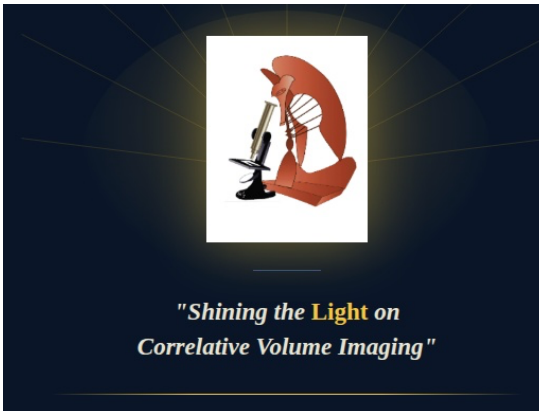
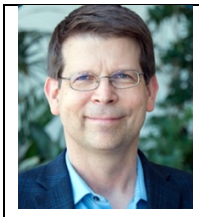
Midwest Microscopy and Microanalysis Fall Meeting

Topic: *Shining the Light on Correlative Volume Imaging*
November 20 , 2026

Host: Baxter Healthcare Headquarters
1 Baxter Parkway, Deerfield IL, 60015



Keynote Speakers



Visualization, quantification and multi-scale approaches for interrogating photosynthetic organisms by 3D conventional and cryo-volume electron microscopy

Kirk J. Czymmek, Ph.D.
Advanced Bioimaging Laboratory, Donald Danforth Plant Science Center, Saint Louis, MO, USA;

Volume electron microscopy (vEM) has the potential to enable new insights into the structure-function relationships of photosynthetic organisms in three dimensions at nanometer resolution. However, plants, algae and cyanobacteria present unique sample preparation challenges due to their waxy cuticles, cell walls, air spaces and large vacuoles, which impede fixation and downstream processing, cause uneven freezing and result in poorly conductive samples. As a result, vEM imaging of photosynthetic organisms is severely lacking compared to their mammalian counterparts. Here, I will describe strategies for both conventional and cryogenic vEM workflows on photosynthetic organisms, with emphasis on the Thermo Scientific Helios Hydra plasma focused ion beam scanning electron microscope (pFIB-SEM).
For conventional resin-embedded samples, improved heavy metal staining combined with the multiple ion species of the Hydra pFIB-SEM allows milling and imaging conditions to be matched to otherwise difficult plant material. Building on this, an artificial intelligence (AI) driven Adaptive Scanning workflow will be described, where a semantic segmentation model trained on a low-resolution scan generates a mask that directs high-resolution imaging only to

targeted regions of each z-slice. Since air spaces, vacuoles and other off-target structures represent a substantial proportion of plant tissue, this multi-resolution strategy realized time savings of up to 50+% while retaining important sample context. Complementary cryo-vEM of vitrified, unfixed and unstained cyanobacteria and alga resolved chloroplast, pyrenoid and thylakoid architecture and the remarkable interconnectivity of organelles in cells. As cryo-vEM becomes more accessible, robust workflows for photosynthetic organisms will be instrumental in supporting the broader plant microscopy community.

Simon Alford, Ph.D.
Sweeney Professor of Basic Sciences and Head
Department of Anatomy and Cell Biology
University of Illinois at Chicago College of Medicine

4D live cell imaging of synapses and the compromises in resolving fast signals in small volumes

Fluorescence imaging, whether of synthetic dyes, or genetically engineered sensors, provides expanding opportunities to probe specific and spatially segregated signaling in cells and tissues. However, signaling events can be in millisecond time domains, particularly in the nervous system. Imaging fast neuronal signaling represents a challenge, pitting spatial versus temporal resolutions of events localized to small subcellular structures. These challenges are particularly profound in quantifying synaptic transmission and the signaling that supports it. For example, in presynaptic terminals both triggers of neurotransmitter release and release of transmitter itself occur within milliseconds. Synaptic dynamics in distinct neuronal populations within intact neural circuits remains challenging due to limitations in acquisition speed, penetration depth, spatial resolution, and spectral cross talk in conventional systems. Fluorescence imaging is also inherently destructive. Fast imaging requires high intensity illumination which bleaches dyes and initiates photodamages. This limits tracking functional changes over time.

Light sheet microscopy overcomes some of these limitations, and by combining the high spatiotemporal resolution available in lattice light-sheet microscopy (LLSM) with chromatically distinct dyes and genetically encoded sensors), it is feasible to resolve cell signaling resolved in 4D with frame rates of 300 Hz or higher. This can be achieved at lateral resolutions of approximately 240 nm and axial resolution of approximately 800 nm with little spectral cross talk and low photo bleaching.

I will demonstrate examples of spectral separation between selectively targeted genetic sensors during high-speed diffraction limited 4D resolved imaging, and some comparative advantages and disadvantages of using genetic sensors and dyes. I will also address challenges in analyses of large datasets with many stochastic events. Solutions using automated ROI detection, transient extraction, double-exponential decay fitting, and comprehensive spectral analysis including power spectral density and time-frequency decomposition.

Opportunities for Optical Characterization at Local Univerisites:
Quadripartite Presentations by R&D Staff :

An outline of the capabilities for Optical Characterization at regional facilities will be presented by R&D Staff of Northwestern Univ., Univ. of Illinois at Chicago, Univ. of Chicago, and Purdue University

Vendor Presentations:

CLEM – Past, Present and Future
Matt Joens/ThermoFisher:

Light and electron microscopy provide complementary views of biological specimens: light microscopy can reveal labeled targets and cellular context, while electron microscopy resolves fine structure. Combining the two has enabled researchers to connect molecular signals with ultrastructure. Originally, CLEM, or comparative light and electron microscopy was developed to guide microscopists to certain features by examining adjacent sections using chromographic stains. As techniques evolved over the years, so did CLEM as it was eventually rebranded as correlative light and electron microscopy. In this new version, samples were prepared in such a way that both light and electron microscopy could be performed on the same sample without needing to modify or re-stain the sample along the way. These protocols were often complex, lengthy, and required multiple tools to be successful. More recently, cryogenic CLEM has made it possible to image vitrified specimens in a near-native state, preserving structural detail and fluorescence signals. In this talk, we will trace the development of correlative techniques and discuss current methods, challenges, and future directions in electron microscopy.

Student Presentations:

From Light to Electrons: Mapping the Cellular Uptake of Biomolecular Condensates Using Correlative Light-Electron Microscopy (CLEM)

Yi Li , Kelley laboratory, Northwestern University

Biomolecular condensates (BMCs) are membraneless assemblies formed through liquid–liquid phase separation, offering a versatile platform for engineering functional biomaterials. Understanding how BMCs enter cells and navigate intracellular compartments is essential for their biomedical applications. In this talk, we will demonstrate a correlative light and electron microscopy (CLEM) workflow that bridges these complementary imaging modalities to investigate BMC uptake and intracellular trafficking. By combining fluorescence-based localization with focused ion beam scanning electron microscopy (FIB-SEM), we achieve three-dimensional ultrastructural reconstruction of BMCs and their surrounding cellular compartments. This approach connects molecular identity with ultrastructural context, offering new insights into condensate–cell interactions and demonstrating the potential of CLEM for studying soft biomaterials

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Characterizing Axonal Golgi Satellites using Live-Cell Imaging, Correlative Electron Microscopy, and Cryo-Electron TomographyFernando

Zacahua – University of Chicago

Abstract - TBA