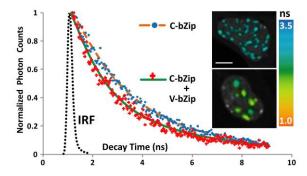
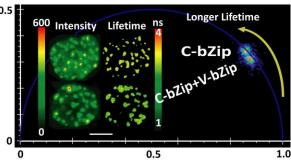


Multi-Photon tissue FRET in traumatic axonal injury. Energy transfer is consistent with BAD (Alexa488)-Bcl-xl (Alexa555) heterodimerization. Differentiation 71: 528-541, 2003





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. R. N. Day. Indiana University

Workshop Co-Director, <a href="mailto:rnday@iupui.edu">rnday@iupui.edu</a>

Dr. M. Barroso, Albany Medical College, NY

Dr. M. Börsch, Jena University, Germany

Dr. J. N. Demas, University of Virginia

Dr. A. Kenworthy, University of Virginia

Dr. S. Vogel, NIAA, NIH

Dr. Yuansheng Sun, ISS Inc., Illinois

Dr. Jin Zhang, UCSD, CA

#### **Guest Lecturers**

Dr. M. Stanley, Chroma Tech., VT

Dr. P. So, MIT, MA

## For more info:

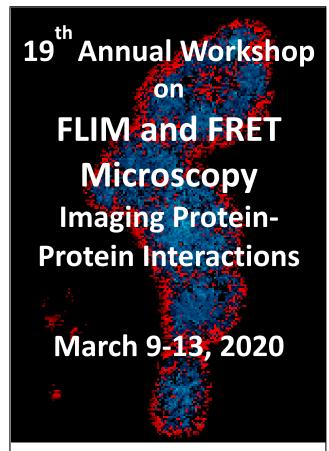
http://www.kcci.virginia.edu/workshop/workshop2020

**FEES**: \$2,200 non-profit organizations \$2,600 for-profit organizations (Includes lodging, breakfast, lunch, dinner, lecture materials)

## **Contact:**

Dr. Ammasi Periasamy Email: ap3t@virginia.edu

Phone: 434-243-7602 or 982-4869



# Hands-on instructions on imaging 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 www.KCCI.virginia.edu/workshop/ workshop2020

### **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 Förster (fluorescence) resonance energy transfer (FRET) and fluorescence lifetime imaging (FLIM) microscopy.

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, handson 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).

**10+** different and advanced microscopy systems will be available for a maximum of 25 students. With 13 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**

### Day 1 (10 AM Noon - 7 PM)\*

- Introduction to workshop
- Basics of Fluorescence, FRET, FLIM, microscope choices
- Meet the experts from Leica, Olympus, Zeiss, Nikon, Chroma Tech, IdeaElan, Applied Precision, Lumen Dynamics, Becker & Hickl, Boston Electronics, ISS, Lambert.

## Days 2-4 (8:30 AM - 12 PM)\*

Short lectures and Q&A on the subjects:

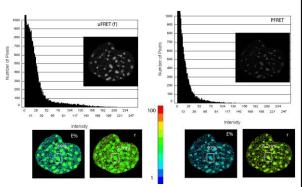
- Confocal/spectral wide-field FRET
- FLIM-FRET
- Fluorophore pairs for FRET/FLIM-FRET,
- Protein fusions
- Imaging live/fixed cells & tissue
- Spectroscopy FRET in suspensions
- Bacterial FRET

## Days 2-4 (1 PM - 9 PM)\*

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

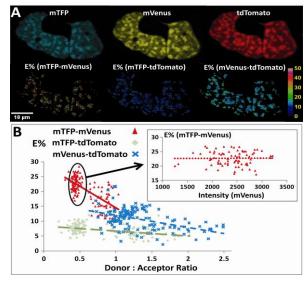
- Anisotropy / Homo-FRET
- TIRF-FRET
- Spectrofluorometer FRET
- FLIM analysis: Fitting and Phasor plots
- Single-molecule FRET
- Fluorescence Correlation / Cross-correlation Spectroscopy
- FRAP
- Working on your instrument of choice after formal curriculum ends

\*including breakfast, breaks, lunch and dinner.



Comparing uFRET (uncorrected for spectral bleed-through - SBT) with PFRET (Processed FRET, corrected for SBT).

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**

Carl Zeiss, Leica Microsystems,
Olympus, ISS, Becker & Hickl, Boston
Electronics,

Chroma Tech, Excelitas Technologies
Corp