Title: Investigating pathophysiological mechanisms of Mast Cell Activation Syndrome (MCAS) with deeper and higher-resolution label-free nonlinear microscopy

Authors: Kasey Shashaty, Kunzan Liu, Honghao Cao, Li-Yu Yu, Randall Dass, Jared Rosenblum, John Biggins, Paolo Bolognese, and Sixian You

Conference BO508 Label-free Biomedical Imaging and Sensing (LBIS) 2025

200–300-word abstract:
Label-free nonlinear microscopy based on optical redox ratio allows for high-resolution and three-dimensional imaging of live biological specimens without the need for exogenous labels. However, its application to thick tissues has been restricted by its limited penetration depth and worsening 3D resolution within 300 microns largely due to light scattering and axial blurring due to diffraction. Here, we demonstrate imaging depth improvement with a multimode fiber source and axial resolution enhancement via physics-based deep learning on surgically excised human tissue. We have recently shown that deeper metabolic imaging of NAD(P)H can be achieved with three photon excitation at 1100 nm, in the form of deep simultaneous label-free autofluorescence-multiharmonic (dSLAM) microscopy, which provides rich characterization of the biological environment by integrating second harmonic generation (SHG) and third harmonic generation (THG) as well as autofluorescence from NAD(P)H and FAD through an excitation band centered at 1110 nm. The higher pulse energy required for deep metabolic imaging was achieved by combining a step-index (SI) multimode fiber (MMF) with a programmable fiber shaper. In collaboration with Mount Sinai, we propose the use of deep and high-resolution label-free autofluorescence-multiharmonic (dSLAM) microscopy in a clinical investigation of tissue from patients with comorbid hypermobile Ehlers-Danlos Syndrome (hEDS) and mast cell activation syndrome (MCAS). We monitor and compare mast cell distribution and tissue structure, cell morphology and metabolism in surgically excised human tissue originating from around the craniocervical junction in patients with comorbid hEDS and MCAS to healthy human tissue.

50-150-word abstract:
Label-free nonlinear microscopy allows for high-resolution and three-dimensional imaging of live biological specimens without the need for exogenous labels. However, its application to thick tissues has been restricted by its limited penetration depth and 3D resolution within 300 microns. Here, we demonstrate imaging depth improvement with multimode fiber-based low-repetition-rate high-peak-power three-photon excitation of NAD(P)H and axial resolution enhancement via physics-based deep learning. Such deeper and higher-resolution label-free nonlinear microscopy is used in a clinical investigation of pathophysiological mechanisms of comorbid hypermobile Ehlers-Danlos Syndrome (hEDS) and mast cell activation syndrome (MCAS). We monitor and compare mast cell distribution and tissue structure, cell morphology

and metabolism in surgically excised human tissue originating from around the craniocervical junction.

Title: AI-assisted isotropic label-free microscopy for human connective tissue disorders

Authors: Kasey Shashaty, Jiashu Han, Kunzan Liu, Randall Dass, Jared Rosenblum, John Biggins, Paolo Bolognese, and Sixian You

Conference BO510 Computational Optical Imaging and Artificial Intelligence in Biomedical Sciences

250-word abstract:

Label-free nonlinear microscopy allows for high-resolution and three-dimensional imaging of live biological specimens without the need for exogenous labels. However, similar to other microscopy technologies, axial resolution in label-free microscopy is fundamentally limited to be 2-5 times worse than lateral resolution. This challenge is particularly pronounced in label-free imaging of thick, heterogeneous tissues, where assumptions about data distribution (e.g. sparsity, label-specific distribution, and lateral-axial similarity) and system priors (e.g. independent and identically distributed (i.i.d.) noise and linear shift-invariant (LSI) point-spread functions (PSFs)) are often invalid. To overcome this challenge, we use a weakly physics-informed, domain-shift-resistant axial deblurring framework for robust isotropic recovery in intact, thick, and heterogeneous tissues. In collaboration with Mount Sinai, we propose the use of AI-assisted 3D isotropic label-free microscopy in a clinical investigation of tissue from patients with comorbid hypermobile Ehlers-Danlos Syndrome (hEDS) and mast cell activation syndrome (MCAS). With 3D isotropic subcellular label-free imaging, we monitor and compare mast cell distribution and tissue structure, cell morphology and metabolism in surgically excised human tissue originating from around the craniocervical junction in patients with comorbid hEDS and MCAS to healthy human tissue.

50-150-word abstract:

Label-free nonlinear microscopy allows for high-resolution and three-dimensional imaging of live biological specimens without the need for exogenous labels. However, axial resolution in optical microscopy is fundamentally limited to be 2-5 times worse than lateral resolution, a challenge further amplified in thick, heterogeneous tissues where assumptions about data distribution and system priors are often invalid. In collaboration with Mount Sinai, we propose the use of a weakly physics-informed, domain-shift-resistant axial deblurring framework for robust isotropic recovery in a clinical investigation of tissue from patients with comorbid hypermobile Ehlers-Danlos Syndrome (hEDS) and mast cell activation syndrome (MCAS). With 3D isotropic subcellular label-free imaging, we monitor and compare mast cell distribution and tissue structure, cell morphology and metabolism in surgically excised human tissue originating from around the craniocervical junction in patients with comorbid hEDS and MCAS to healthy human tissue.