



# Oklahoma Microscopy Society Spring Meeting 2025 Program



University of Oklahoma, Forum building, 1704 Asp Ave, Norman, OK 73072

April 18, 2025

Welcome to the Oklahoma Microscopy Society Spring Meeting 2025, hosted at the beautiful University of Oklahoma campus in Norman. We are thrilled to gather once again in person to celebrate the diverse and cutting-edge microscopy research happening across our state.

This year's theme, "Advances in Microscopy in Oklahoma," reflects the powerful momentum we're witnessing in imaging sciences—from AI-powered analysis to atomic-resolution electron tomography. We are especially proud to feature talks from rising student researchers as well as keynote insights from internationally respected leaders in the field.

As we look forward to a day filled with insightful talks, engaging discussions, and hands-on lab tours, we encourage you to take full advantage of the opportunities to network with fellow researchers, vendors, and educators. Whether you're here to learn, share, explore instrumentation, or spark new collaborations, your participation helps strengthen our state's thriving microscopy community.

We are grateful to our speakers, sponsors, volunteers, and attendees who make this meeting possible. Thank you for joining us—and enjoy the day!

With warm regards,  
The Oklahoma Microscopy Society Organizing Committee

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# Welcome

This year's Spring Meeting brings together a diverse array of microscopy-focused research from across Oklahoma and beyond. Our invited speakers represent the cutting edge of imaging science, featuring experts in artificial intelligence-driven S/TEM analysis, high-resolution cryo-electron microscopy of protein-nucleic acid interactions, CRISPR-Cas system structure and fidelity, X-ray microanalysis, and electron energy loss spectroscopy (EELS).

A major focus of this year's event is the next generation of instrumentation at the University of Oklahoma, including the **JOEL NeoARM STEM**—a state-of-the-art aberration-corrected scanning transmission electron microscope (arriving soon)—and the newly installed **ThermoFisher FEI Tundra Cryo-TEM**, an essential tool for cryo-EM grid screening and single-particle analysis. Many of our sessions are designed to engage and inform current and future users of these facilities, highlighting their research applications and potential for interdisciplinary collaboration across the state.

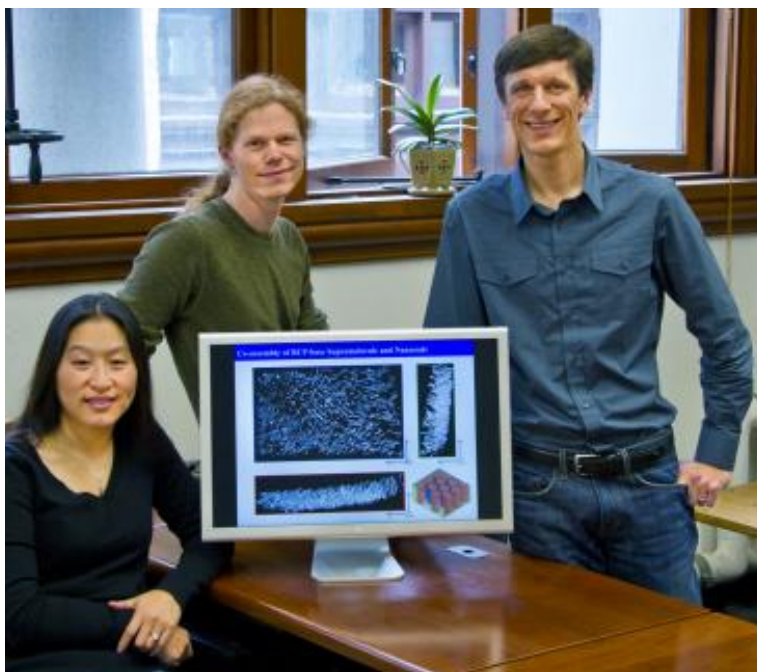
## Speakers

Keynote Speaker: Dr. Peter Ercius

**Dr. Peter Ercius**

**Staff Scientist, National Center for Electron Microscopy (NCEM)  
Lawrence Berkeley National Laboratory**

Dr. Peter Ercius is a leading expert in electron tomography and atomic-resolution imaging. He earned his B.S. and Ph.D. in Applied and Engineering Physics from Cornell University, focusing his doctoral research on 3D electron tomography of semiconductor devices using scanning transmission electron microscopy (STEM). Currently, Dr. Ercius leads the electron tomography program at NCEM, overseeing the dual aberration-corrected TEAM 0.5 microscope. His research encompasses atomic-scale materials characterization, 4D-STEM scanning diffraction, in situ liquid TEM, electron energy loss spectroscopy (EELS), and automated experimentation. His work has significantly advanced the field of nanoscale imaging, including the first 3D image of atoms in an amorphous solid and real-time visualization of atomic nucleation processes.



### **Dr. Len Thomas**

#### **Facility Director, Biomolecular Structure Core University of Oklahoma**

Dr. Len Thomas is a research scientist and facility director at the University of Oklahoma, where he oversees the Biomolecular Structure Core (BSC). In this role, he manages advanced cryo-electron microscopy (cryo-EM) instrumentation, including the ThermoFisher FEI Tundra 100kV Cryo-TEM, which is pivotal for single-particle cryo-EM grid analysis and screening. Dr. Thomas's work supports a wide range of structural biology research, facilitating high-resolution imaging of macromolecular structures. His expertise ensures that researchers have access to state-of-the-art equipment and methodologies for their investigations.

### **Dr. Rakhi Rajan**

#### **Associate Professor, Department of Chemistry & Biochemistry University of Oklahoma**

Dr. Rakhi Rajan is a structural biologist whose research focuses on the molecular mechanisms of CRISPR-Cas systems, the adaptive immune systems in bacteria and archaea. Her laboratory investigates protein-nucleic acid interactions, particularly those involving Cas9 and Cas12a proteins, which are pivotal in gene editing technologies. By employing techniques such as X-ray crystallography, molecular biology, and bioinformatics, Dr. Rajan aims to elucidate the atomic-level interactions and conformational changes of Cas protein-nucleic acid complexes. Her work not only advances our understanding of CRISPR-Cas systems but also contributes to the development of high-fidelity genome editing tools and diagnostics.

Dr. Rajan earned her B.S. from Kerala Agricultural University, her Ph.D. from Ohio State University, and completed postdoctoral research at Northwestern University. She currently serves as an Associate Professor at the University of Oklahoma.

### **Dr. Ritesh Sachan**

#### **Assistant Professor Oklahoma State University**

Dr. Ritesh Sachan is an Assistant Professor in the School of Mechanical and Aerospace Engineering at Oklahoma State University, where he leads the Nanofusion Laboratory. His research focuses on nanomaterials, surface engineering, and atomic-scale characterization using advanced techniques such as 4D-STEM and EELS. With degrees from IIT Varanasi, KIST, and the University of Tennessee, Dr. Sachan has earned numerous accolades including the NSF CAREER Award and the R&D 100 Award. His recent efforts have helped bring Oklahoma its first aberration-corrected electron microscope, advancing the state's nanoscience capabilities.

**Dr. Iman Ghamarian**

**Assistant Professor**

**University of Oklahoma**

Dr. Iman Ghamarian is an Assistant Professor in the School of Aerospace and Mechanical Engineering at the University of Oklahoma, where he leads the Accelerated Materials Development Lab (AMDL). His research focuses on understanding and optimizing the processing-structure-property relationships in structural materials, with a strong emphasis on additive manufacturing, advanced microscopy, and machine learning. A recipient of the SAMPE Outstanding Paper Award and a DURIP grant for electron microscopy instrumentation, Dr. Ghamarian is advancing materials science through high-throughput methods and multiscale modeling.

 [Scan for Details / Registration](#)



## Schedule Format

Time	Description / Who	Title / notes
7:15 - 8:30 AM	Room open for setup (food, vendors, and posters etc)	
8:30 - 9:00 AM	Continental breakfast, registration, visit with vendor tables, poster setup	
9:00 - 9:30 AM	Opening remarks	Dr. Tingting Gu (OU, SRNML, OMS); Dr. Ann West (OU, OVPRP); Mason Rhue (OU, MSA student council)
9:30 - 10:00 AM	Dr. Rakhi Rajan	Understanding the DNA Cleavage Fidelity Mechanisms of CRISPR-Cas Nucleases Using Cryo-EM
10:00 - 10:30 AM	Dr. Len Thomas	New and Unique to Oklahoma: Tundra Cryo-TEM for Your Protein Structure Needs
10:30 - 10:45 AM	Dr. Sonika Robertson (Oxford)	Pushing boundaries in X-ray microanalysis with technological advancements
10:45 - 11:00 AM	John Haritos (Ametek)	Clarity Direct Detection EBSD for low kV and beam current EBSD analysis

11:00 – 12:15 PM	<b>Timpano Award talks with student presenters</b>	
11:00 - 11:15 AM	Donny Han	The Molecular and Physiological Mechanisms of Electric Signal Elaboration During Early Development in the South American Weakly Electric Fish, <i>Brachyhypopomus gauderio</i>
11:15 - 11:30 AM	Sooraj Patel	Towards grain boundary engineering of Protonic Ceramic Cell Electrolytes using Orientation-Microscopy Assisted Grain Boundary Character Distribution Analysis
11:30 - 11:45 AM	Itunu Apalara	Understanding the Impact of a Modern Euxinic Spring on Sediment Using Bulk Sediment and Smectite Clay-Preserved Proxies
11:45 - noon	Chhandosee Ganguly	Structural insights into a high-fidelity CRISPR-Cas12a variant revealed using optimized graphene oxide cryo-EM grids
Noon – 12:15 PM	Vikas Reddy Paduri	Bimetallic AgCo Nanoparticle Synthesis via Combinatorial Nanosecond Laser-Induced Dewetting of Thin Films
12:15 - 12:45 PM	lunch	Lunch will be included in meeting registration, also the Cross Center restaurants are directly across Asp Street
12:45 - 1:30 PM	Tours	Choose between Tundra cryo-TEM, NeoARM aberration-corrected STEM, or main SRNML core facility
1:30 - 2:00 PM	Student/researcher poster session, networking and vendor tables	
2:00 - 3:00 PM	<b>Plenary Keynote:</b> Dr. Peter Ercius (LBNL)	Imaging Atomic Structure, Strain, and Disorder By Atomic Electron Tomography
3:00 - 3:30 PM	Dr. Ritech Sachan (OSU)	Unveiling Swift Heavy Ion Track Morphology in Sr-Based High-Entropy Perovskites
3:30 - 4:00 PM	Dr. Iman Ghamarian (OU)	Machine Learning-Assisted Quantitative Microscopy to Accelerate Microstructure-Centric Design and Manufacturing
4:00 - 4:15 PM	Break	
4:15 - 4:30 PM	Awards and closing	

## Abstracts:

## KEYNOTE SPEAKER

### **Imaging Atomic Structure, Strain, and Disorder By Atomic Electron Tomography**

*Peter Ercius*

National Center for Electron Microscopy, Molecular Foundry, Lawrence Berkeley National Laboratory

Knowledge of the atomic structure of materials is critical to understanding their functionality in many fields such as biology, microelectronics, condensed matter, and nanotechnology. Scanning transmission electron microscopy (STEM) is an effective method for measuring the atomic structure of materials based on so-called Z-contrast where the image intensities are proportional to the atomic number of the material being imaged. However, by its nature STEM is limited to producing only two-dimensional projections of a structure. Electron tomography is a technique that can reconstruct the three-dimensional structure of unique nanoscale objects from a series of two-dimensional images acquired at different viewing angles. Utilizing the sub-Angstrom real-space resolution of STEM with advanced tomographic reconstruction algorithms we have extended tomography to atomic resolution to solve the structure of materials at the single atom level without averaging or the assumption of crystallinity. This talk will present the abilities of AET to reconstruct order and disorder in materials with 20 picometer precision. Our success in resolving chemical order/disorder in metallic FePt nanoparticles was extended to include the time domain by capturing nucleation and growth of ordered phases in the same nanoparticle. Also, the ability to reconstruct atomic structure without the need for averaging or crystallinity led to the achievement of directly imaging three-dimensional atomic arrangements in amorphous solids. AET has become an important technique in the field of atomic structural characterization.

## Understanding the impact of a modern euxinic spring on sediment using bulk sediment and smectite clay-preserved proxies

*Itunu T. Apalara<sup>1\*</sup>, Preston R. Larson<sup>2</sup>, Julian E. Sabisch<sup>2</sup>, Caitlin Hodges<sup>1</sup>, Megan E. Elwood Madden<sup>1</sup>, Andrew S. Elwood Madden<sup>1</sup>*

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Modern euxinic (sulfidic and anoxic) environments are useful proxies for early Earth. Therefore, we sought to understand the morphological and chemical proxies preserved in sediments impacted by a modern euxinic spring as compared to adjacent unimpacted sediments. Two sets of samples were studied: euxinic-spring-impacted creek bank sediment (ICS) and unimpacted creek bank sediment (UCS) at the surface, and euxinic-spring-impacted soil (ISL) and unimpacted soil (USL) samples at 220 cm depth. We used X-ray diffraction (XRD) to analyze the mineralogy of the bulk sediment and isolated clay fraction. We further examined clay nanoparticles using Scanning/Transmission Electron Microscopy (S/TEM) and Energy-Dispersive X-ray spectroscopy (EDX) to study clay morphology and chemistry. The bulk mineralogy of the impacted sediments includes pyrite and barite in addition to plagioclase, quartz, calcite, and K-feldspar, which were present in the unimpacted sediment. However, dolomite was absent only in ICS, while gypsum and ankerite were present in ISL only. While the sediments currently in contact with the euxinic water are much darker and appear to have a texture indicating they are richer in clays compared to the unimpacted sediments, the XRD results of clay-oriented mounts show the impacted and the unimpacted clay fractions are both comprised of smectite, chlorite, kaolinite, illite, smectite/chlorite mixed layer clays. We chose to focus additional analyses on smectite clay nanoparticles because of their known potential for redox reactivity; TEM images of these smectites reveal platy and cornflake textured smectites in all the samples. However, EDX results reveal differences between the smectites in the impacted and unimpacted samples; unimpacted smectites have Si, Al, Fe, Mg, whereas impacted smectites additionally have S, Ba, and Sr. The unimpacted smectites contain Ca and K as interlayer cations while Ca, K, and Na, with traces of Ba are seen in the impacted smectites. Furthermore, STEM-EDX shows evidence of nanoscale barite and pyrite precipitation on the impacted smectites. These nanoprecipitates were also supplied by the euxinic spring. Ultimately, this study demonstrates that (1) sulfide and sulfate minerals can co-exist in modern euxinic sediments whether in contact with the atmosphere or not and (2) smectite nanoparticles primarily respond to euxinic conditions by immobilizing cations by adsorption or cation exchange rather than significantly changing their octahedral sheet cation composition.

## **Glaucoma-induced Blood-Retinal Barrier Leakage by Transcytosis**

*Jennifer D. Ballheim<sup>1</sup>, Chi Zhang<sup>3,4</sup>, Marina Simón<sup>3,4</sup>, Haeyn Lim<sup>3,4</sup>, Michael H. Elliott<sup>1,2</sup>, Simon W.M. John<sup>3,4</sup>*

<sup>1</sup>Department of Biochemistry & Physiology, University of Oklahoma Health Sciences Center, Oklahoma City

<sup>2</sup>Department of Ophthalmology, Dean A. McGee Eye Institute, University of Oklahoma Health Sciences Center, Oklahoma City

<sup>3</sup>Department of Ophthalmology, Vagelos College of Physicians and Surgeons, Columbia University Irving Medical Center, New York

<sup>4</sup>Mortimer B. Zuckerman Mind Brain Behavior Institute, Columbia University, New York

**Purpose:** Glaucoma is a leading cause of blindness worldwide, with vision loss resulting from intraocular pressure (IOP)-induced damage to optic nerve cells. Recent studies have shown that elevated IOP disrupts venous blood-retinal barrier (BRB) in peripheral venules across three distinct glaucoma mouse models, contributing to optic nerve/retinal ganglion cell degeneration (Zhang et al., 2024). Whether IOP-induced BRB leakage occurs through junctional, transcellular, or both leakage pathways is unclear. To investigate this, we used transmission electron microscopy (TEM) to examine the tight junctions and transcellular vesicles in peripheral retinal venules.

**Methods:** DBA/2J mice, a glaucoma model with ocular hypertension, and strain and age matched normotensive control mice (D2-Gpnmb+) were tail-vein injected with horseradish peroxidase (HRP). After 30 minutes of HRP circulation, the retinas were dissected, fixed, and incubated with 3-3' diaminobenzidine (DAB) to visualize the retina vasculature. Peripheral venules were dissected, dehydrated, and embedded in Eponate/Araldite epoxy resin. Ultrathin (100nm) sections containing the venule diameter were imaged at 7,000× magnification using a Hitachi H7600 TEM at 100 kV with an AMT Nanosprint 1200 camera. As a positive control for junctional leakage, wild-type mice were intravitreally injected with lipopolysaccharide (LPS) and processed using the same HRP/TEM method.

**Results:** The number of vesicles per  $\mu\text{m}^2$  was significantly increased in DBA/2J mice compared to controls ( $n=11$ , Welch's t-test,  $p=0.005$ ) indicating increased transcytosis. Tight junctions appeared normal and intact in both hypertensive and control mice ( $n>50$  tight junctions), while ~60% of junctions in LPS-treated mice exhibited HRP leakage ( $n=41$  junctions).

**Conclusion:** These results indicate that BRB leakage in DBA/2J glaucoma occurs via increased transcytosis rather than junctional leakage. This provides new insight into the retinal vascular dysfunction mechanisms that contribute to vision loss in glaucoma.

Chi Zhang, Haeyn Lim\*, Jennifer D. Ballheim\*, Marina Simón\*, Rui Fu, Nicholas G. Tolman, Logan Horbal, Felicia A. Juarez, Qing Wang, Aakriti Bhandari, Christa Montgomery, Ling Zhu, Jonathan G. Crowston, Nicolas Robine, Michael P. Fautsch, Michael H. Elliott, Simon W.M. John. "IOP-induced blood-retinal barrier compromise contributes to RGC death in glaucoma." Manuscript submitted for publication. \*Authors contributed equally.



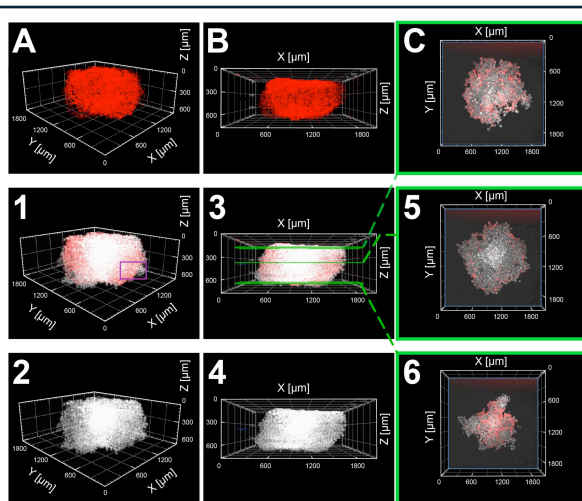
## Visualizing Nanoparticle Spatiotemporal Distributions in Entire Tumor Spheroids Using Expansion Microscopy

Connor Baroody<sup>1</sup>, Vinit Sheth<sup>1</sup>, Reto Fiolka<sup>2</sup>, Kevin M. Dean<sup>2</sup>, Stefan Wilhelm<sup>1</sup>

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<sup>2</sup>Lyda Hill Department of Bioinformatics, The University of Texas Southwestern Medical Center, Dallas, Texas, 75390, USA

Microscopy techniques are widely used for studying the spatiotemporal distributions of nanoparticles across tumor microenvironments towards the development of safer and more efficacious anti-cancer nanomedicine formulations. However, conventional microscopy techniques have several limitations that prevent an in-depth analysis of the interactions between nanoparticles and cancer cells. For example, light microscopy methods lack sufficient spatial resolution to fully visualize nanoparticles intracellular trafficking mechanisms, while electron microscopy methods cannot easily image large three-dimensional (3D) volumes. Here, we apply expansion microscopy (ExM), a hydrogel-based, 3D super-resolution light microscopy method to visualize the distribution of gold nanoparticles throughout entire cultured cancer spheroids with confocal laser scanning microscopy (CLSM) and axially swept light-sheet microscopy (ASLM). We further integrated this method with light scattering imaging to achieve label-free visualizations of intracellular gold nanoparticles to avoid potential experimental confounds from the addition of labels on the surface of the nanoparticles (**Figure 1**). This method provides a robust tool that allows for studying nanoparticle-cellular interactions at high resolutions and in large 3D volumes. The successful application of this method will empower more informed research into cancer nanomedicine for the development of safer and more efficacious cancer therapies.



**Figure 1. Combining Axially Swept Light-sheet (ASLM) imaging and label-free imaging of AuNPs within a mouse tumor cell spheroid (4T1).** Red represents the AuNP light-scattering signal, and white represents the bulk stain (PAN) NHS ester 488. **(A)** Whole spheroid imaging using an ASLM, 3x magnification, viewed from a 45-degree angle and **(B)** from the side view. A1 and B3 show the merged image of the two channels. (C-C6) Sectional slices shown of the spheroid from the bottom, middle, and top with both channels present (merged). The relative locations of the slices can be seen coming from B3. The software Arivis was used to generate the renderings

## **Sample Preparation Solutions Induce Nanoparticle Formation, Deposition, and Grid Contamination in Transmission Electron Microscopy Analysis**

*Em G. Elder<sup>1</sup>, Katherine A. Sluder<sup>1</sup>, Preston R. Larson<sup>2</sup>, Julian E.C. Sabisch<sup>2</sup>, Andrew S. Elwood Madden<sup>1</sup>*

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Extraction of nanoparticles from soil samples often involves partial digestion of the material with a range of acidic, basic, redox-reactive, rinsing, and/or surface-active solutions. Our initial study found that the contact of soil digest solutions with the Transmission Electron Microscopy (TEM) grids led to contamination and sample preparation artifacts. The question was then raised about the susceptibility of different types of TEM grids to different solutions commonly used in sample preparation that may be causing results not indicative of the sample, but of the procedure. The primary goal of this research is to document and observe how different solutions used in these processes may create nanoparticles and other solution precipitates. We investigated how different solutions react with different grid materials through Scanning Electron Microscopy (SEM) and Energy-Dispersive X-Ray Spectroscopy (EDS). Ultimately, we aim to create a point of reference to help prevent the contamination of future samples.

We considered how exposure time, concentration, type of solution, and exposure method may affect the creation of nanoparticles. We began by testing the effects of eight different solutions on copper, molybdenum, and gold grids for three different reaction times. SEM and EDS results showed that the mere exposure of the grids to solutions during routine sample preparation procedures did generate particulate matter on the grid. Different solutions and times produced different sizes, shapes, and amounts of material; however, EDS spectra revealed that the particles and precipitates typically consisted of solution + grid material + elements associated with the production of the grid or the SEM itself. On the other hand, spectra from different solution + grid combinations were all unique, suggesting there are different mechanisms at play in each. In addition to the grid experiments, we also tested how copper and gold nanoparticles reacted when deposited onto grids using five different solutions. Through these trials, we were able to observe how the depositing solution affects clumping, particle size, and the chemical make-up of standardized nanoparticles. Going forward, we will continue to explore the connections between TEM grids and sample preparation solutions through TEM analysis, hoping to uncover the different mechanisms that are leading to these reactions.

## **Structural insights into a high-fidelity CRISPR-Cas12a variant revealed using optimized graphene oxide cryo-EM grids**

*Chhandosee Ganguly, Swarmistha Aribam, Leonard Thomas, Rakhi Rajan*

Department of Chemistry and Biochemistry, University of Oklahoma

CRISPR-Cas12a has become a widely used tool for genome editing and molecular diagnostics. However, unintended off-target DNA cleavage by Cas12a remains a key limitation. To address this, our lab engineered a Cas12a variant by modifying the bridge helix—a structurally important element—resulting in markedly reduced off-target activity.

To understand the structural basis of this enhanced specificity, we used cryo-electron microscopy (cryo-EM) to determine high-resolution structures of the variant. Initial grid preparation presented challenges in sample distribution and reproducibility. We overcame these by deriving a reproducible method for preparing graphene oxide (GO) coated Quantifoil holey carbon copper grids to achieve sufficient area covered with GO monolayer. These GO-coated grids significantly improved particle density within the holes while maintaining optimal ice thickness. This protocol may benefit other cryo-EM studies involving difficult samples. The resulting structures reveal novel conformational features of the bridge helix variant, offering mechanistic insights into how DNA cleavage specificity is modulated. These findings not only advance our understanding of Cas12a function but also provide a structural framework for rational design of next-generation genome editing tools with improved precision due to their reduced off-target effects.

## **Machine Learning-Assisted Quantitative Microscopy to Accelerate Microstructure-Centric Design and Manufacturing**

*Iman Ghamarian*

Department of Aerospace and Mechanical Engineering, University of Oklahoma

Advanced characterization techniques, such as scanning transmission electron microscopy, are critical for developing and optimizing materials used across diverse sectors like energy, aerospace, and healthcare. These techniques enable comprehensive mapping of vast material regions, resulting in massive datasets containing rich composition, crystallographic, and microstructural information. However, extracting meaningful insights from such extensive datasets poses significant analytical challenges. Leveraging advanced data analytics and machine learning algorithms becomes essential to distill critical structural information efficiently and accurately from the collected microscopy data. In this presentation, I will discuss integrating machine learning methods with orientation microscopy to address these analytical challenges. First, I will demonstrate how statistical analyses of grain boundary distributions can inform the enhancement of proton conductivity in protonic ceramic fuel cells. I will present a generative machine learning approach for diffraction pattern denoising, significantly improving data interpretability and reliability. Finally, I will illustrate the application of graph neural networks for accurate crystal structure identification from complex diffraction patterns. These examples highlight how machine learning-assisted quantitative microscopy can drive the accelerated development of materials with targeted microstructural properties.

## **The molecular and physiological mechanisms of electric signal elaboration during early development in the South American weakly electric fish, *Brachyhypopomus gauderio***

Donglin Han and Michael Markham

Cellular and Behavioral Neurobiology Program, School of Biological Science, University of Oklahoma, Norman, OK

Active sensory and communication systems such as echolocation and electrolocation allow animals to successfully inhabit visually unfavorable environments that are inaccessible to visually guided competitors. These active sensory and communication signals, however, are detectable by predators, thereby creating predation risks associated with active sensing. Many active-sensing species exhibit behavioral and physiological adaptations to conceal their sensory signals from predators, but the degree of signal concealment often varies based on social/environmental conditions and sometimes changes during development.

Weakly electric fish evolved an active sensing method of generating electric organ discharges (EODs) from an electric organ (EO) to enable navigation and communication in dark and turbid waters. The electric fields created by the EOD are detected by electroreceptors on the skin and central neural circuits analyze distortions of the electric fields in response to nearby objects or specifics to serve navigation and communication, respectively. Many fish that do not generate electric signals also possess electroreceptors that detect low-frequency, low-amplitude electric energy.

In all Gymnotiform fish studied to date, juveniles produce monophasic EODs that transition to more complex EOD waveforms as the fish mature (Carmpton et al. 2011). The cellular and physiological mechanisms that cause these developmental changes in EOD waveform are still not known. The South American weakly electric fish *Brachyhypopomus gauderio* demonstrates an EOD waveform that changes from monophasic to biphasic in early development, with important implications for predation risk because monophasic EODs have significant low-frequency signal energy that is detectable by electroreceptive predators while biphasic EODs suppress this low-frequency component of the signal energy. *B. gauderio* is the only Gymnotiform reliably bred in captivity, and therefore serves as an accessible model to investigate developmental changes in the EOD waveform.

Gymnotiform EOD waveforms are determined by the morphology and ion channel expression patterns in the electric organ cells (electrocytes). It is known that biphasic EODs are produced by electrocytes that produce two sequential action potentials on the opposing posterior and anterior electrocyte membranes, whereas in many species with monophasic EODs, only the posterior electrocyte membrane produces an action potential. The transition from monophasic EODs to biphasic EODs could be accomplished by initiating action potentials on the anterior membrane of electrocytes and coordinating action potentials on the posterior membrane of electrocytes, **but this possibility has not been directly investigated.**

In this research, we will test the following three different aims. **Aim 1:** Determine of distribution of voltage-gated sodium channels on the electrocyte membrane and the morphology of electrocytes during the change from monophasic to biphasic EODs. **Aim 2:** Determine the activation stages and movements of voltage-gated sodium channels (Nav) on the membrane of electrocytes during the change from monophasic to biphasic EODs. **Aim 3:** Determine the mechanism of modulating the firing time intervals between two sides of the membrane in electrocytes. Currently results indicate

that the electric organ developed and assembled prior to EOD generation, around 5-6 days after hatching; there are no voltage-gated sodium channels transported to the membrane of electric organs when fish start to generate EODs around 9 days after hatched. Moreover, most voltage-gated sodium channels are located in the lumen of electrocytes at same time. Roughly 25 days after hatched, a few voltage-gated sodium channels are transported to the membrane of electrocytes (Figure 1.).

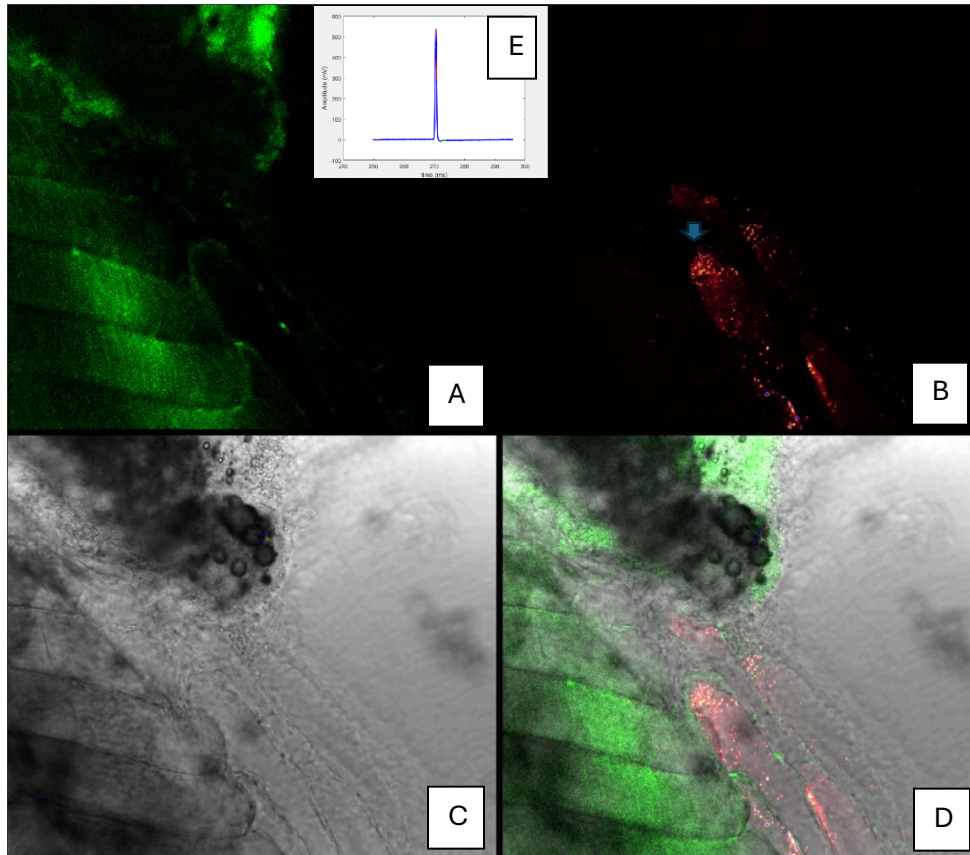


Figure 1. The location of voltage-gated sodium channels on electric organ membrane of a 25 days old larval *Brachyhypopomus gauderio* (A indicates outlets of electric organ cells; B indicates voltage-gated sodium channels labeled by Anti-pan antibodies; C indicates transmitting light image of larval fish; D indicates overlays of all A, B, C images; E indicates EOD waveform recording of these 25 days old larval fish).

## **Determining the structure of CRISPR type II-A spacer acquisition**

*Kole Long<sup>1</sup>, Swarmistha Aribam<sup>1</sup>, Saadi Rostami<sup>1</sup>, Chhandosee Ganguly<sup>1</sup>, Nathan Burrows<sup>2</sup>, Oliver Wu<sup>1</sup>, Rakhi Rajan<sup>1</sup>*

<sup>1</sup>Department of Chemistry and Biochemistry, The University of Oklahoma, <sup>2</sup>Stanford-SLAC CryoEM Center (S<sup>2</sup>C<sup>2</sup>), Division of CryoEM and Bioimaging, Stanford Synchrotron Radiation Light source, SLAC National Accelerator Laboratory, Menlo Park, CA United States

CRISPR-Cas systems provide adaptive immunity in prokaryotes by capturing and integrating fragments of foreign DNA, known as spacers, into the host genome. This process, called spacer integration, is catalyzed by the Cas1-Cas2 complex during the adaptation phase. This study focuses on the structural characterization of Cas1-Cas2 complexes from Type II-A CRISPR systems using cryo-electron microscopy (cryo-EM). Preliminary 3D reconstructions of the Cas1-Cas2-DNA complex have been obtained; however, the quality and resolution of these reconstructions are currently limited by particle heterogeneity. Variability analysis and 3D classification reveal distinct conformational states, suggesting the presence of mobile domains within the complex. Addressing this heterogeneity is crucial for achieving high-resolution structural models. A multifaceted strategy is being employed to improve complex homogeneity, including the use of freshly purified protein, immediate grid preparation following size exclusion chromatography, testing of cross-linking conditions, and optimization of grid-coating techniques. In parallel, advanced data processing techniques such as improved 3D classification and refinement strategies are being explored to better resolve structural variability. These efforts aim to capture stable conformations of the Cas1-Cas2-DNA complex, enabling detailed mechanistic insights into spacer integration. Ultimately, this work will enhance our understanding of the molecular basis of CRISPR adaptation and inform future applications in genome engineering.

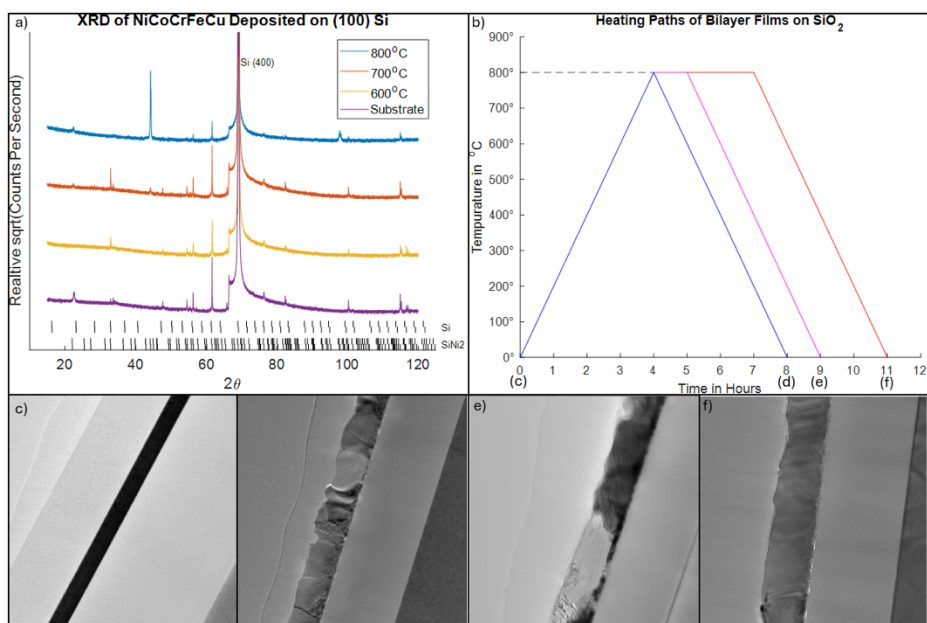
## Thermal Annealing-Driven Multi-Principal Element Silicide Thin Film Formation

Joshua Marvin, Lance Speigel, Dr. Ritesh Sachan

*Department of Mechanical and Aerospace Engineering, Oklahoma State University*

Multi-Principal Elemental Silicides (MPESs) are a novel family of ceramics being explored as environmentally stable, radiation-resistant thermal barrier coatings in critical applications for aircraft engines and extraplanetary devices. These materials consist of multiple elements substituted into the same lattice site of a parent structure; however the synthesis remains a critical challenge due to their complex structures. In the present study, thin films of MPESs are prepared by pulsed laser deposition of alloy thin films onto a (100) Si single Crystal substrate using a NiCoCrFeCu target under high vacuum at temperatures ranging from ambient to 800°C. These films are compared using X-Ray diffraction to observe the formation of distinct crystal structures and at 800°C strong peaks matching a Ni silicide structure were observed. However, these films are observed to delaminate from the substrate with extended annealing. Therefore, to accomplish the synthesis, MPES thin films are prepared by depositing bilayers of NiCoCrFeCu and Si onto an inert amorphous SiO<sub>2</sub> substrate. These samples are subsequently annealed under vacuum in a tube furnace at 800°C for different lengths of time. These samples are characterized using scanning electron microscopy and atomic force microscopy for investigating the surface morphology and transmission Electron microscope (TEM) to cross-sectionally understand the silicide formation, crystal structure and elemental distribution as a function of annealing time and temperature. The imaging, diffraction and elemental mapping are conducted in TEM with energy dispersive x-ray spectroscopy (EDS), selected area electron diffraction (SAED), electron energy-loss spectroscopy (EELS), and high-resolution scanning transmission electron microscopy (STEM). The bright-field TEM images obtained from each film show a progression in the reaction of the Si layer with the underlying metallic layer seeming to be consumed after 3 hours held at 800°C with a reduction in thickness correlating to an increase in density. In the case of the film as it was deposited the layers are entirely distinct giving a near perfect starting point for the reactions study. The images obtained from each film show no reaction with the

SiO<sub>2</sub> substrate nor any delamination enforcing the utility of this method for the study of this system. HR-STEM and EDS results after 3 Hours at 800°C reveal that Ni, Co, and Fe are forming a silicide phase with Cu concentrated at the boundaries of the grains while a Cr rich phase is distinctly separating and forming a silicide with the parent structure of CrSi<sub>2</sub>.





## Investigation of Airborne Particulate Matter from 4<sup>th</sup> of July Fireworks in Norman, OK Using Drone-Based Sampling

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Fireworks are a core component of celebrations and festivals across the world and have been used for hundreds of years in the commemoration of these special events. While fireworks bring joy and excitement to those in attendance, they are also a well-known pollution emitter. Fireworks mainly contribute to heightened levels of airborne particulate matter (PM) pollution as well as volatile organic compounds (VOCs) and other atmospheric pollutants. PM pollutants are a major concern for members of the population with preexisting health conditions or considered to be at risk for medical episodes.

Past research into the pollutants generated by large scale firework use has been centered around ground-based techniques including passive samplers, optical measuring tools, and pumping systems with filters used to trap particulates. To potentially gain a novel understanding of these pollutants, a drone-based sampling approach was developed and used to sample particulate matter from a fireworks display hosted in Norman, OK for the 4<sup>th</sup> of July (Independence Day) celebrations. Samples consisted of aluminum disks with double sided carbon tape as the method for trapping particulate matter as the drone was held within the plume of smoke generated from the fireworks display. Samples were then transferred to a Thermo-Fisher Scientific Quattro E-SEM for in-depth analysis.

SEM analysis revealed diverse physical structure and elemental makeup across the samples, with trends and differences noted between samples taken at lower altitudes (LAS) and those taken at higher altitudes (HAS). Lower altitude samples were, on average, smaller and more chemically diverse than higher altitude samples. LAS particulates ranged from “rocky” shape structures to spherical ones while HAS particulates were almost exclusively spherical in structure and made of various metals that are used in fireworks to create the effects they are so well known for. Through this analysis, a proposed mechanism of formation for these particulates has been created and, in the future, it can be used to better understand the behavior of these pollutants.

*\*\* We gratefully acknowledge the support for this work from the National Science Foundation REU-Site Award Number: 2150365.*

# Bimetallic AgCo Nanoparticle Synthesis via Combinatorial Nanosecond Laser-Induced Dewetting of Thin Films

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Herein, a combinatorial approach is developed to conduct high-throughput studies on the nanosecond pulsed laser-induced dewetting phenomenon of bilayer Ag–Co metallic thin films. Laser irradiation results in the spontaneous rupture of these films in nanosecond timescale, forming bimetallic nanoparticles (NPs) through intermediate stages of hole formation and bicontinuous nanostructures. This approach utilizes bilayer thin films with thickness gradients in both Ag and Co layers (referred to as bigradient samples) while maintaining a constant overall thickness. The laser irradiation on such bigradient bilayer films facilitates control on Ag and Co ratio in the thin films, thus enabling material libraries of Ag–Co NPs covering a large compositional variation. The evolution of NPs with a correlation between NP diameter and interparticle spacings is further studied. The study reveals monotonic increase in NP size and interparticle spacing in Co/Ag bilayer arrangement, while an increase and subsequent decrease in the NP size is observed at 50% Ag in Ag/Co bigradient films. A transition from intermediate stage hole formation to bicontinuous nanostructures with changing composition is also observed. These changes in intermediate stage morphologies and dewetting mechanism are attributed to variation of the free energy of the bilayer system dominated by intermolecular interaction forces.

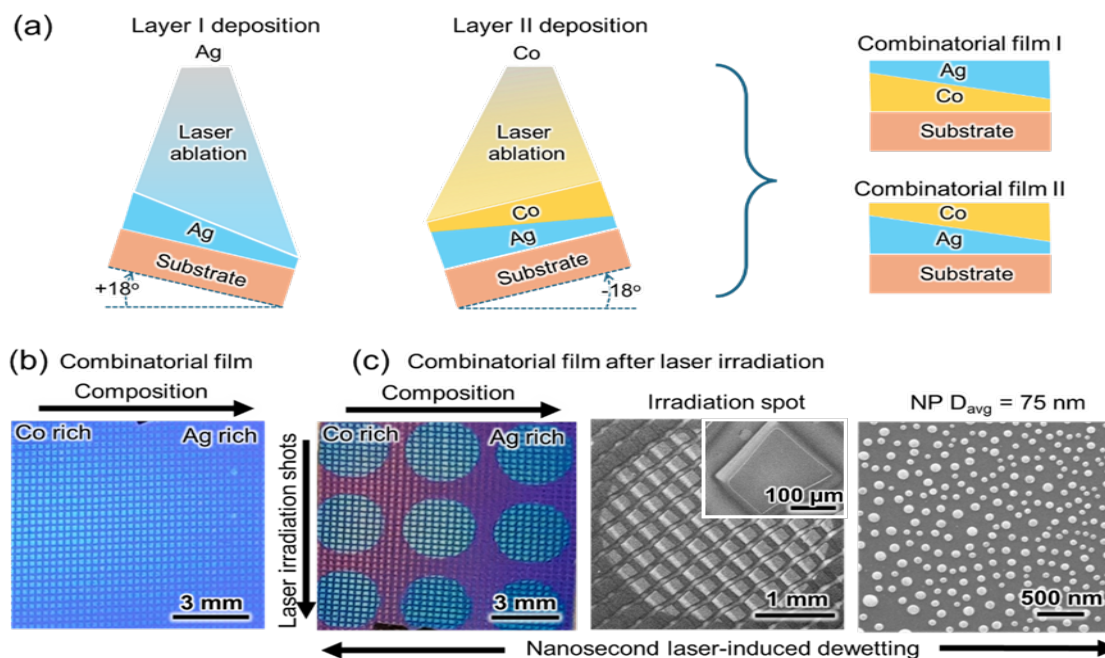


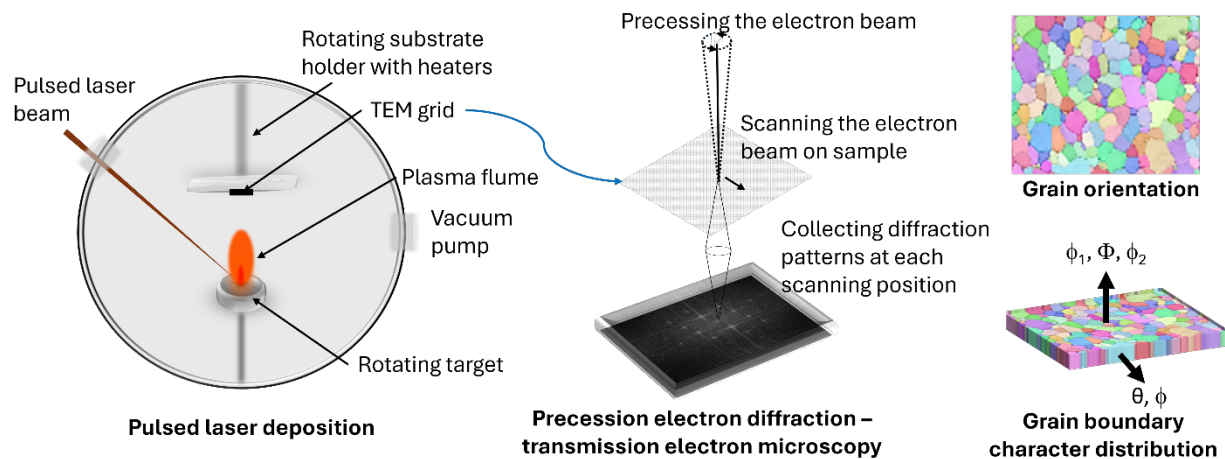
Figure 1. a) Schematic of the experimental setup showing combinatorial bigradient thin-film generation with a tilt-enabled substrate holder, b) a representative optical image of sample with bilayer islands after the deposition, and c) optical image with a matrix of laser irradiation spots showing Co-rich to Ag-rich regions; an SEM image showing a single-irradiation spot of 3mm diameter (inset showing a single nano island of 250 x 250 μm) and magnified SEM image illustrating final NPs after complete PLiD.

## Towards Grain Boundary Engineering of Protonic Ceramic Cell Electrolytes using Orientation-Microscopy Assisted Grain Boundary Character Distribution Analysis

Sooraj Patel, Iman Ghamarian

Accelerated Materials Development Lab, School of Aerospace and Mechanical Engineering, University of Oklahoma

Global demand for sustainable energy solutions has elevated interest in protonic ceramic electrochemical cells (PCECs) as next-generation devices. This work focuses on grain boundary engineering to optimize ionic conductivity in  $\text{BaCe}_{0.4}\text{Zr}_{0.4}\text{Y}_{0.1}\text{Yb}_{0.1}\text{O}_{3-\delta}$  (BCZYYb) thin films, a promising PCEC electrolyte. In this work, nanocrystalline BCZYYb thin films with columnar morphology are synthesized using pulsed laser deposition. The five-parameter grain boundary character distribution is evaluated based on orientation data acquired through precession electron diffraction in transmission electron microscopy. The study identifies dominant grain boundary orientations and population densities of grain boundary plane normal for specific misorientations. The findings lay the groundwork for designing high-performance, durable electrolytes by addressing a non-uniform grain boundary character distribution. This research aims to accelerate the development of efficient electrochemical devices, contributing to clean energy generation and supporting the broader transition to sustainable power solutions. It fosters new pathways for tailoring grain boundary structures, enabling conduction enhancement for future clean energy applications.



*Experimental workflow illustrating PCEC electrolyte thin film deposition, orientation mapping using precession electron diffraction – transmission electron microscopy, and subsequent grain boundary character distribution analysis.*

## Understanding the DNA Cleavage Fidelity Mechanisms of CRISPR-Cas Nucleases Using Cryo-EM

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Cas12a, a protein belonging to the CRISPR-Cas system, uses the sequence complementarity of RNAs called as CRISPR-RNAs (crRNAs) to target DNA and introduce sequence-specific double-stranded DNA cleavage. This property has been harnessed for genome applications and molecular diagnostics. Yet, it suffers from off-target DNA cleavage, where DNA with partial complementarity with the crRNA are also cleaved, along with the desired on-target DNA cleavage. This compromises the safety and sensitivity of Cas12a-based tools. Our lab has been focusing on protein engineering to remove off-target DNA cleavage by Cas12a.

Cas12a protein has a bi-lobed architecture and the two lobes are connected by a conserved, arginine-rich helix called the bridge helix (BH). Previous studies from our lab had shown that amino acid substitutions that impairs the loop-to-helical transition of the BH improve the DNA cleavage fidelity of Cas12a. Here, we used cryo electron microscopy (cryo-EM) and biochemistry to decipher the mechanism of BH-mediated off-target discrimination of Cas12a by comparing the effects of deformity introduced by proline substitutions versus stability imparted by alanine substitutions within the BH. The results show that proline substitutions in the BH impairs helical transitioning of the BH, which in turn compromises the structural stability of the RNA-DNA hybrid thereby making the proline variant very sensitive to off-target DNA cleavage. Biochemical studies show that switching proline with alanine substitutions increases the off-target DNA cleavage, potentially due to the stability offered to the BH by alanine substitutions. BH-mediated DNA cleavage selectivity can be broadly applied to other BH-containing Cas enzymes (e.g., Cas9) and arginine-rich helix containing RNA-binding proteins.

## Unveiling Swift Heavy Ion Track Morphology in Sr-Based High-Entropy Perovskites

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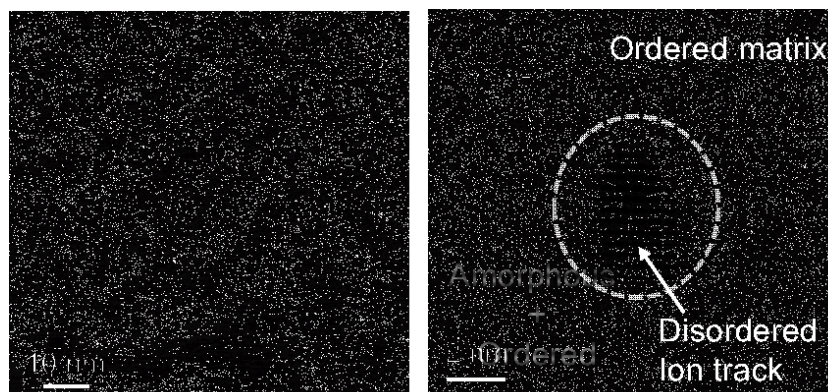
<sup>7</sup>Department of Mechanical and Materials Engineering, Queen's University, Kingston Ontario, Canada

High entropy oxides (HEOs) have attracted significant scientific interest due to their compositional complexities, which can unlock novel or enhanced properties. The incorporation of multiple elements into a cation lattice site in HEOs influences atomic-level responses to energetic ion irradiation, though these effects remain poorly understood.

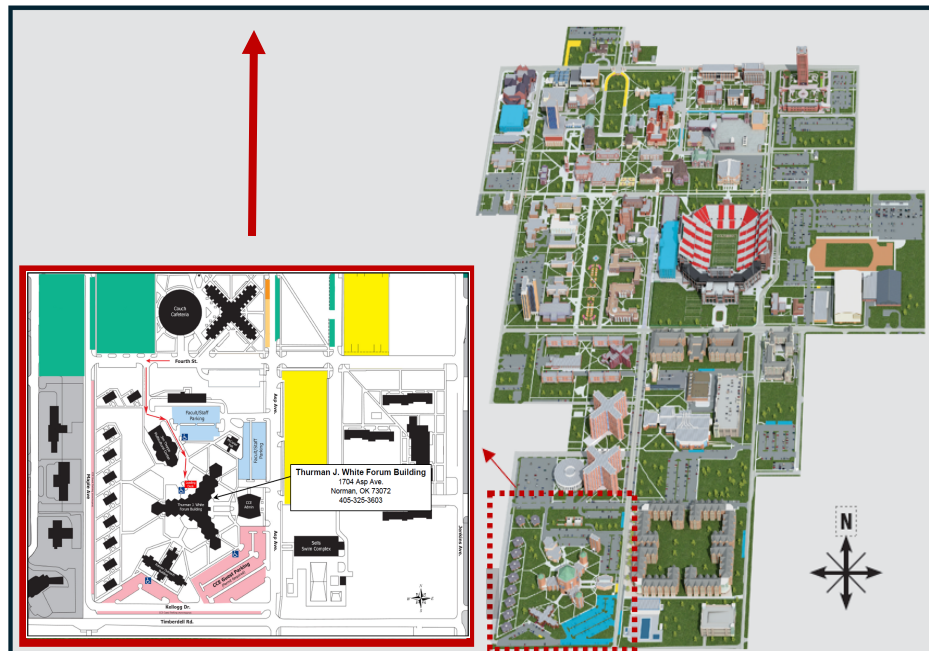
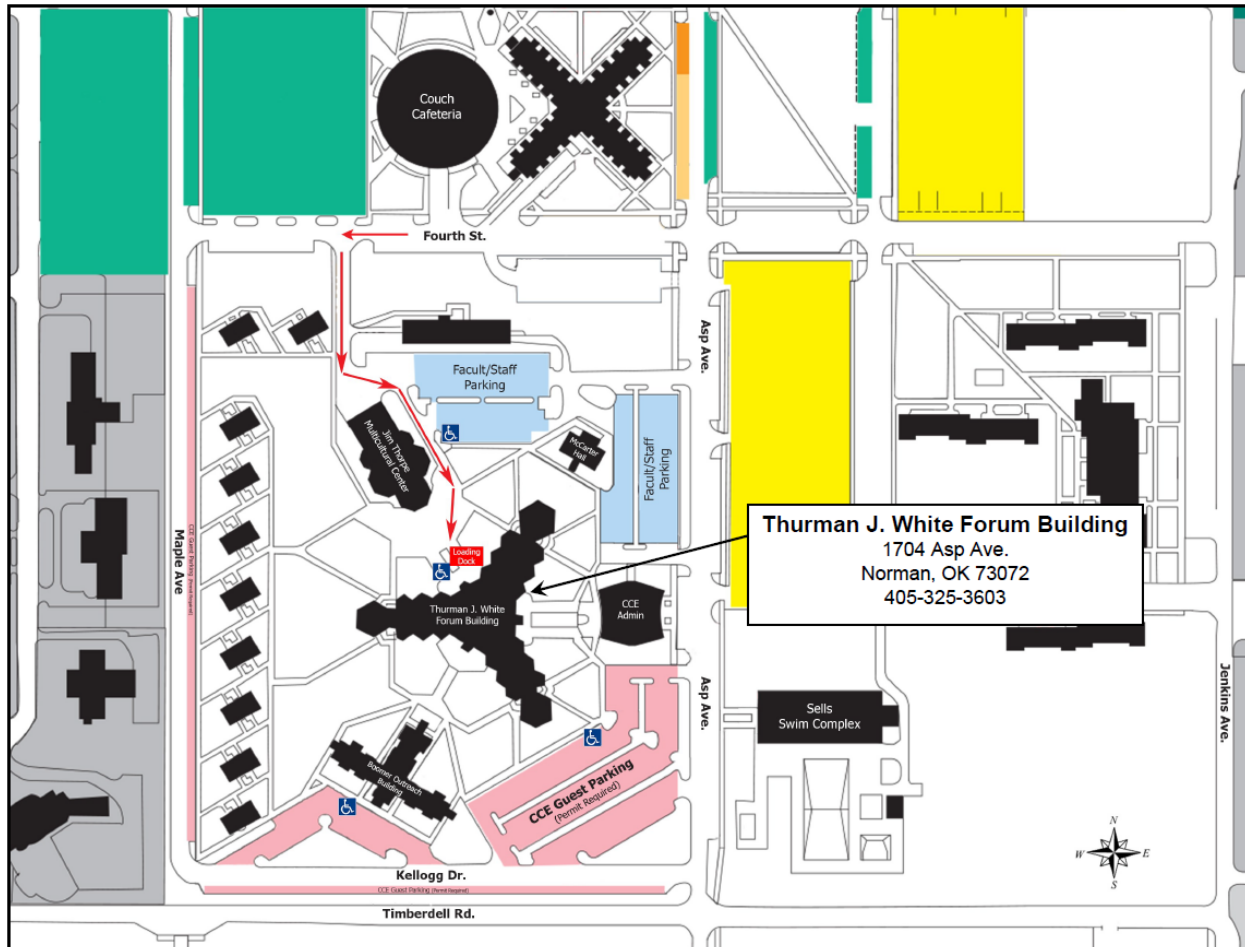
This study investigates nanoscale phase transitions in perovskite-structured HEO  $\text{Sr}(\text{Zr}_{0.2}\text{Sn}_{0.2}\text{Ti}_{0.2}\text{Hf}_{0.2}\text{Nb}_{0.2})\text{O}_3$  under swift heavy ion irradiation (774 MeV Xe). Unlike typical amorphous ion-tracks, Sr-HEO exhibits discontinuous and partially recrystallized tracks. The presence of multiple cations at the B-site reduces ion track diameters compared to  $\text{SrTiO}_3$  under the same energy-loss conditions. Additionally, while  $\text{SrTiO}_3$  tracks display a core-shell structure with a disordered interface, Sr-HEO tracks exhibit minimal lattice distortion, restricted within  $\sim 2\text{--}3$  monolayers.

Atomic-resolution observations reveal that the amorphous phase in Sr-HEO remains stable under electron irradiation, unlike  $\text{SrTiO}_3$ , which readily recrystallizes. This stability is attributed to structural complexities resisting recrystallization, outweighing electron-induced excitations and local heating effects that facilitate defect migration in  $\text{SrTiO}_3$ .

Theoretical calculations using the inelastic thermal spike model offer insights into the differing track formation mechanisms in these materials, emphasizing the role of compositional disorder in modifying irradiation responses.



## Campus Map:





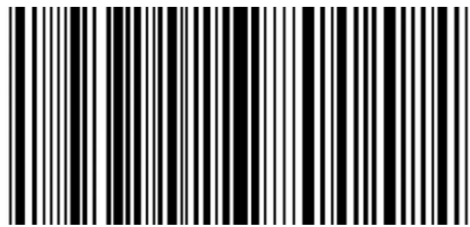


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