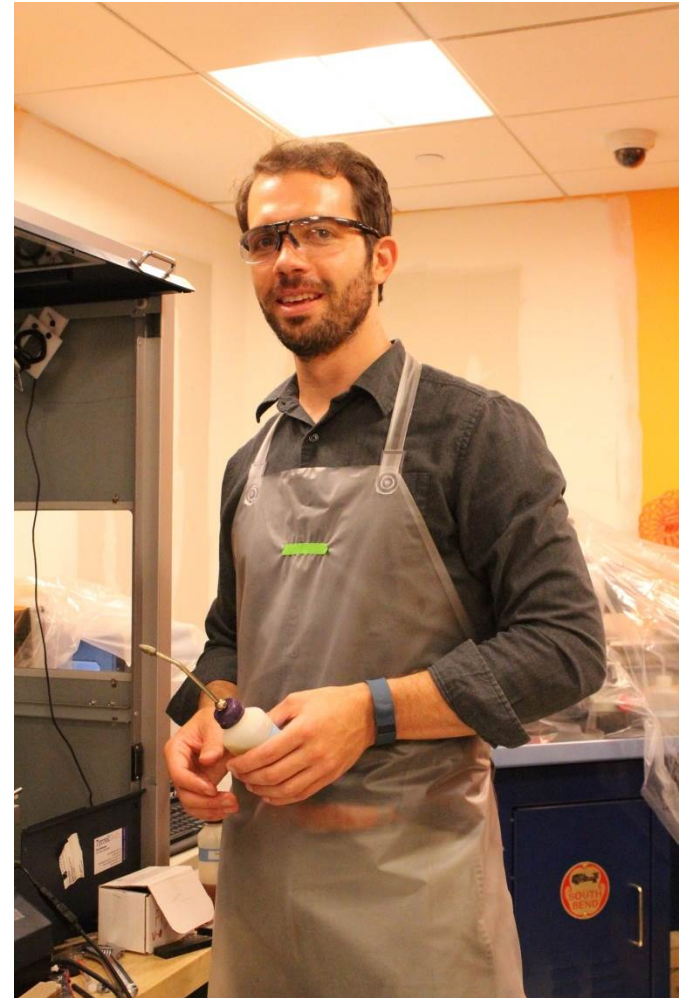
inside.rockefeller.edu/fabrication/

Contact:

Jim Petrillo

Instrumentation Engineer

PFRC Director





Resource Center Locations

- **Bio-Imaging (DWB 201-203)**
- **Antibody and Bioresource Core Facility (DWB 415)**
- **Cryo-Electron Microscopy (CRC B13)**
- **Electron Microscopy (RRB 120)**
- **Flow Cytometry (DWB 205-211)**
- **Genomics (WRB 725)**
- **High Throughput Screening (DWB 219)**
- **Proteomics (RRB 157)**
- **Comparative Bioscience Center**
- **Laboratory of Comparative Pathology (MSK)**
- **Gene Targeting (DWB 703)**
- **Transgenics (CBC 542-546)**
- **Structural Biology (RRB 1st floor)**
- **Precision Fabrication Facility (Greenberg Building – C Floor)**