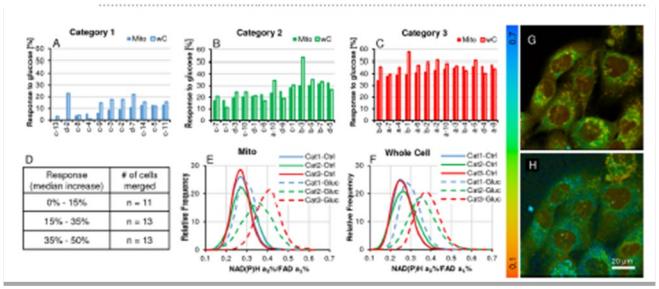
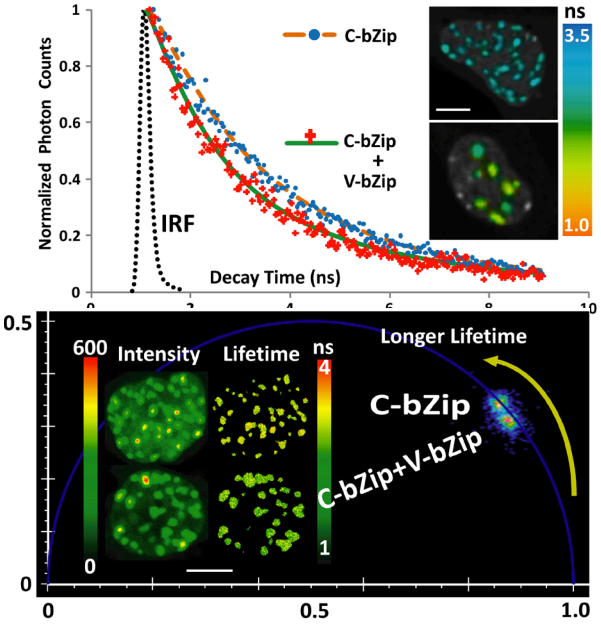


Metabolic response of segmented prostate cancer cells after treatment, based on FLIRR (Fluorescence Lifetime Redox Ratio) –NAD(P)H-a2%/FAD-a1%. Scientific Reports | *(2018) 8:79,* DOI:10.1038/s41598-017-18634-x



FLIM-FRET Microscopy to investigate proteins dimerizing in the nucleus. Top: Time correlated single photon counting (TCSPC) method (Becker & Hickl). Bottom: Frequency-domain technique (ISS). Nature Protocols 6 (9), 1324-1340, 2011

Systems and live model cell line - transfected with above proteins will be available at workshop for the practical.

Faculty

Dr. A. Periasamy, University of Virginia
Workshop Director, ap3t@virginia.edu
Dr. M. Barroso, Albany Medical College, NY
Dr. M. Börsch, Jena University, Germany
Dr. J. N. Demas, Chemistry, UVA
Dr. M. Digman, Univ. of California-Irvine
Dr. A. Kenworthy, MPBP, UVA
Dr. A. Rück, Universität Ulm, Germany
Dr. S. Vogel, NIAAA, NIH
Dr. A. Walsh, Texas A & M University
Dr. Jin Zhang, UC at San Diego

Guest Lecturers

Dr. Huiwang Ai, MPBP, UVA
Dr. K. Siller, Research Computing, UVA
Dr. M. Skala, Univ. Wisconsin
Dr. P. So, MIT
Dr. M. Stanley, Chroma Tech.

For more info:

Partial Tuition fees Scholarships available, visit the link below or contact ap3t@virginia.edu
https://kcci.virginia.edu/workshop-2025

TUITION FEES:
$2,500 non-profit organizations
$2,900 for-profit organizations
(**Includes lodging, breakfast, lunch, dinner, lecture materials**)

Contact:
Prof. Ammasi Periasamy
ap3t@virginia.edu

22nd Annual Workshop on FLIM and FRET Microscopy and Label-free NAD(P)H/FAD Metabolic Imaging

https://kcci.virginia.edu/workshop-2025

June 9-13, 2025

Hands-on instructions on microscopy systems
*** Internationally recognized Faculty**
*** Best imaging and analysis solutions**
*** Personal attention for a maximum of 25 participants**
*** Individual problem solving**

W.M. Keck Center for Cellular Imaging, University of Virginia
https://kcci.virginia.edu/workshop-2025

AIM

The W.M. Keck Center for Cellular Imaging (KCCI), a university imaging center at the University of Virginia, is sponsoring an advanced practical course on (a) Förster (fluorescence) resonance energy transfer (FRET) for confocal and fluorescence lifetime imaging microscopy (FLIM-FRET); and (b) label-free FLIM microscopy of NAD(P)H and FAD to analyze the Redox metabolic states (FLIRR) in live cells before and after treatment.

Attendees are expected to be familiar with the basics of fluorescence microscopy. The curriculum, after a brief introduction to the principles of fluorescence, microscopy, fluorophores, FRET and FLIM, will concentrate on the practical aspects, hands-on individual instruction at the instruments followed by data analysis and interpretation.

Lectures and after dinner problem-solving discussions will address questions of fluorophore choices, the most suitable systems to achieve specific research objectives, qualitative vs. quantitative analysis and many more related subjects. Participants will also be introduced to a unique image processing and analysis software (PFRET) and Python code for FLIRR.

10+ different and advanced microscopy systems will be available for a maximum of 25 students. With internationally recognized faculty in attendance, there is ample opportunity to interact with experts formally or informally.

Live-cell specimens are provided.

Participant's own specimens are welcome.

PROGRAM SCHEDULE

Lectures by leading scientists
(Monday 10-6pm theory; other days
Theory 8:30 AM – 12 PM)

- Introduction to workshop
- Basics of Fluorescence, FRET, FLIM, NADH, FAD, Trp, microscope choices
- Meet the experts and Q&A on the subjects:
- Confocal/spectral FRET
- FLIM-FRET, NAD(P)H-TRP FRET
- Fluorophore pairs for FRET/FLIM-FRET
- FLIM-FRET,
- Redox states analyzed by NAD(P)H & FAD
- Metabolic Imaging
- Imaging live/fixed cells & tissue
- Spectroscopy FRET in suspensions
- Bacterial FRET

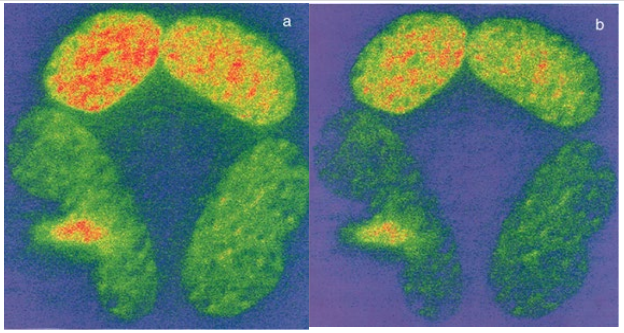
June 10-12 Lab (1 PM – 5 PM)*

Hands-on practical instruction on various systems, data analysis, special demos, and general problem-solving discussions on

- Anisotropy / Homo-FRET
- NAD(P)H-TRP FRET
- FLIM analysis: Fitting and Phasor plots
- Single-molecule FRET
- Metabolic Imaging
- FRAP
- Working on your instrument of choice after formal curriculum ends

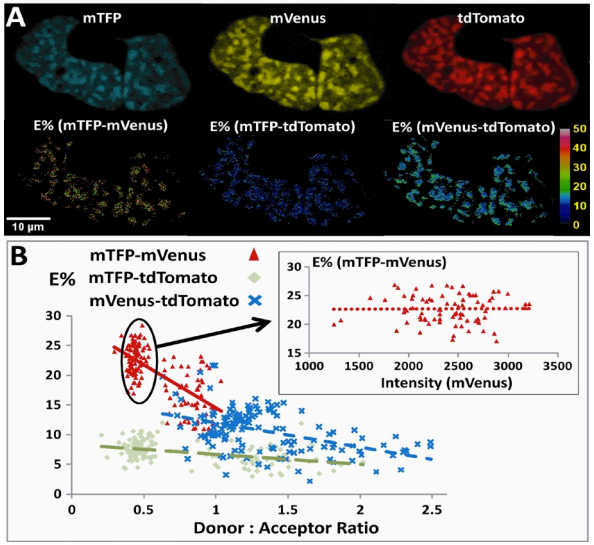
June 13th 8:00 to 12pm Lab; 1:00 to 3pm
Theory and certificate distribution.

***Including breakfast, coffee breaks, lunch, and dinner.**



BFP-GFP-Pit1 Protein Dimerization. (a) Before correction. (B) after PFRET correction.

This PFRET correction software will be available for workshop participants.



An expanded PFRET software analyzes 3 FRET-interacting, labeled proteins simultaneously in live cells – a Keck Center for Cellular Imaging development. Biophys. J. 99, 1274-1283, 2010

Participating Instrument Companies

Becker & Hickl, Boston Electronics, Carl Zeiss, Chroma Tech, ISS Inc., Leica Microsystems, Olympus.