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Princeton-Nature Conference

4th Frontiers in Electron and Scanning Probe Microscopy for the Physical and Life Sciences

October 19-21, 2026

Princeton University, New Jersey

Program and Abstracts

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4th Frontiers in Electron and Scanning Probe Microscopy for the Physical and Life Sciences

October 19-21, 2026

Princeton University
Maeder Hall
86 Olden Street
Princeton, NJ 08544

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Welcome

Dear Colleagues and Friends,

We are pleased to welcome you to the 2026 Nature Conference, "**Frontiers in Electron and Scanning Probe Microscopy for the Physical and Life Sciences**." The fourth conference in this series, initiated in 2018 through a partnership between Princeton University and Nature, continues our shared goal of bringing together leading scientists and emerging researchers across disciplines to explore new frontiers in imaging, analysis, and discovery.

This year marks an important expansion of the conference. In addition to electron microscopy, we are extending the scientific program to include scanning probe microscopy, bringing together two powerful and complementary families of techniques for observing, measuring, and manipulating matter at increasingly small scales. By connecting these communities with researchers in the physical and life sciences, we hope to encourage new interactions, exchange ideas across traditional disciplinary boundaries, and inspire approaches that might not emerge within any single field.

Microscopy has profoundly changed the way we see and understand the world. Electron microscopy has advanced from early imaging of materials and biological structures to atomic-resolution characterization, cryogenic imaging, tomography, spectroscopy, in situ observation, and increasingly automated and AI-assisted experimentation. At the same time, scanning probe microscopy has enabled researchers not only to visualize surfaces with atomic and molecular precision, but also to probe electronic structure, measure forces and quantum phenomena, and manipulate individual atoms and molecules.

These developments are increasingly interconnected. Advances in instrumentation, computation, artificial intelligence, sample preparation, and data analysis are creating opportunities that cross conventional boundaries between materials science, physics, chemistry, biology, engineering, and medicine. Bringing these perspectives together is therefore especially timely. We believe that some of the most exciting advances will emerge where different methods, disciplines, and scientific communities meet. Over the three days of this conference, the program will highlight important developments in cryo-electron microscopy and tomography, artificial intelligence and computational microscopy, scanning probe microscopy, and advanced electron microscopy, together with emerging applications across the physical and life sciences. Invited and keynote presentations will be complemented by flash talks, poster sessions, discussions with Nature Portfolio editors, career conversations, and informal opportunities for scientific exchange.

An equally important purpose of this conference is to strengthen our research community. We hope that established investigators and early-career scientists alike will use this opportunity to exchange knowledge, challenge existing ideas, discover complementary expertise, and begin collaborations that extend well beyond these three days. The history of microscopy repeatedly demonstrates that advances in one field can transform another when knowledge and technologies are shared openly. We are delighted to welcome you to Princeton and deeply grateful to our speakers, participants, scientific committee, Nature Portfolio colleagues, sponsors, and everyone whose efforts have made this conference possible.

We hope that the 2026 conference will be scientifically rewarding, intellectually stimulating, and personally enjoyable - and that the discussions and connections formed here will help shape the next generation of imaging and analysis. Welcome to Princeton, and to the 2026 Nature Conference.

Conference Co-Chairs, on behalf of the Scientific Committee,

Nan Yao, Princeton University **Daniel McNally**, Nature Research

General Information

Conference Name Badges

You must wear your name badge at all times for the entire duration of the conference. If you misplace yours, please go to the registration desk.

Connecting to WiFi

Princeton University provides wireless internet service to campus visitors through either PUVisitor or Eduroam. Visitors who are in the Eduroam community should connect to Eduroam. All other visitors should use the visitor wireless service.

How to connect to PUVisitor

When you first attempt to connect, you will be directed to register your device:

1. Enter your name. Your mobile number is optional.
2. Enter a valid email address to which we can send your password.
3. Read and accept the terms of use.

Scientific Session Protocol

Photography, audio or video recording by audience members of the scientific sessions is not permitted.

Our conferences have an opt-out social media policy. If a speaker or presenter opts out, requesting to keep their presentation closed and in confidence to the conference attendees, honor their request and do not post or share information from their presentation.

Also note that abstracts may not be re-printed, and poster presentations are also considered personal communications. Information in these may not be publicly shared without prior permission of the presenter.

Remember to turn off cell phones or set to vibrate or silent during the general sessions.

Filming

There will be a film crew from Nature Conferences taking footage throughout the duration of the program. If you do not wish to be recorded or photographed, please see Colleen Seaver at the registration desk.

E-Program Book

This program book is available as an e-book. There will be a limited quantity of printed copies onsite. The most updated version can always be found on the conference app, Whova, under the Documents section.

Poster Session

The poster session will be taking place the evening of **Tuesday, October 20th from 5:00 – 6:30 p.m. ET at the Friend Center Convocation Room.**

Board number assignments appear in the Poster Presenter List section of this book. Please check the Whova app for the final list.

Poster presenters can hang their posters starting at 3:00 p.m. on Tuesday. Please do not bring your posters before Tuesday, as we will be unable to store them for you. Boards will be numbered and thumbtacks will be available. Posters must be taken down at the close of the poster session or else they will be discarded.

Conference App - Whova

All attendees have access to the conference app called Whova. You should have received an email with instructions for downloading and creating your account. If you did not receive the email or need us to resend it, please email colleen.seaver@springernature.com.

At the registration desk, there will be QR codes posted, which link to the Whova app. You will be able to check in yourself and then take your name badge.

Certificate of Attendance

If you need a Certificate of Attendance – or a Certificate of Poster or Flash Talk Presentation, please email colleen.seaver@springernature.com after the conference's conclusion. Please note that only presenting authors of a poster are provided with a Certificate of Poster Presentation.

Dietary Requests

We can only accommodate requests communicated to us in advance of the conference. Last-minute requests cannot be accommodated onsite.


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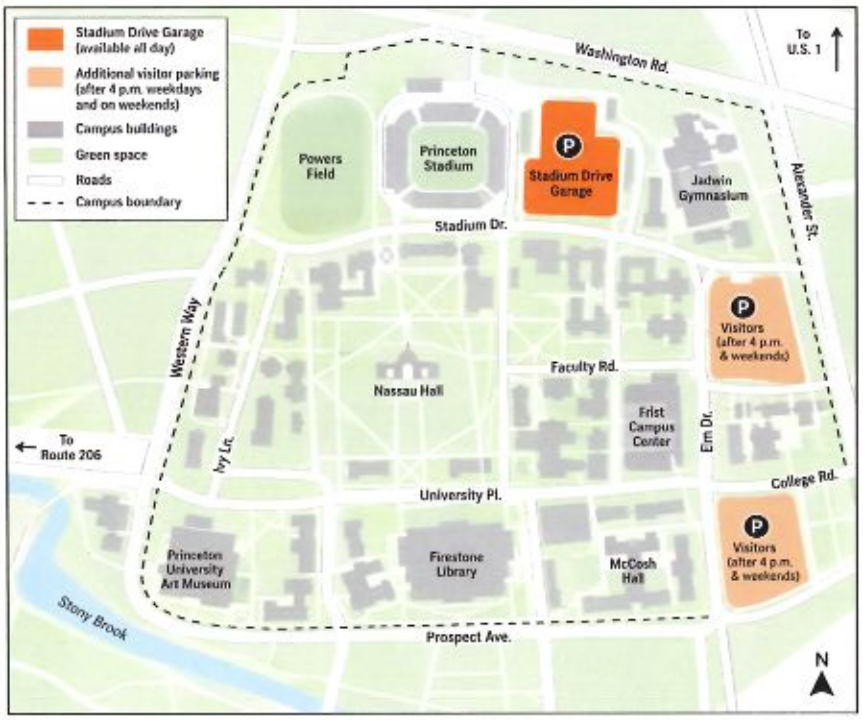
Visitors must register their vehicle before arrival.

Daily permits are free.

Scan to register.

WHERE TO PARK

- Visitors may park in Stadium Drive Garage all day.
- Additional parking (highlighted) is available after 4 p.m. on weekdays and on weekends.
- For overnight parking, contact The Service Point, 609.258.8300, thepoint@princeton.edu.



PRINCETON UNIVERSITY — A HIGHER PURPOSE

Conference Program

DAY 1

Monday, October 19, 2026

8:00 – 8:50 a.m.
Registration check-in, and Breakfast

8:50 – 9:00 a.m.
Opening Remarks
Andrew Houck, School of Engineering & Applied Science, Princeton University
Richard A. Register, Princeton Materials Institute, Princeton University

SESSION I: **Cryo-EM**
Chaired by Allison Doerr, Nature Methods

9:00 – 10:00 a.m.
KEYNOTE: Making sense of disappearing densities in single particle cryo-EM
Yifan Cheng – University of California San Francisco/Howard Hughes Medical Institute, USA

10:00 – 10:30 a.m.
Atomistic Cryo-EM Structure Determination of Small RNA Guests by Leveraging the Symmetry of an Oligomeric Host
Adrian Ferré-D'Amaré– National Institutes of Health, USA

10:30 – 11:00 a.m.
Coffee Break

11:00 – 11:30 a.m.
New frontiers for cryo-EM in energy and health
Yuzhang Li – University of California Los Angeles, USA

11:30 a.m. – 12:30 p.m.
Meet the Nature Editors

Kevin Cannon – Associate Editor, Nature Communications
Stacey Chin – Senior Editor, Nature Materials
Allison Doerr – Chief Editor, Nature Methods
Zachary Mansley – Associate Editor, Nature Communications
Benjamin Martindale – Senior Editor, Nature Catalysis
Florian Ullrich – Senior Editor, Nature

Moderator: **Daniel McNally** – Senior Editor, Nature Materials

12:30 – 2:00 p.m.
Lunch

SESSION II: **AI**
Chaired by Benjamin Martindale, Nature Catalysis

2:00 – 2:30 p.m.
Self-Driving Microscopy and What It Should Answer
Ju Li (Massachusetts Institute of Technology, USA)

2:30 – 3:00 p.m.
Fluxionality And Functionality: Probing Atomic-Level Spatiotemporal Surface Dynamics In Catalytic Nanoparticles
Peter Crozier (Arizona State University, USA)

3:00 – 3:30 p.m.
From Ex Situ to Operando: The role of Electron Microscopy in Accelerating Battery Innovation
Zhao Liu (Thermo Fisher Scientific, USA)

3:30 – 4:00 p.m.
Coffee Break

4:00 – 4:30 p.m.
AI Methods for High-Throughput Biomolecular Structure Determination in Cryo-EM
Ellen Zhong (Princeton University, USA)

4:30 – 5:00 p.m.
Building a fully automated cryo-EM data processing pipeline with foundation models
Huaizong Shen (Shenzhen Medical Academy of Research and Translation (SMART), China)

5:00 – 6:30 p.m.
Welcome Reception

DAY 2

Tuesday, October 20, 2026

8:00 – 9:00 a.m.
Registration check-in, and Breakfast

SESSION III: Scanning Probe
Chaired by Daniel McNally, Nature Materials

9:00 – 10:00 a.m.
Keynote: Quantum sensing of quantum matter
Amir Yacoby (Harvard University, USA)

10:00 – 10:30 a.m.
Scanning SQUID microscopy
Kathryn Ann Moler (Stanford University, USA)

10:30 – 11:00 a.m.
Coffee Break

11:00 – 11:30 a.m.
40 Years of Atomic Force Microscopy – Seeing Dents in the Atom
Franz Giessibl (University of Regensburg, Germany)

11:30 a.m. – 12:30 p.m.
Flash Talks
Flash talks will be chosen from our pool of abstract submissions and will be 3-5 minutes in length.

12:30 – 2:00 p.m.
Lunch

SESSION IV: Scanning EM
Chaired by Stacey Chin (Nature Materials)

2:00 – 2:30 p.m.
High-Throughput Electron Tomography for Brain Mapping
Aaron Kuan (Yale University, USA)

2:30 – 3:00 p.m.
Scanning Nanodiffraction of Beam-sensitive Materials
Karen Bustillo (Lawrence Berkeley National Laboratory, USA)

3:00 – 3:30 p.m.
A High-Throughput Approach to Identify and Characterize Molecular Glues Stabilizing WEE1-MG-CRBN-DDB1 Complex
Wenfei Song (Thermo Fisher Scientific, USA)

3:30 – 4:00 p.m.
Coffee Break

4:00 – 4:30 p.m.
Shelly Conroy (Imperial College London, England)

4:30 – 5:00 p.m.
In-situ (scanning) transmission electron microscopy to understand electrocatalyst materials
Drew Higgins (McMaster University, Canada)

5:00 – 6:30 p.m.
POSTER SESSION & RECEPTION

DAY 3

Wednesday, October 21, 2026

8:00 – 9:00 a.m.

Registration check-in, and Breakfast

SESSION V: Cryo-ET

Chaired by Florian Ullrich (Nature)

9:00 – 10:00 a.m.

Keynote: Electron Videography of Soft and Energy Matter at the Nanoscale

Qian Chen (University of Illinois at Urbana-Champaign, USA)

10:00 – 10:30 a.m.

Structural Biology of Membrane Fission

John Jimah (Princeton University, USA)

10:30 – 11:00 a.m.

Coffee Break

11:00 – 11:30 a.m.

Benchmark Specimens for Quantitative Evaluation of the CryoET Pipeline, from Data Collection to Subtomogram Averaging

Reza Paraan (Biohub, USA)

11:30 a.m. – 12:30 p.m.

Career Conversations: Where Microscopy Can Take You

Panelists:

Tim Maitland (Senior Sales Account Manager, Life Sciences Electron Microscopy, Thermo Fisher Scientific)

Paul P. Shao (IAC Senior Research Specialist, Princeton University)

Qian Chen (Professor, Department of Materials Science and Engineering, University of Illinois Urbana-Champaign)

Allison Doerr (Chief Editor, Nature Methods)

Moderator:

Daniel McNally, (Senior Editor, Nature Materials)

12:30 – 2:00 p.m.

Closing Remarks & Lunch

Nan Yao (Imaging and Analysis Center, Princeton University, USA)

Speaker Bios

KAREN BUSTILLO **Lawrence Berkeley National Laboratory, USA**



Karen Bustillo is a staff member at the National Center for Electron Microscopy (NCEM), Molecular Foundry, Lawrence Berkeley National Laboratory in Berkeley, California. At NCEM, Karen collaborates with scientists worldwide through the U. S. Department of Energy Molecular Foundry User Program to characterize materials with transmission electron microscopy. Dr. Bustillo's research interests include developing advanced TEM techniques for imaging electron beam-sensitive materials such as polymers, organic small molecules, framework materials, and battery materials. She also has expertise in situ experiments including cryo, heating, mechanical testing, liquid cell, and gas cell. She pursues characterization of nanostructures using the spectroscopy capabilities of the TEM especially with respect to elemental quantification and data analysis.

QIAN CHEN **University of Illinois at Urbana-Champaign, USA**



Professor, Department of Materials Science and Engineering, Department of Chemistry (Affiliated), Department of Chemical and Biomolecular Engineering, Materials Research Laboratory (Affiliated), Beckman Institute for Advanced Science and Technology (Affiliated), Carle Illinois College of Medicine (Affiliated), Carl R. Woese Institute for Genomic Biology (Affiliated), and CZ Biohub Chicago Investigator

YIFAN CHENG **University of California San Francisco and Howard Hughes Medical Institute, USA**



Dr. Yifan Cheng is currently an Investigator of Howard Hughes Medical Institute and a Professor of Biochemistry and Biophysics at University of California San Francisco (UCSF). He received his Ph.D. degree in 1991 from Institute of Physics, Chinese Academy of Sciences (CAS). From 1991 to 1996, he continued his research in solid state physics and electron microscopy as a postdoctoral fellow at University of Oslo (NTNF Fellow) and Max-Planck-Institute of Metal Research (Alexander von Humboldt Fellow). In 1996, he changed his research direction to structural biology, and received further training in cryo-electron microscopy (cryo-EM) from Professors Kenneth Taylor at Florida State University and Yoshinori Fujiyoshi at Kyoto University. In 1999, he joined the laboratory of Thomas Walz to setup a cryo-EM operation at Harvard Medical School. He started his independent career in 2006, joining the faculty of UCSF and has stayed there ever since. He has been an HHMI Investigator since 2015. He is the recipient of the Christian B. Anfinsen Award from The Protein Society (2018) and Anatrase Membrane Protein Award from Biophysical Society (2026), elected members of the American Academy of Arts and Science (2019) and National Academy of Sciences (2020).

His laboratory uses cryo-EM to study structures of biological macromolecules, particularly integral membrane proteins and dynamic complexes. In addition, development of cryo-EM methodology for structural biology is also a long-lasting interest of his laboratory. Previous works of his laboratory include developments of algorithms to correct electron beam-induced image motion and determination of the first atomic structure of TRPV1 by single particle cryo-EM, etc.

SHELLY CONROY **Imperial College London, England**



Dr. Shelly Conroy is Associate Professor in Functional Thin Films and Microscopy, Royal Society Tata University Research Fellow, and Principal Investigator of the ERC Consolidator Grant DISCO. Her research focuses on the atomic scale design and characterisation of functional materials for semiconductor, quantum, and energy applications. Dr. Conroy is Co-Director of the CDT in Materials 4.0 and Royce Imperial Academic Champion for Thin Film Growth. She holds a joint appointment at Tyndall National Institute as a Principal Researcher and is a Mercator Fellow at Friedrich Alexander Universität Erlangen Nürnberg. She also serves as Leader of the Focused Interest Groups of the Microscopy Society of America. Dr. Conroy previously worked at Pacific Northwest National Laboratory as a permanent staff scientist before joining the University of Limerick as a Science Foundation Ireland Analog Devices Research Fellow and principal investigator. She holds a PhD in AlN thin film growth for optoelectronic device applications and in situ TEM from Tyndall National Institute and University College Cork, Ireland. Her PhD was funded by a Science Foundation Ireland PhD scholarship and seed funded by Intel Ireland.

PETER CROZIER **Arizona State University, USA**



Peter A. Crozier is a Professor of Materials in the School for Engineering of Matter, Transport and Energy at Arizona State University. He has extensive experience in characterizing ceramics, metal nanoparticles and catalytic materials for applications related to energy and the environment. He has expertize in developing and applying advanced techniques of transmission electron microscopy, including in situ and operando methods to problems in ion conductors and catalytic materials. He is also applying electron energy-loss spectroscopy to determine the optical and vibrational properties of nanomaterials. Over the last 5 years, he has explored the effectiveness of various deep learning models for removing noise in electron images, spectra and diffraction patterns. He is a Fellow of the Microscopy Society of America and also a member of the Materials Research Society, the North American Catalysis Society and the American Ceramics Society. He served as the President of the Microscopy Society of America. He has organized numerous workshops, schools and symposia and has published over 200 archival journal papers and book articles.

ADRIAN FERRÉ-D'AMARÉ **National Institutes of Health, USA**



Adrian R. Ferré-D'Amaré graduated from the Instituto Tecnológico y de Estudios Superiores de Monterrey in Mexico in 1990 with a B.S. in chemistry. He earned a Ph.D. in molecular biophysics from The Rockefeller University in 1995. He was a Jane Coffin Childs postdoctoral fellow at Yale University from 1995 to 1999. Dr. Ferré-D'Amaré was a Member of the Fred Hutchinson Cancer Research Center and an Affiliate Professor of Biochemistry at the University of Washington, Seattle, from 1999 to 2011. He was also an Investigator of the Howard Hughes Medical Institute (HHMI) from 2008 to 2011. He resigned from HHMI and joined the NHLBI in 2011 as a Senior Investigator and Chief of the Laboratory of RNA Biophysics and Cellular Physiology. Dr. Ferré-D'Amaré was a Rita Allen Foundation Scholar from 2001 to 2004, received the Eli Lilly and Company Research Award from the American Society for Microbiology in 2004 and was a Distinguished Young Scholar in Medical Research of the W.M. Keck Foundation from 2003 to 2008. Dr. Ferré-D'Amaré has authored or coauthored more than 100 papers. He is on the editorial board of the journal RNA and has served on many review panels for the National Institutes of Health, the National Science Foundation, as well as foreign and international scientific organizations.

FRANZ GIESSIBL **University of Regensburg, Germany**



Franz J. Giessibl holds the chair for quantum nanoscience at the University of Regensburg in Germany and is a research professor at Kanazawa University in Japan. He is a pioneer in atomic force microscopy (AFM) who started as the PhD student of Nobel Laureate Gerd Binnig at the IBM Physics Group Munich at LMU in 1988. In his PhD thesis, he built the first AFM for low temperatures and ultrahigh vacuum (UHV) together with Gerd Binnig and Christoph Gerber and obtained true atomic resolution on an ionic crystal. After that, he joined an AFM manufacturer in Silicon Valley, designed and built a UHV AFM and launched the field of atomically resolving noncontact AFM by demonstrating true atomic resolution on the 7x7 reconstruction of silicon for the first time. In 1995, he joined McKinsey as a management consultant and invented the qPlus sensor, a novel core for the AFM, in his home laboratory on weekends. He joined Augsburg University in 1997 where he obtained his habilitation in 2001. An extended version of his habilitation thesis was published in Reviews of Modern Physics in 2003 with currently more than 3500 citations – it is generally considered one of the foundational review papers in AFM.

In 2005, he received offers for chairs at the University of Bristol and University of Regensburg, joining the latter in 2006. Since then, he did key AFM experiments from atomic manipulation, subatomic spatial resolution, artificial atoms, topological insulators and others. Important results were also obtained in collaborations with the IBM labs in Almaden and Zurich, NIST in Gaithersburg, the National University of Singapore, Kanazawa University and others. His main invention, the qPlus sensor, is standard today for low temperature AFMs, it is used by seven international AFM manufacturers and operates in more than 500 low temperature AFMs globally today. In 2026, the AFM market leader launched an AFM that uses the qPlus sensor in ambient conditions. Franz J. Giessibl is a Fellow of the American Physical Society, the European Academy for Sciences and Arts in Salzburg, the European Academy of Sciences in Brussels, the Japanese Vacuum- and Surface Science Society and a distinguished Fellow of the International Engineering and Technology Institute. He has won several international science prizes, including the Feynman Prize on Nanoscience, the Heinrich Rohrer Grand Medal, the NIMS Award, the APS Keithley Prize and the MRS Innovation in Materials Characterization Award. He also worked with an eminent visual artist as published in his 2022 book "First View Inside an Atom. Encounters with Gerhard Richter between Art and Science". In 2026, his new book "Atomic Force Microscopy and Atomic Manipulation. An Inquiry into Matter" will appear with Springer Nature.

DREW HIGGINS **McMaster University, Canada**



Drew is an Associate Professor and Tier 2 Canada Research Chair in the Department of Chemical Engineering at McMaster University, where their research focuses on sustainable electrochemical energy technology development. Particularly, their team focuses on applications including electrochemical CO2 conversion, rechargeable zinc-ion batteries, supercapacitors, water electrolyzers and biomass valorization processes. Their research covers the synthesis of new nanostructured electrode materials and their integration into electrochemical technologies, as well as developing advanced understanding of material performance through the development and implementation of in situ characterization techniques. Previously Drew completed their PhD in Chemical Engineering at the University of Waterloo in 2015, with their thesis involving the synthesis, characterization, and device integration of nanostructured oxygen reduction catalysts for low temperature fuel cells. During this time, they spent approximately one year at the Los Alamos National Laboratory working. In 2015, Drew started a Banting Postdoctoral Fellowship at Stanford University in the Department of Chemical Engineering and in 2017 was promoted to an Associate Staff Scientist at Stanford University / SLAC National Accelerator Laboratory, where they oversaw research activities focusing on discovering and understanding new electrocatalyst compositions and structures for a variety of important electrochemical reactions, including water oxidation, CO2 reduction, oxygen reduction and methane activation. More details are available at: https://www.higginslab.com/.

JOHN JIMAH **Princeton University, USA**



John Jimah is an Assistant Professor of Molecular Biology at Princeton University. His laboratory investigates the molecular mechanisms of membrane remodeling in human cells and eukaryotic parasites, using cryo-electron microscopy and tomography (cryo-EM and cryo-ET) alongside biochemical and cell biological approaches.

He has contributed to our understanding of the structural mechanisms by which membrane architecture is maintained or remodeled, including how human dynamin assembles to drive membrane fission during endocytosis, how dynamin organizes actin during cell-cell fusion, and how the bacterial β-barrel assembly machinery mediates outer membrane protein folding. He also studied the malaria parasite protein CelTOS, a multistage vaccine target, and how it disrupts host membranes to enable parasite traversal through host cells. Dr. Jimah earned his B.A. in Molecular Biology from Colgate University and his Ph.D. in Biology and Biomedical Sciences from Washington University in St. Louis, and he completed postdoctoral training at the National Institutes of Health. His work has been recognized by the Alfred P. Sloan Research Fellowship and the NIGMS MOSAIC K99/R00 award. Overall, his research seeks to define structural principles of membrane remodeling across diverse cellular contexts.

AARON KUAN **Yale University, USA**



Aaron Kuan investigates how circuit connectivity within and between brain areas supports a diverse array of functions. His lab aims to apply circuit-mapping techniques at brain-wide scales to understand how brain wiring underlies cognition and neurological diseases. Kuan develops new electron microscopy and X-ray imaging techniques to enable 3D imaging of neuronal circuits with unprecedented resolution and scale and combines these approaches with behavior and functional imaging to reveal how circuit connectivity underlies brain activity and, ultimately, cognition. The long-term research goal is to extend high-resolution circuit mapping techniques to brain-wide scales to unite "top-down" and "bottom-up" views of brain anatomy and function.

JU LI **Massachusetts Institute of Technology, USA**



Ju Li is Carl Richard Soderberg Professor of Power Engineering at MIT Department of Nuclear Science and Engineering and Materials Science and Engineering, as well as a core faculty member of MIT Schwarzman College of Computing (CCSE). Previously he has held faculty positions at the Ohio State University, and the University of Pennsylvania. His group (http://Li.mit.edu) investigates the mechanical, electrochemical and transport behaviors of materials as well as novel means of energy storage and conversion.

Ju was elected Fellow of the American Physical Society in 2014, a Fellow of the Materials Research Society in 2017 and a Fellow of AAAS in 2020. Li is the chief organizer of MIT A+B Applied Energy Symposia that aim to develop solutions to global climate change challenges with "A-Action before 2040" and "B-Beyond 2040" technologies.

YUZHANG LI **University of California, Los Angeles, USA**



Yuzhang Li is an Associate Professor in Chemical and Biomolecular Engineering at UCLA. He received his bachelor's in Chemical Engineering from UC Berkeley and his Ph.D. in Materials Science and Engineering from Stanford University. The long-term goal of the Li group@UCLA is to invent new tools and materials that address important challenges in sustainability and health. For example, we are leading efforts to leverage the powerful cryogenic electron microscopy (cryo-EM) tool to address grand challenges in sustainability. These efforts have led to significant breakthroughs in our understanding of batteries (Science 358, 506, 2017; Science 375, 66, 2022) and electrocatalysts (Nature Energy 8, 138, 2023), which represent important clean energy technologies necessary for securing energy resilience and security. New insights then inform parallel efforts in materials innovations (Nature 620, 86, 2023; Nature Energy 10, 502, 2025) that will enable transformative technologies. Yuzhang's research has been highlighted by news media including Forbes, Popular Mechanics, and ABC7 Bay Area, while also being recognized with several awards, including the Packard Fellowship, Dreyfus Teacher-Scholar Award, Forbes 30 Under 30, and young investigator awards from the NSF, DOE, NIH, DOD, ACS, and ECS.

ZHAO LIU **Thermo Fisher Scientific, USA**



Zhao Liu is a Senior Market Development Manager at Thermo Fisher Scientific, based in Oregon, USA, specializing in the battery market. He earned his PhD in Materials Science and Engineering from Northwestern University and later joined Thermo Fisher Scientific. He also serves as an Affiliated Professor in the Materials Science & Engineering Department at the University of Washington, helping strengthen academia–industry collaboration. His work focuses on advancing battery characterization metrology across R&D and manufacturing, addressing analytical challenges, and driving practical solutions through strategic partnerships with academic and industrial stakeholders.

KATHRYN ANN MOLER **Stanford University, USA**



Kathryn A. "Kam" Moler is the Marvin Chodorow Professor at Stanford University, where she previously served as Vice Provost and Dean of Research from 2018 to 2023 and currently serves as Vice President for SLAC. A physicist with both a B.S. and Ph.D. from Stanford, her work has been recognized by her election to the National Academy of Sciences and as a Fellow of the American Physical Society. Her other notable honors include the Presidential Early Career Award for Scientists and Engineers (PECASE) and the William L. McMillan Award for outstanding contributions in condensed matter physics.

REZA PARAAN **Biohub, USA**



Reza Paraan is a team leader at biohub (formerly known as CZ Imaging Institute). His research focuses on benchmarking the cryoET pipeline from data collection to sub-tomogram averaging (STA) of protein complexes. For this, they develop novel benchmark samples such as the phantom (https://www.nature.com/articles/s41592-025-02800-5) that was used for benchmarking particle picking (https://www.biorxiv.org/content/10.1101/2025.11.03.686153v1), and other samples such as E.coli overexpressing virus-like particles (https://cryoetdataportal.czscience.com/datasets/10455) to study the effects of tomogram quality on STA. They collaborate with cryoET tool developers and machine learning developers to push the limits of sub-tomogram averaging and in situ structural biology.

HUAIZONG SHEN **Shenzhen Medical Academy of Research and Translation (SMART), China**



Dr. Huaizong Shen leads the Laboratory of Artificial Intelligence and Biological Structure at the Shenzhen Medical Academy of Research and Translation (SMART). His research integrates structural biology with artificial intelligence to decipher the structure and function of pharmaceutically important ion channels and membrane receptors. By developing novel AI-driven methods, his team aims to democratize cryo-EM technology and accelerate the discovery of novel therapeutic targets and drug precursors. Dr. Shen has authored landmark studies in top-tier journals and is a recipient of China's Excellent Young Scientist Fund and national high-level talent awards.

WENFEI SONG **Thermo Fisher Scientific, Netherlands**



Wenfei Song is a Product Specialist at Thermo Fisher Scientific in Eindhoven, the Netherlands, specializing in cryo-electron microscopy (cryo-EM), with a strong focus on single-particle analysis (SPA). She leads SPA-related application work and has experience across the full workflow, from sample preparation and screening to data collection, image processing, and high-resolution structure determination.

Her work also includes integrated structural biology, combining cryo-EM with mass spectrometry to better understand sample quality, protein behavior, and structure. She also has strong experience in cryo-electron tomography (cryo-ET), with hands-on experience across the full workflow from sample preparation and data collection to tomographic reconstruction and structural analysis.

ELLEN ZHONG **Princeton University, USA**



Ellen Zhong is an Assistant Professor of Computer Science at Princeton University. Her research sits at the intersection of AI and the molecular sciences, where she develops algorithms that drive new scientific discoveries. Her group's research spans methodological research in AI and computer vision, as well as close collaboration with experimentalists in structural biology and chemistry. Previously, she has worked on the AlphaFold team at Google DeepMind and at D. E. Shaw Research on molecular dynamics for drug discovery. She obtained her Ph.D. from MIT in 2022 before joining the Princeton faculty.

Speaker Abstracts (in Presentation Order)

MAKING SENSE OF DISAPPEARING DENSITIES IN SINGLE PARTICLE CRYO-EM

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In the era of single particle cryo-EM and AI driven protein structure prediction, determining high-resolution structure is no longer a difficult but an achievable goal. However, study and understand protein dynamics is still a challenging task. Using TRPM8 and αvβ8 mediated L-TGF-β activation as examples, I intend to illustrate the significance and challenging of studying protein dynamics, particularly when such dynamics play significant role in protein functions.

ATOMISTIC CRYO-EM STRUCTURE DETERMINATION OF SMALL RNA GUESTS BY LEVERAGING THE SYMMETRY OF AN OLIGOMERIC HOST

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Numerous RNAs of biological or biotechnological importance autonomously adopt complex 3D folds. Many such RNA domains are comprised of 100 nucleotides or less (molecular mass < 34 kDa), and current cryogenic sample electron microscopy (cryoEM) does not readily yield their atomic-resolution structures *de novo*. We have developed a new host-guest approach to RNA structure determination by leveraging the symmetric oligomerization of the frameshifting stimulatory element (FSE) of the Rous sarcoma virus (RSV). While the FSE is functionally a monomer in the ~10 kilobase RSV genome, our ~120 nucleotide constructs serendipitously assemble into C2-symmetric dimers and D2-symmetric tetramers (dimers of dimers) *in vitro*. Combined X-ray crystallography and cryoEM structures of the FSE revealed helical extensions at the periphery of the oligomers suitable for replacement by guest RNA domains. Use of the FSE as a host molecule for *de novo* cryoEM structure determination of a guest RNA domain would benefit from prior knowledge of the structure of the host at high resolution, the increase in mass of the guest by fusion to the host, and the symmetry of the resulting host-guest chimera. Previously, large, monomeric RNAs and ribonucleoproteins have been repurposed as hosts for structure RNA determination. Molecular engineering of small RNAs in order to induce their oligomerization has also been proposed to facilitate cryoEM analysis. Our host-guest methodology marries both approaches, exploiting the intrinsic oligomerization of the compact FSE domain, but without altering the (*a priori* unknown) structure of the guest. This host-guest approach has resulted in the *de novo* structure determination of several RNAs. Thus, the aptamer domain of the engineered quinine aptamer in complex with the alkaloid was determined at 2.9 Å resolution (2.7 Å resolution for the host), sufficient to determine the mode of binding of the small molecule. Likewise, this approach revealed the structure of the bacterial 8-oxoguanine riboswitch aptamer domain at ~2.9Å resolution (2.5Å resolution for the host). Optimization of our host-guest approach in order to drive higher throughput RNA structure determination aims to achieve more complete tetramerization of the FSE through a combination of structure-guided engineering and *in vitro* evolution (the first-generation host forms a mixture of monomers and dimers), and to develop "sticky" ends so that the host and guest RNAs can be made to associate in *trans*. Work supported by the intramural program of the NIH.

NEW FRONTIERS FOR CRYO-EM IN ENERGY AND HEALTH

Yuzhang Li[1]

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Electrified interfaces govern processes spanning energy storage, electrochemistry, and biology, yet their transient structures are difficult to observe directly. In this talk, I will discuss our efforts to extend cryogenic electron microscopy beyond static structure determination toward the visualization of dynamic, electrically driven interfaces. We have developed electrified cryo-EM (eCryo-EM), an approach that rapidly freezes electrochemical and electrically driven processes while they occur, kinetically trapping transient states for direct nanoscale imaging. In energy-storage systems, we use this platform to interrogate phenomena ranging from the electric double layer (EDL) to lithium electrodeposition and formation of the solid-electrolyte interphase (SEI). Surprisingly, analysis of SEI growth reveals that two interphases with markedly different chemistry and electrochemical performance exhibit effective ionic diffusivities within approximately 10% of one another. In contrast, their reaction-limited growth rates differ by roughly threefold, suggesting that suppressed interfacial reactivity, rather than enhanced ion transport, is primarily responsible for the improved Coulombic efficiency of the high-performance electrolyte. Beyond batteries, we are extending this framework to biological interfaces. I will present new, unpublished work using eCryo-EM to capture structural transformations of lipid membranes during electroporation, providing a direct view of how electric fields perturb and reorganize soft biological matter. Together, these studies point toward a broader frontier for cryo-EM: a transition from determining equilibrium structures toward capturing transient states of matter under active perturbation. By integrating controlled stimuli with rapid cryogenic trapping, cryo-EM may provide a general framework for connecting nanoscale structure to transport, reaction, and function across systems ranging from electrochemical interfaces to biological membranes.

SELF-DRIVING MICROSCOPY AND WHAT IT SHOULD ANSWER

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Abstract unavailable at time of print.

FLUXIONALITY AND FUNCTIONALITY: PROBING ATOMIC-LEVEL SPATIOTEMPORAL SURFACE DYNAMICS IN CATALYTIC NANOPARTICLES

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Many energy conversion and storage options depend on functionalities of materials such as transport and reactivity. For example, in a heterogeneous catalyst, nanoparticle surfaces and interfaces orchestrate the kinetic pathways for chemical conversion processes which are controlled by charge/mass transport and reactivity at so-called active surface sites. Functionalities such as transport and reactivity involve motion of atomic species in or on the materials and may lead to time-varying perturbations, distortions and destabilizations of the surface structure. There is an increasing recognition that in order to develop a deep understanding of transport and reactivity, it is critical to characterize local structural dynamics, often termed fluxionality. Fluxional atomic motion and migration are now observable with advanced *in situ* transmission electron microscopes, and recent developments in electron detectors and artificial intelligence have made it possible to track the fluxional behavior on nanoparticle surfaces with spatial and temporal resolutions of 1 Å and 10 ms, respectively (at a moderate electron dose). Surfaces showing high degrees of fluxionality behave in a stochastic manner where the system transitions from metastable states to a series of rapidly changing unstable configurations. Under some conditions, such as Pt in CO, it can be possible to correlate the fluxionality of the surface metal atoms with the surface adsorbate configuration. These observations highlight the importance of developing suitable descriptors for atomic-level structural dynamics such as those based on Markovian behavior or persistent homology. Some aspects of the structural dynamics appear to correlate with surface chemical interactions and catalytic functionality, but, as is the case for enzymes, the complexity of the behavior makes it challenging to distinguish functional surface motifs from spectator motifs.

FROM *EX SITU* TO *OPERANDO*: THE ROLE OF ELECTRON MICROSCOPY IN ACCELERATING BATTERY INNOVATION

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The rapid expansion of electric mobility, renewable-energy storage, and emerging electrified technologies is driving unprecedented demand for batteries with higher energy density, improved safety, longer lifetime, and lower cost. At the same time, the battery industry is diversifying beyond conventional lithium-ion chemistries toward silicon-rich anodes, lithium-metal and solid-state batteries, sodium-ion systems, and other emerging technologies. Translating these materials innovations into reliable, manufacturable cells remains challenging because battery performance and degradation are governed by complex and dynamic processes occurring across multiple length and time scales.

Electron microscopy (EM) provides a powerful means of linking battery performance to structural, chemical, and interfacial evolution across the battery development cycle, from R&D through manufacturing and from individual particles and interfaces to electrodes [1–3]. In this presentation, I will discuss several case studies demonstrating how complementary 2D and 3D imaging [4–5], in situ reaction studies [6], and operando electrical biasing [7] can address key characterization challenges and help accelerate battery innovation. These capabilities will be illustrated through applications to emerging battery materials and architectures, including dry-processed electrodes, lithium-metal batteries, single-crystal cathodes, and solid-state batteries. I will also highlight the importance of standardized workflows for sample preparation, data acquisition, and analysis in improving reliability reproducibility of results [8].

By bridging ex situ, *in situ*, and *operando* measurements across relevant length scales, EM is evolving from a tool for observing battery materials into a quantitative platform for establishing structure–process–performance relationships. Such insights can help identify degradation mechanisms, guide materials and electrode design, and ultimately shorten the development cycle for next-generation batteries.

AI METHODS FOR HIGH-THROUGHPUT BIOMOLECULAR STRUCTURE DETERMINATION IN CRYO-EM

Ellen Zhong[1]

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Structural biology has been transformed by breakthroughs in deep learning methods for protein structure prediction. In parallel, cryo-electron microscopy (cryo-EM) has made it possible to reconstruct molecular structures and movies with unprecedented detail. As machine learning continues to transform structural biology, what are the next frontiers for biomolecular structure determination? In this talk, I will present recent advances in AI for cryo-EM that push the boundaries of both throughput and resolving conformational dynamics. I will highlight generative modeling frameworks for 3D reconstruction, such as cryoDRGN and its extensions to in situ and complex mixtures, as well as emerging approaches that integrate protein structure prediction models to advance resolution and interpretability.

BUILDING FULLY AUTOMATED CRYO-EM DATA PROCESSING PIPELINE WITH FOUNDATION MODEL

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Cryo-electron microscopy (cryo-EM) has emerged as a cutting-edge technology for determining high-resolution structures of biological macromolecules, yet its widespread application remains limited by the demand for specialized expertise in data processing. To overcome this barrier, we leveraged an unsupervised learning strategy to develop Cryo-IEF, a foundation model for cryo-EM image evaluation, pre-trained on a massive dataset of approximately 134 million particle images. Cryo-IEF demonstrates outstanding performance across multiple cryo-EM data processing tasks, including classifying particles with distinct structures, clustering particle orientations, and assessing image quality. Fine-tuning further enhances its accuracy and efficiency in evaluating particle image quality. Building on this capability, we constructed CryoWizard, a fully automated single-particle cryo-EM data processing pipeline. Together, Cryo-IEF and CryoWizard represent a synergistic innovation that makes cryo-EM technology more accessible, efficient, and robust, promising to accelerate research across diverse fields in the life sciences.

QUANTUM SENSING OF QUANTUM MATTER

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Abstract unavailable at time of print.

SCANNING SQUID MICROSCOPY

Kathryn Ann Moler [1]

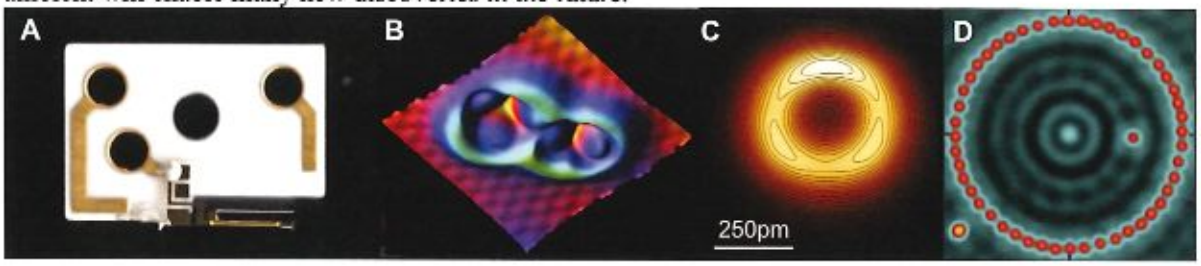
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Abstract unavailable at time of print.

40 YEARS OF ATOMIC FORCE MICROSCOPY – SEEING DENTS IN THE ATOM

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When Binnig et al. resolved the silicon 7x7 reconstructed surface with the scanning tunneling microscope (STM) [1], it came as a surprise. STM relies on a quantum effect, electron tunneling, with its exponential current increase at a factor of ten per Å (100 pm) of distance reduction. This strong increase is key to the high spatial resolution of STM. In 1986, Binnig, Gerber and Quate introduced atomic force microscopy (AFM) [2], a method that also images samples with, ideally, the single front atom of a sharp tip. Richard Feynman believed that the atomic hypothesis ("...attracting each other when they are a little distance apart, but repelling upon being squeezed into one another") contains the most scientific information in the fewest words and started chapter 1 of his Lectures [3] with this statement. The AFM uses exactly these properties of atoms to create images – it senses a surface with a sharp tip mounted on a cantilever. The challenge is that this sensor needs to be able to probe forces small enough such that neither the tips front atom nor the sample are damaged on an atomic level. The silicon 7x7 surface was first resolved by AFM in a noncontact mode [4], where the frequency shift of an oscillating cantilever was used as a feedback mechanism [5]. Further progress was enabled by the invention of the qPlus sensor, a quartz cantilever that is self sensing and oscillates at sub-Angstrom amplitudes [6]. Outfitting its tip with a CO molecule allows to image organic molecules at unprecedented resolution [7]. Today AFM, not only resolves single atoms but images the bonding strength at subatomic spatial resolution ("dents in the atoms") [8,9]. The quantum corral, introduced by Crommie, Lutz and Eigler in 1993 [10], was revisited by AFM in 2021, showing that the 102 electrons that the corral contains can be viewed as the shell of a two-dimensional atom with similar bonding properties to AFM tips as a natural atom [11]. AFM is now a powerful tool for the surface science of insulators in ultrahigh vacuum [12] that precisely records the evolution of chemical bonds [13]. Most applications of AFM are in ambient conditions and we have first obtained true atomic resolution in ambient with a qPlus microscope in 2013. Recently, our homebuilt qPlus based Orpheus II microscope [14] was turned into a professional tool (NX1) by Park Systems. We trust AFM at low temperatures and in ambient will enable many new discoveries in the future.



A qPlus sensor. **B** AFM image of a Fe trimer next to a Fe dimer on Cu(111). **C** AFM image of a single Fe atom on Cu(111) [8]. **D** Quantum corral with one perturbing atom [11].

References

1. G. Binnig et al., *Phys. Rev. Lett.* **50** 120 (1983).
2. G. Binnig et al., *Phys. Rev. Lett.* **56** 930 (1986).
3. R. P. Feynman et al., The Feynman Lectures on Physics, Addison Wesley 1963.
4. F. J. Giessibl, *Science* **267**, 68 (1995).
5. T. R. Albrecht et al., *J. Appl. Phys.* **69**, 668 (1991).
6. F. J. Giessibl, *Rev. Sci. Instrum.* **90**, 011101 (2019).
7. L. Gross et al., *Science* **325** 1110 (2009).
8. M. Emmrich et al., *Science* **348** 308 (2015).
9. F. Huber et al., *Science* **366** 235 (2019).
10. M.F. Crommie, *Science* **262** 218 (1993).
11. F. Stilp et al., *Science* **372** 1196 (2021).
12. J. Hütner et al., *Science* **385** 1241 (2024).
13. M. Weiss et al., *Phys. Rev. B* **113** 245402 (2026).
14. K. Pürckhauer et al., *Rev. Sci. Instrum.* **91**, 083701 (2020)

HIGH-THROUGHPUT ELECTRON TOMOGRAPHY FOR BRAIN MAPPING

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Abstract unavailable at time of print.

SCANNING NANODIFFRACTION OF BEAM-SENSITIVE MATERIALS

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Scanning nanodiffraction, a subset of 4D-STEM, is a powerful technique to elucidate structure from materials that are typically considered "beam-sensitive" due to their tendency to lose long-range order during exposure to an electron beam. The technique is inherently low-dose, in large part because it is easy to control the fluence and acquire diffraction patterns from unexposed areas. The rich datasets allow us to not only reconstruct meaningful structure maps across many length scales, but also to make statistical correlations between samples because of the large number of frames.

The featured example is a homopolymer that conducts both ions and electrons – an organic mixed ionic-electronic conductor (OMIEC) for use in flexible electronics and energy storage. During device operation, the polymer is present in a liquid electrolyte and experiences charging/doping and discharging/de-doping. Transport characteristics vary depending on the doping state, and it is hypothesized that this reflects an underlying change in polymer structure. Scanning nanodiffraction was used to probe the structure dry, in the electrolyte environment, and at different stages of charging. To make these operando measurements, a plunge-freezing tool was modified to accommodate a charging apparatus to prepare the TEM samples; the grids were cryo-transferred into a side-entry cryogenic holder. As is typical with many semicrystalline polymers, the diffraction signal is weak requiring direct electron detectors and benefitting from machine learning to improve peak-finding routines. These measurements reveal that structural disorder develops non-uniformly across the film and across different crystallographic directions, providing a spatially resolved signature of charge-induced reorganization that is not accessible via common bulk-averaged techniques.

A HIGH-THROUGHPUT APPROACH TO IDENTIFY AND CHARACTERIZE MOLECULAR GLUES STABILIZING WEE1- CRBN-DDB1 COMPLEX

Wenfei Song[1], Kheewoong Baek[2], Weijing Liu[3], Eric Fischer[2], Christopher Mullen[3], Yuan Xiong[2], Ken Durbin[4], Rosa Viner[3], Thomas Moehring5 , Oliver Raschdorf5

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Targeted protein degradation of conventionally undruggable proteins represents a groundbreaking strategy in drug discovery. This approach leverages molecular glues (MGs) to enhance the weak intrinsic interactions between targeted proteins with E3 ligases by modifying the interaction surface, thereby facilitating their ubiquitin-proteasome-mediated degradation without relying on the strong binary affinity. Wee-like protein kinase (WEE1), a key cell cycle regulator, and the CRBN-DDB1 E3 ubiquitin ligase complex are compelling targets for MG-mediated stabilization, offering novel strategies to disrupt cell cycle checkpoints in cancer.

We report a high-throughput molecular glue screening approach using native mass spectrometry to identify molecular glues that enhance WEE1 binding to CRBN-DDB1 complex. This method enables the screening and analysis of over 600 molecular glue compounds per day. Six promising molecular glues were identified and further characterized by cryo-electron microscopy (Cryo-EM). Due to the challenges in obtaining high quality cryo-EM map arising from preferred particle orientation, extensive sample screening and tilted data acquisition were required. Multiple Cryo-EM datasets were collected and analyzed to further characterize the six promising molecular glues, resulting in three high-resolution structures which enable direct visualization of the binding interface. Structural analysis of the ternary complexes of WEE1-MG-CRBN-DDB1 offers detailed mechanistic insights into how these molecular glues facilitate and stabilize the interaction between WEE1 and the CRBN-DDB1 complex, which is essential for drug design and optimization. The integration of native mass spectrometry with cryo-EM enables both rapid screening and high-resolution structural validation, offering a powerful workflow for molecular glue discovery and mechanism elucidation.

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Abstract unavailable at time of print.

IN SITU ELECTRON MICROSCOPY AND X-RAY SPECTROMICROSCOPY TO UNDERSTAND ELECTROCATALYTIC MATERIALS AND PROCESSES

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Electrochemical CO_2 conversion offers a route to use renewable sources of electricity to convert CO_2 into valuable carbon-based fuels and chemicals, including carbon monoxide, ethanol and ethylene. For electrochemical CO_2 conversion technologies to become viable component of future sustainable energy infrastructures, the development of improved performance catalyst materials and reactor designs are needed to achieve high conversion rates, selectivity and single-pass utilization of CO_2. This talk will focus on the development of techniques to characterize the properties of electrochemical CO_2 conversion materials under reaction conditions that will guide the design of next-generation materials and reactors. The talk will focus primarily on in situ transmission electron microscopy (TEM) and related spectroscopic techniques including select area electron diffraction, along with synchrotron-based methods including soft scanning transmission X-ray microscopy (STXM).

ELECTRON VIDEOGRAPHY OF SOFT AND ENERGY MATTER AT THE NANOSCALE

Qian Chen[1]

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I will discuss my group's recent progress on adapting a suite of electron microscopy methods (e.g., liquid-phase TEM, electron tomography, 4D-STEM) and machine-learning based data-mining to synthetic soft and energy related systems. In the first direction, we focus on the phase behaviors of nano-sized building units as they are dispersed in solution. As a proof-of-concept, we directly image the crystallization pathways of nanosized colloids into superlattices, where the discreteness and multi-scale coupling effects complicate the free energy landscape. Single particle tracking and simulations combined unravel a series of interesting pathways at this length scale, such as non-classical crystallization, size-dependent crystal growth habits of superlattices, and moiré patterning, enabling advanced crystal engineering. In the second direction, we further push direct imaging to separation membranes and multivalent ion batteries, where the strain embedded heterogeneously within leads to morphogenesis and distinct charge transport properties. We foresee our suite of "electron videography" tools to provide crucial and complementary insights in various materials systems, with the common theme of imaging and manipulating materials in space and time at the nanoscale.

STRUCTURAL BIOLOGY OF MEMBRANE FISSION

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Membrane fission is fundamental to cellular life and central to processes including organelle division and endocytosis. Membranes substrates undergoing fission span a remarkable range of sizes and topologies, from narrow vesicle necks to large multi-membrane organelles. Dynamin-related proteins (Drps) are large GTPases that catalyze membrane fission by assembling into spiral scaffolds around membrane substrates and constricting in a GTP-dependent manner. Atomic models of human dynamin 1, the founding member of the Drp superfamily, have revealed key principles of assembly and constriction during narrow vesicle neck fission, including our structures capturing the late stages of constriction and in situ cryo-electron tomography (cryo-ET) of dynamin assemblies on membranes. In contrast, high-resolution structures of organelle-scale Drp spiral filaments are lacking, leaving unclear how Drps assemble large-diameter filaments for organelle fission. To address this gap, we investigated an essential Drp required for the division of endosymbiotic organelles in malaria parasites. We show that this Drp forms large spiral filaments with diameters compatible to its target organelles and it is sufficient to catalyze membrane fission. Cryo-EM structures identify key oligomerization interfaces, and comparison with other Drps suggests that repositioning of α-helices contributes to the formation of larger-diameter assemblies. Cryo-ET further reveals that the protein forms spiral filaments in an unexpected rung-offset organization with implications for the mechanism of membrane constriction. Together, these findings reveal structural principles that enable membrane fission Drps to assemble spiral filaments of distinct diameters for fission across membrane substrates of different scales.

BENCHMARK SPECIMENS FOR QUANTITATIVE EVALUATION OF THE CRYOET PIPELINE, FROM DATA COLLECTION TO SUBTOMOGRAM AVERAGING

Reza Paraan[1], Ariana Peck[1], Joshua Hutchings[1], Mallak Ali[1], Yue Yu[1], Mykhailo Kopylov[1], Anchi Cheng[1], Pavel Olshin[1], Elizabeth Montabana[1], Utz Ermel[1], Tahiti Dutta[2], M.G. Finn[2], Joshua Dickerson[3], Eric Cooper[4], Bronwyn Lucas[3], Holger Muller[4], Matthias Haury[1], David Agard[1,5], Bridget Carragher[1] [1]Biohub, Redwood City, California, USA; [2]Chemistry, Georgia Tech, Atlanta, GA, USA; [3]MCB, UC, Berkeley, Berkeley, CA, USA; [4]Physics, UC, Berkeley, Berkeley, CA, USA; [5]Biochemistry & Biophysics, UC, San Francisco, CA, USA

Cryo-electron tomography (cryoET) can reveal macromolecular structures within their native cellular environments. However, because in situ samples rarely provide a complete ground truth, an objective comparison of new methods and technologies remains difficult. We have developed benchmark specimens that enable quantitative assessments of the cryoET pipeline from data collection through subtomogram averaging.

We developed a community particle-picking challenge where participants identified five molecular species across hundreds of experimental tomograms. The methods developed for this challenge surpassed existing approaches and revealed important lessons about data augmentation and the sensitivity of subtomogram averaging to false-positive picks. We also developed a resolution standard using PP7 virus-like particles (VLPs) expressed in *Escherichia coli*. This controlled in situ system enables direct comparisons of specimen preparation, data acquisition, and refinement strategies. Evaluations of both plunge-frozen and high-pressure frozen specimens demonstrated that the VLP resolution provides a sensitive diagnostic of the full cryoET workflow. The high symmetry of VLPs also supports high-resolution averaging from limited data, particularly useful for technically demanding microscopy experiments.

The *E. coli*-PP7 system was subsequently used to benchmark the first tomography dataset collected with a crossed laser phase plate (xLPP). Its known structure and reproducible cellular context enabled direct assessment of particle detectability, low-spatial-frequency contrast, alignment accuracy, imaging artifacts, and downstream subtomogram averaging. Together these benchmarks provide a quantitative link between physical performance of emerging technologies and their ultimate structural output under realistic cryoET conditions.

Poster Abstracts

(in Alphabetical Order by Presenting Author's Last Name)

SMARTSCOPE: A SELF-TUNING WORKFLOW FOR HIGH-THROUGHPUT CRYO-EM

Jonathan Bouvette[1], **Laura He**[2], **Mario Borgnia**[3], **Alberto Bartesaghi**[2]

[1]Princeton University; [2]Duke University; [3]NIEHS/NIH;

SmartScope has already made cryo-EM screening and data collection faster and more accessible by automating specimen evaluation. Here, we close the loop between acquisition and analysis with real-time feedback. As data streams in, quality metrics steer the microscope toward the best regions on the grid while steering it away from areas resembling prior failures. Sessions on unproductive specimens are automatically abandoned in favor of better candidates. And once enough particles have been collected to reach a target resolution, the microscope knows to stop. The result is a fully autonomous, sample-to-structure pipeline that eliminates wasted microscope time and puts high-resolution structure determination within reach of non-experts.

TOPOGRAPHIC CONTROL OF DEFECTS IN MONOLAYER WS2

Satya Butler[1], Guangming Cheng[1], Nan Yao[1], Saien Xie[1]

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Single-crystalline thin films are defined on flat substrates with translational symmetry. Curved substrates break this symmetry, providing a unique platform for engineering crystalline thin film growth, tuning defects, and controlling material properties. Two-dimensional (2D) materials such as transition metal dichalcogenides (TMDs) that can withstand extreme levels of strain provide an ideal platform for studying the interplay between defect formation and lattice strain in crystals. This, in turn, allows for curvature engineering of the electronic and optical properties of 2D materials. While there has been substantial effort towards crystalline TMD growth, controlled synthesis of dislocations in these materials remains an unsolved challenge. Here, we present the topography engineered synthesis of monolayer TMDs with controlled morphology and defect density. Atomic-resolution high-angle annular dark-field scanning transmission electron microscopy (HAADF-STEM) reveals arrays of defects in a single-crystalline domain grown over a non-flat surface. These crystalline domains can merge and form a continuous film which, when suspended, preserves its 3D morphology and demonstrates its mechanical strength and robust structure. Our work establishes topography engineering as a powerful approach for controlling defect formation in thin film synthesis, providing a new method for creating functional materials for future electronics and optoelectronics.

THE JOINT OPTIMIZATION OF PARTICLE LOCALIZATION AND TILT-SERIES ALIGNMENT FOR CRYOET

Jackson Carrion[1], Joseph Davis[2]

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High-resolution subtomogram averaging in cryo-electron tomography (cryoET) depends on high-quality tilt-series alignment, as alignment errors propagate through tomogram reconstruction and limit the performance of downstream 3D particle picking methods. Existing approaches often rely on volumetric template matching or tomogram-based particle detection, which can be computationally expensive and sensitive to reconstruction artifacts. Here we present JOLT [Joint Optimization of Particle Localization and Tilt-series alignment], a template-free cryoET particle localization framework that operates directly on the 2D tilt-series data. JOLT uses lightweight 2D CNN particle pickers to detect candidate features in individual low-dose tilt images, then identifies geometrically consistent features across multiple tilt images by establishing 2D particle trajectories with optimal transport. These resulting trajectories provide a sparse set of data-derived fiducials that support the joint refinement of 3D particle positions and tilt-series alignment through differentiable bundle adjustments. By formulating particle localization and tilt-series alignment as a single geometric optimization over 2D point clouds, JOLT avoids expensive volumetric template searches while providing a computationally efficient framework for template-free 3D particle localization and tilt-series geometry refinement.

TANTALUM AND TANTALUM-ALLOYED THIN FILMS FOR QUANTUM INFORMATION SYSTEMS

Guangming Cheng[1], Nan Yao[1]

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Superconducting qubits are among the most advanced platforms for quantum information processing, offering potential for scalability and compatibility with standard device fabrication techniques. Among the materials investigated for superconducting qubits, Tantalum (Ta) has emerged as a compelling candidate due to its self-terminating stoichiometric surface oxide and chemical resistance to harsh post-fabrication processes. Efforts to mitigate surface loss by treating surface oxides and using high-resistivity silicon substrates have led to Ta-based qubits with lifetimes and coherence times exceeding 1 ms. Guided by these successes in reducing dielectric loss in Ta-based devices, we are interested in exploring material engineering approaches that enhance the superconducting gap. Recently, we explored superconducting gap engineering in Ta-based devices as a powerful strategy for expanding the range of suitable host materials. Here, we demonstrate an approach for tuning deposition conditions to control the orientation and transport properties of Ta-IIf alloy thin films. We investigate the microstructure and composition distribution of the thin films by using high-resolution scanning transmission electron microscopy (STEM) with a super-X EDS system. Our structural analysis using X-ray diffraction (XRD), TEM diffraction, and STEM confirms a BCC structure across all films, and direct current (DC) transport measurements reveal a 40% enhancement in Tc relative to undoped Ta (Tc = 4.3 K). We monitored oxide growth over an 18-day period following film deposition to better understand how Hf incorporation influences surface oxides. In both Ta and Hf spectra, the fraction of fully oxidized (Ta2O5 and HfO2) states gradually increased during the 18-day exposure, indicating progressive surface oxidation. The analysis reveals that the oxide thickness for the 1 ks Ta-Hf alloy film is approximately 3 nm in thickness, comparable to the 3 nm oxide observed on native Ta. The measured surface loss tangent of the Ta-Hf alloy remains comparable to that of native Ta. This result suggests that the surface oxide of the Ta-Hf alloy, which contains a measurable fraction of IIfOx, does not introduce additional dielectric loss. By engineering the alloy composition and film deposition, this study achieves both improved superconducting properties and compatibility with fabrication processes. These findings show that material doping and alloying can strategically expand the suite of viable materials for superconducting quantum devices. This approach provides a pathway for tailoring material properties to support the next generation of quantum technologies and offers broad relevance to researchers in materials science, quantum information, and device engineering.

MINT: RAPID AI SEGMENTATION AND ULTRASTRUCTURAL ANALYSIS FOR CRYO-ET

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In electron microscopy, the slowest step is no longer data collection but analysis. A single tomogram can take hours or days to annotate by hand, and this bottleneck blocks the generation of quantitative results from most tomography data. We built MINT (Multimodal Imaging Neural Toolkit), an open-source desktop application that can take data from raw tilt series through reconstruction, segmentation, and quantitative analysis in a single interface. In MINT, you segment by describing what you want. Type "mitochondrial cristae", "single-membrane vesicles", or "ribosomes" and the program returns color-coded masks in under four minutes per tomogram. MINT leverages multiple AI models, including one fine-tuned in-house on tomograms, volume-EM data, and single-particle micrographs. It works across diverse sample types and can label multiple features simultaneously. It reviews its own masks across multiple passes, provides tools for manual review and editing, and segments an entire set of volumes in a single batch. Once a tomogram is segmented, MINT measures the segmentations, reporting volume, surface area, sphericity, diameter, contact analysis, curvature, and multiple other measurements from per-feature meshes, so changes in ultrastructure across sample types can be quickly observed. For cryo-ET, MINT converts segmented particles into coordinate files for subtomogram averaging. These particles are also used to train a CNN for your specific target and dataset. Because MINT reads the microscope's atlas and search-map metadata, tomograms, search maps, and overview images are overlaid back onto the lower-magnification images using stage positions and cross-correlation, and every tomogram can be placed where it was collected, with its segmentations in context. Segmentations of lower-magnification search maps can also be performed to track the relative positioning of organelles within the sample. MINT can also automatically render publication-ready 3D movies of the results. To make this whole workflow straightforward, a built-in assistant, driven by a large language model, answers questions about the software and queues and executes jobs on your behalf, performing reconstruction, segmentation, ultrastructural analysis, and the rest of MINT's pipeline. This allows an entire analysis to be run in plain language. We have used this pipeline across cultured fibroblasts, neuronal models of Parkinson's disease, glioblastomas, Chlamydomonas, viral particles, and autopsy tissue. In human fibroblasts, we can track cristae remodeling with age and disease, as well as changes in inter-organelle contacts. In each sample type, the time from raw data to a tomogram measurement drops from days to minutes.

INTEGRATIVE STRUCTURAL BIOLOGY TO DISSECT MECHANISMS OF LIPID-DEPENDENT ANTIFUNGAL DRUG ACTION

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Fungal β-1,3-glucan synthase (GS), the membrane protein complex responsible for synthesizing a major component of the fungal cell wall, is the target of echinocandin antifungal drugs. Although recent high-resolution cryo-EM structures of GS have revealed atomic details of its catalytic and drug-binding sites, the mechanisms underlying drug inhibition and resistance remain incompletely understood, in part because the dynamic membrane environment and lipid–protein interactions are difficult to recapitulate in vitro. To address this gap, we integrated cryo-electron tomography (cryo-ET) with biochemical, biophysical, and microbiological approaches to investigate echinocandin action in native fungal plasma membranes. Echinocandin exposure alters membrane protein organization and membrane biophysical properties, suggesting that drug binding to GS may perturb the dynamics of the GS complex and its local lipid network. Consistent with this model, perturbations in lipid composition are associated with changes in membrane biophysical properties and modulated GS susceptibility to echinocandins, supporting a role for the local lipid environment in drug action. In Cryptococcus neoformans, which exhibits intrinsic echinocandin resistance, we find that disrupting lipid flipase function perturbs plasma membrane asymmetry and sensitizes cells to echinocandins. Cryo-ET further reveals nanoscale changes in plasma membrane organization associated with altered drug susceptibility, further supporting the model in which the membrane lipids modulate echinocandin action. Together, our studies establish a structural framework linking native membrane organization to antifungal drug action and resistance. Our findings identify membrane organization and lipid composition as key determinants of echinocandin susceptibility and highlight the membrane environment as a potential therapeutic vulnerability for overcoming antifungal resistance. This work also demonstrates the power of integrative structural biology to investigate dynamic molecular organization and drug-target interactions within native cellular membranes.

LEARNING HIGH-ENTROPY-ALLOY NANOPARTICLE MICROSTRUCTURE ACROSS LENGTH SCALES

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Microstructure is typically captured by microscopy at select fields of view. However, the number and scales of measurements required to recover a material's structure are chosen heuristically, obscuring whether a microscopy campaign is complete. This is especially limiting for hierarchical microstructures, where distinct morphologies and structure–property relationships emerge across scales, and exhaustive imaging is costly. A quantitative description of microstructure therefore requires not only property statistics, but also quantifying much independent information is contributed by microscopy at each length scale. Here, we formulate multiscale microstructure characterization as a generative image modeling problem. We captured more than 20,000 scanning electron microscopy images of carbothermal-shock high-entropy-alloy nanoparticles on carbon nanofiber supports, spanning five field widths from microns to millimeters, and 270 ternary synthesis recipes across seven elements. We train generative models directly on these images to learn the scale-dependent distributions of experimental microstructure, generating full-resolution SEM images at specified field widths. We further train the model to generalize to previously unseen nanoparticle chemistries: given a sparse set of experimental images acquired across multiple field widths, it generates realistic microstructures at a requested field width. Because this inference does not require spatially registered images or identical acquisition conditions across scales, it accommodates experimental variation in exposure, stage position, and carbon-nanofiber substrate. We assess whether these generated images reproduce microstructure rather than merely SEM-like appearance using physically interpretable structural distributions. Generated samples recover the experimental distributions of high-entropy-alloy nanoparticle radii, nanoparticle aspect ratio and eccentricity, and carbon-nanofibre widths and orientations. The ability to reconstruct these statistics from sparse measurements provides an operational test of how completely a set of microscopy observations specifies a material. We systematically vary the field widths supplied to the model and those withheld for reconstruction. Cross-scale reconstruction reveals that microstructural information is distributed non-uniformly across length scales: observations made at one field width can contain substantial information about structure at another, and measurements at the target scale are not always the most informative measurements for reconstructing it. These experiments empirically demonstrate how structural information is shared across the mesoscale. Finally, by varying model capacity, compute, and experimental sampling, we establish generative-model scaling relations for microstructure reconstruction. These relations connect reconstruction fidelity to measurement budget, providing the first quantitative route to determine how many SEM images, at which field widths, are needed to statistically characterize a material. This recasts microstructure as a measurable information-acquisition problem for finite experimental microscopy campaigns.

OBSERVATION OF CHEMICAL BONDS AND COLLECTIVE SURFACE PHENOMENA IN REAL SPACE WITH SCANNING PROBE MICROSCOPY

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Scanning probe microscopy (SPM) enables the direct observation of atomic and molecular structures in real space. Beyond structural imaging, it can also provide insights into chemical bonds and collective surface phenomena. We combine atomic force microscopy (AFM), scanning tunneling microscopy (STM), density functional theory (DFT), and statistical analysis to study these effects at different length scales. High-resolution AFM images often show bond-like features between atoms, but whether these features represent true chemical bonds remains unclear. Using a CO-functionalized tip, we find that apparent bond-like features form when the interatomic separation is below an upper limit of 4.4–5.1 Å. This threshold holds across nonbonded CO clusters, an ionic NaCl surface, and a hydrogen-bonded water hexamer. True bonds are detected only when their electron density is comparable to or greater than that above adjacent atoms, as demonstrated for the triple bond in tolane. We further propose AFM line profiles and the overlap of the tip's van der Waals surface with that associated with the bond as practical criteria for distinguishing true bond contributions from steric artifacts [1]. We further investigate CO adsorption on Cu(111), a model system in surface science and heterogeneous catalysis. CO adsorption consistently saturates at approximately 26%, significantly below the expected 33% coverage. This anomalous saturation persists across a wide range of dosing amounts and deposition temperatures, indicating an intrinsic thermodynamic constraint rather than kinetic trapping. Statistical analysis of the images reveals an effective interaction radius of approximately 5.3 nm. DFT calculations show that increased coverage confines Cu(111) surface-state electrons and produces overlapping elastic strain fields, both of which weaken CO binding. These substrate-mediated interactions collectively enforce a self-limiting adsorbate density [2]. These results demonstrate that SPM can move beyond structural imaging to reveal local chemical interactions and collective surface behavior across different length scales. [1] P. Chen, D. Fan, A. Selloni, and N. Yao, Cell Rep. Phys. Sci. 7, 103483 (2026). [2] P. Chen, D. Fan, A. Selloni, J. Fan, and N. Yao, J. Am. Chem. Soc. 148, 360−367 (2026).

INVESTIGATING THE ROLE OF SPHINGOLIPIDS IN THE HIGHER-ORDER ORGANIZATION OF PLASMA MEMBRANE PROTEINS IN CANDIDA GLABRATA.

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Fungal plasma membrane proteins are important therapeutic targets for antifungal drugs, making it essential to understand how their organization contributes to drug resistance. Our previous work demonstrated that caspofungin, an echinocandin antifungal that inhibits β-(1,3)-glucan synthase (GS), alters the higher-order organization of plasma membrane proteins in Candida glabrata, a common opportunistic fungal pathogen. Interestingly, disruption of mature sphingolipid biosynthesis, which is critical for lipid raft formation and plasma membrane organization, increases membrane tension and enhances caspofungin resistance. These findings suggest that sphingolipids play an important role in maintaining membrane organization, however, the structural basis for this phenotype remains unknown, as the plasma membrane architecture has not been directly visualized in sphingolipid-deficient cells. Cryogenic electron tomography (cryo-ET) enables visualization of membrane architecture in native membranes, providing a powerful approach to investigate how sphingolipid depletion alters plasma membrane organization. We hypothesize that depletion of mature sphingolipids disrupts the higher-order organization of plasma membrane proteins, thereby remodeling the membrane landscape in ways that contribute to antifungal resistance. To test this hypothesis, we are using cryo-ET to examine crude plasma membrane extracts from Candida glabrata lacking the FEN1 gene, using GS and the proton pump Pma1 as molecular landmarks to characterize changes in membrane organization. By directly visualizing the membrane landscape, this work aims to reveal the structural mechanism by which sphingolipid depletion reorganizes plasma membrane proteins and contributes to echinocandin resistance.

QUANTITATIVE SCANNING NEAR-FIELD OPTICAL SPECTROSCOPY ACROSS MATERIALS IN AIR AND LIQUID

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Scanning near-field optical microscopy (SNOM) enables chemical and optical characterisation at spatial resolutions below 50 nm, but quantitative interpretation remains challenging. Measured spectra arise from a convolution of intrinsic material response, resonant absorption, scattering, re-radiation and probe-induced tip–sample coupling. These effects distort spectral amplitudes and lineshapes, obscuring the relationship between near-field contrast and fundamental optical properties. Here, we introduce a unified framework for retrieving the complex permittivity of a sample directly from tip-scattered SNOM spectra. The method converts instrument-dependent near-field signals into intrinsic, physically interpretable optical constants and is applicable across measurement environments without requiring modification of the microscope hardware. We validate the framework using inorganic and organic reference materials spanning strong, moderate and negligible optical dispersion. For both the real and imaginary components of the permittivity, the retrieved values show close agreement with reference values obtained by far-field ellipsometry. We further demonstrate that the same framework operates in both air and liquid, where changes in dielectric background and tip–sample interactions make quantitative analysis particularly challenging. As representative applications, we recover molecular fingerprints of nanoplastics dispersed in liquid and analyse nanoscale spectroscopic signatures associated with the molecular composition and protein secondary structure of Escherichia coli. These results show that near-field spectra acquired from chemically and structurally distinct samples can be interpreted within a common quantitative framework. The approach can be applied to existing and future SNOM datasets, including measurements of unknown or heterogeneous specimens in their native environments. By separating intrinsic material response from probe-mediated contrast, the framework reduces the technical burden of reconstructing nanoscale optical constants and supports comparative measurements across samples and environments. It therefore provides a broadly applicable route towards quantitative near-field spectroscopy for the physical, materials and life sciences, with potential applications ranging from nanoscale analysis of environmental contaminants to investigations of functional biomaterials and living systems.

RESOLVING THE CONDUCTIVE ULTRASTRUCTURE OF CABLE BACTERIA BY ELECTRON MICROSCOPY

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Cable bacteria are multicellular organisms that transport electrons over centimeter-scale distances, far exceeding the length scales typically associated with biological electron transfer. This long-distance electron transport is mediated by highly conductive periplasmic fibers, with reported conductivities of 10-2–102 S cm^{-1}, among the highest observed for biological materials. Despite their remarkable electrical properties, the molecular and ultrastructural organization of these conductive fibers remains poorly understood, limiting our understanding of the mechanism underlying long-distance charge transport.

Here, we investigate the ultrastructural and molecular organization of cable bacteria conductive fibers using a combination of high-resolution scanning Electron Microscopy (SEM), Raman Spectroscopy, and Atomic Force Microscopy (AFM). Measurements were performed on both intact cable bacteria filaments and fiber networks isolated using an SDS–EDTA extraction procedure, enabling the direct interrogation of the physical and chemical structures of these conductive materials at multiple levels of organization.

Our observations support a core–shell organization of the periplasmic fibers, consisting of a conductive proteinaceous core surrounded by a comparatively insulating protein shell. Spectroscopic characterization further indicates that the conductive core contains a sulfur-coordinated nickel cofactor, providing a potential molecular basis for the unprecedented electronic properties of the fibers. Together, these results connect the nanoscale structural organization of cable bacteria fibers with their chemical composition and electrical properties.

Ongoing work aims to resolve the conductive fibers at higher spatial resolution. Focused ion beam (FIB) and selective chemical or enzymatic treatments will be used to isolate and expose individual periplasmic fibers, followed by Transmission Electron Microscopy (TEM) and cryogenic Electron Microscopy (cryo-EM) to investigate their molecular organization and nickel–sulfur coordination environment. Temperature dependent four-probe resistivity measurements will further characterize the mechanisms of their intrinsic conductivity. These complementary structural, spectroscopic, and electrical measurements will help establish a molecular-level model for long-distance electron transport in cable bacteria.

AB INITIO SUBTOMOGRAM AVERAGING OF COMPLEX MIXTURES FROM CRYO-ET

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The direct atomic-scale observation of biomolecules with cryo-electron tomography (cryo-ET) can reveal how structural interactions enable specific cellular processes in situ. However, existing methods for high-resolution structure determination from cryo-ET are not robust to the extreme heterogeneity of real biological systems and often rely on prior templates for classification and subtomogram averaging (STA). Here, we extend cryoDRGN-AI, a neural framework for ab initio heterogeneous 3D reconstruction, to unfiltered experimental cryo-ET datasets with conformational and compositional variability. We first demonstrate ab initio reconstruction for multiple samples directly from aligned tilt series, including thyroglobulin and yeast 80S ribosomes across translation states. CryoDRGN-AI achieves single-shot, fully de novo 3D reconstruction to subnanometer resolution, whereas comparable results from a typical STA workflow require iterative reference-based refinement. Finally, in collaboration with the Chan Zuckerberg Biohub, we share preliminary results for simultaneous reconstruction of mixed molecular species from cellular lysate, advancing toward high-resolution "visual proteomics" with cryo-ET.

4D-STEM AND OPERANDO X-RAY DIFFRACTION STUDIES OF DYNAMIC SURFACE RECONSTRUCTION OF SINGLE-CRYSTAL LANIO3 CATALYSTS

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Nickel-based perovskite oxides, particularly nickelates containing Lanthanum as the A site alkaline-earth metal, are considered promising catalysts for the oxygen evolution reaction (OER), which exhibit comparable activity to benchmark Ru- and Ir-based materials. Despite their potential, their catalytic mechanism remains insufficiently understood due to material complexity arising from substantial structural reconstruction and following elemental leaching during the OER process. Such dynamic behavior complicates the understanding of the OER mechanism and thus inhibits rational catalyst design. Therefore, direct structural characterizations of these transformations are critically needed. This work combines atomic-resolution scanning transmission electron microscopy (STEM), four-dimensional STEM (4D-STEM), and operando synchrotron surface X-ray diffraction to characterize post-reaction structural changes and track dynamic surface reconstruction during oxygen evolution. Epitaxial single-crystal LaNiO3 thin films grown by molecular beam epitaxy (MBE) provide a well-defined model system for examining surface reconstruction and investigating catalytical mechanisms. Atomic-scale STEM reveals amorphization of the thin film surface, while STEM-EDX identifies Ni depletion within the corresponding reconstructed layer. 4D-STEM further maps local strain induced by the reconstruction process. Complementary operando surface X-ray diffraction based crystal truncation rod (CTR), tracks the structural evolution of the film with monolayer-scale sensitivity under oxygen-evolution conditions. Together, these measurements connect atomic-scale structural, compositional and strain changes in the recovered material with the dynamic surface reconstruction observed during electrochemical operation.

MULTIMODAL IMAGING OF PROTEIN-MEMBRANE INTERACTIONS IN NEURODEGENERATION

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Intrinsically disordered proteins (IDPs) play central roles in cellular organization through dynamic interactions with biomolecular assemblies, membranes, and protein networks. Dysregulation of these proteins is a hallmark of numerous neurodegenerative diseases, including Huntington's and Parkinson's diseases, where aberrant protein assembly and membrane remodeling are thought to contribute to disease progression. However, the structural heterogeneity and transient interactions of IDPs have made them difficult to study using conventional structural biology approaches. Cryo-electron tomography (cryo-ET) provides a unique opportunity to visualize these proteins directly within their native cellular environment, preserving their interactions with membranes, organelles, and other cellular components. Our laboratory integrates cryo-ET with multimodal imaging approaches, including cryo-correlative light and electron microscopy (cryo-CLEM), to investigate the structure, dynamics, and cellular organization of disease-associated IDPs. Using huntingtin and α-synuclein as model systems, we investigate how disease-associated protein assemblies interact with biological membranes, remodel membrane architecture, and reorganize intracellular ultrastructure. Our studies reveal membrane deformation associated with huntingtin assemblies in model membrane systems and membrane-associated α-synuclein-rich assemblies within cells, providing structural insight into protein-membrane interactions that are difficult to capture using conventional methods. Together, these findings demonstrate the power of multimodal imaging to bridge molecular structure and cellular function, advancing our understanding of the mechanisms by which intrinsically disordered proteins contribute to neurodegenerative disease.

HIGH-FREQUENCY NANOMECHANICAL SCANNING PROBE SENSOR AT THE QUANTUM LIMIT

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We introduce a nanomechanical atomic force microscopy sensor operating at frequencies above 10^{8} Hz and potentially into the gigahertz range. We formulate a figure of merit that sets the force-gradient sensitivity of mechanical-resonator-based scanning probes, and use it to devise an optimization strategy that exceeds the state of the art by three orders of magnitude. We discuss the capability of high-frequency sensors to probe local dynamics and achieve quantum-limited measurements. The on-chip design brings the toolbox of nanoengineering to scanning probe sensors, unlocking the potential to study a wide range of systems in materials and device physics.

THEORY-GUIDED ATOMIC-SCALE STEM IMAGING OF BIMETALLIC THIN FILMS FOR AMMONIA PRODUCTION

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Ammonia is one of the world's most important industrial chemicals, with the majority of its production used for fertilizer manufacturing. However, natural nitrogen cycling cannot efficiently convert the excessive nitrate accumulated in groundwater into ammonia, resulting in widespread nitrate contamination of water resources. Electrochemical nitrate reduction (NO_3RR) offers a promising and sustainable strategy to simultaneously mitigate nitrate pollution and produce ammonia using renewable electricity. Cu-based bimetallic catalysts have emerged as an attractive platform for optimizing catalytic activity and selectivity toward NO_3RR. In this work, we employ adatom formation energy as a theoretical descriptor to evaluate the stability of surface-active sites across different Cu-based bimetallic compositions. Density functional theory calculations indicate that alloying Cu with secondary metals leads to composition-dependent adatom formation energies. Incorporating relatively noble or coinage metals, such as Ag, Au, and Zn, lowers the adatom formation energy, facilitating the formation of surface adatoms that have recently been recognized as highly active catalytic sites. To experimentally validate this hypothesis, we fabricate high-throughput composition-spread Cu-based bimetallic thin films using magnetron sputtering. Each sample set consists of seven electrodes with compositions ranging from 5 to 95 at.% of the secondary metal, enabling rapid screening of catalytic performance. Electrochemical NO_3RR measurements reveal that Cu-based bimetallic thin films exhibit a volcano-shaped dependence of ammonia Faradaic efficiency on alloy composition, suggesting the existence of an optimal balance between catalytic activity and surface stability. To understand the origin of this composition-dependent behavior, we performed a comprehensive structural characterization of the catalysts before and after electrochemical testing. Scanning electron microscopy (SEM) was used to monitor changes in surface morphology and roughness, while plasma focused ion beam (PFIB) milling was employed to prepare cross-sectional TEM lamellae for atomic-resolution analysis. High-angle annular dark-field scanning transmission electron microscopy (HAADF-STEM) was used to investigate grain boundaries and nanoscale structural evolution, whereas energy-dispersive X-ray spectroscopy (EDX) and electron energy-loss spectroscopy (EELS) provide elemental and chemical mapping of the bimetallic catalysts. These characterizations reveal the formation of a thin surface oxide layer, which likely corresponds to the electrochemically activated surface, as well as composition-dependent elemental redistribution that may contribute to the distinct catalytic behaviors observed across different Cu-based bimetallic systems

REVEALING DISSOLUTION AND REPRECIPITATION BEHAVIORS OF NANO-SCALE INERT PHASES IN LPBF STRUCTURES VIA EXTRACTION REPLICATION

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Inert particles like carbides, nitrides, and oxides significantly influence the microstructure and properties of alloys processed by fusion-based additive manufacturing, acting as inoculants, Zener pinning particles, and dispersoids that strengthen the material. To understand these phases' origins and evolution, detailed characterization of the feedstock, which is particularly challenging for laser powder bed fusion (LPBF) powders, as traditional thin-foil specimens require hours of focused ion beam preparation for each foil, target only a few particles, and are affected by matrix interference, so the nanoscale particle population in a powder lot has largely remained uncharacterized statistically. Here, it is demonstrated that these inert phases can be analyzed across populations through a multi-stage extraction replication process. Powder particles are embedded in electroplated Ni to create composite foils, which are then electropolished to expose particles in cross-section, leaving the inert phases in relief for extraction on carbon replicas. A single run yields thousands of matrix-free particles from many independent powder particles on one replica, enabling morphological, chemical, and crystallographic measurements on a much larger scale than serial thin-foil methods allow—facilitating automated data collection and analysis. This method was used to compare inert phases in powders and LPBF builds from a Ti-modified triplex steel. The only inert phase in the atomized powders is cuboidal TiN up to 150 nm across. In contrast, the builds contain smaller cuboidal γ-Al_2O_3/Ti(C,N) core/shell particles, free spheroidal γ-Al_2O_3 particles, and intermediate structures. This indicates that TiN dissolves in the melt during LPBF, γ-Al_2O_3 forms by reaction of Al with residual O, and upon cooling, it acts as a nucleant for Ti(C,N) precipitation, often forming a single-crystal cuboidal shell around the γ-Al_2O_3 core. These findings show how high-throughput extraction replication can reveal inert-phase development in LPBF builds and provide the statistical particle data needed for feedstock qualification and data-driven process optimization.

AUTOMATED SEGMENTATION AND VISUALIZATION OF SUBCELLULAR FEATURES IN HUMAN POSTMORTEM RETINAL TISSUE USING CRYOGENIC VOLUME ELECTRON MICROSCOPY AND MACHINE LEARNING

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We present a cryogenic volume electron microscopy (cryo-vEM) workflow for unfixed postmortem human retina that integrates high-pressure freezing, serial plasma-FIB/SEM imaging, image filtering, and automated segmentation. Sequential destriping, median filtering, and unsharp masking preserved subcellular features, including mitochondria's cristae, rhodopsin disks, and nuclear pores. An iterative human-in-the-loop nnU-Net bootstrapping strategy, seeded with complete mitochondrial annotations from three rod cells, reduced annotation effort by half and achieved a mean Dice score of 0.86. The model transferred successfully to an independently acquired volume. We segmented and morphometrically characterized 874 mitochondria across 47 rod cells. All data are publicly available at NanoAtlas (www.nanoatlas.org). Cryogenic volume electron microscopy (cryo-vEM) merges focused ion beam (FIB) milling with scanning electron microscopy (SEM) to visualize vitrified biological tissues, without chemical fixation, dehydration, or heavy-metal staining. We present a newly developed cryo-vEM workflow applied to postmortem human retina obtained at autopsy, encompassing high-pressure freezing, serial plasma-FIB milling combined with SEM imaging at 5x5x20 nm anisotropic voxel size, artifact correction, and automated segmentation. Sequential wavelet-Fourier destriping, median filtering, and unsharp masking suppressed milling and charge-induced artifacts while preserving membrane boundaries in rod cells. These computational procedures resulted in resolving mitochondria and their cristae topology, stacked rhodopsin discs, and nuclear pores in the nuclear membrane envelope. Starting from three manually annotated mitochondria in a subvolume (6.6 pm x 33.0 pm x 4.5 pm) of the first data set (63.1 x 51.2 x 5.1 pm), we trained nnU-Net models through an iterative human-in-the-loop bootstrapping strategy that progressed from 2D to 3D and reduced manual annotation time by approximately 50%. The final 3D model reached a mean Dice coefficient of 0.86 on withheld regions and transferred to a second, independently acquired volume containing the rod cell somata. Applying this workflow across the entire dataset 1, we segmented 874 mitochondria within 47 rod cells and quantified volume, surface area, and shape, revealing substantial morphological heterogeneity within individual inner segments. We further introduce NanoAtlas (www.nanoatlas.org), a web-based platform for interactive sharing of these volumes and the associated organelle annotations. Together, this study demonstrated the feasibility of using cryo-vEM as a practical route to nanometer-scale mitochondria analysis in unfixed human tissue. We hereby also embarked on generating a publicly accessible database of structural informatics on retinal mitochondria across physiological states, relevant to fundamental vision biology in health and disease.

A META-ALGORITHM FOR AB INITIO RECONSTRUCTION OF COMPLEX MIXTURES IN CRYO-EM

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We describe a systematic approach for spawning and aggregating multi-class cryo-EM reconstruction jobs. This approach formalizes standard ad hoc strategies of iterative classification and filtering typically used by practitioners to sort impure, heterogeneous samples. To our knowledge, this is the first method that can successfully perform *ab initio* reconstruction on datasets containing dozens of distinct species. We obtain 97% accuracy on *ab initio* reconstruction of a 45-class subset of Tomotwin-100, 75% accuracy on the full Tomotwin-100 dataset, and demonstrate recovery of ribosomal assembly states from an unfiltered experimental cryo-EM dataset. Our approach's capability scales with compute and lays the foundation for automated cryo-EM workflows in modern experimental settings.

NANOSCALE CHEMICAL IMAGING OF SINGLE CELLS WITH NEAR FIELD INFRARED SPECTROSCOPY

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Near-field Infrared (IR) spectroscopy provides a unique, label-free path to visualize small molecules inside single cells—such as metabolites and drugs—at resolutions far beyond the diffraction limit. This capability is potentially useful for multiplexed quantification of molecular distributions without the use of function-altering fluorescent tags. However, the complex physics of scanning probe detection has traditionally introduced significant variability resulting in a tradeoff between resolution and accurate chemical signals. To overcome this tradeoff, we developed a novel control strategy for near-field IR imaging that reduces measurement variability by 24x. We demonstrate this system's performance through multiplexed measurements of model cell samples, visualizing multiple molecular species simultaneously in their native state. Our second-generation system features seamlessly integrated hardware and controls, enabling data acquisition at the physical limits of the detector with improvements in reliability, functionality, sensitivity, and resolution. By bridging the gap between complex, high-resolution physics and quantitative biology, these advancements position label-free chemical imaging as a standard tool for uncovering molecular heterogeneity in single-cell biology.

VISUALIZING DEACETYLATION INDUCED SURFACE DEFECTS IN SCALABLE TOP-DOWN ISOLATION OF CHITIN NANOCRYSTALS

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The precise defect engineering of crystalline nanomaterials is a mature paradigm in solid-state chemistry. However, extending this control to soft matter, specifically the top-down isolation and modification of native biopolymer crystals like chitin, is hindered by a severe analytical gap. Conventional ensemble-average and dry-state characterization tools cannot resolve the delicate surface features of fragile organic crystals. Consequently, the relationship between harsh chemical processing and localized topological lattice disruptions remains largely unmapped. This study presents a methodology that bridges scalable polysaccharide isolation with high resolution imaging. To overcome the analytical barrier of observing these topochemically modified surfaces, in situ liquid-state high-resolution atomic force microscopy (HR-AFM) is deployed. Operating at sub-molecular resolution in the liquid state, this technique successfully captures previously obscured topological phenomena without the destructive artifacts of traditional dry-state methods. The HR-AFM imaging provides direct visualization of deacetylation-induced surface defects, revealing that topochemical treatments drive a stochastic surface chain dissolution that preferentially nucleates at pre-existing molecular defects along the fibrils. By resolving these structural alterations, this work establishes the molecular-level understanding necessary to correlate macroscopic processing conditions with localized surface topography and optimize the design of sustainable functional nanomaterials.

CRYO-EM BEYOND IMAGING: QUANTITATIVE REAL-SPACE STRUCTURAL ANALYSIS BY ELECTRON PAIR DISTRIBUTION FUNCTION

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Transmission electron microscopy (TEM) has evolved into an indispensable platform for imaging, crystallography, and chemical analysis at the atomic scale. Yet, much of the structural information contained in scattered electrons remains underexploited. Electron Pair Distribution Function (ePDF) analysis addresses this challenge by transforming electron diffraction into a quantitative real-space probe capable of determining local atomic arrangements beyond the limits of conventional crystallography. By exploiting both Bragg and diffuse scattering, ePDF enables structural characterization of nanocrystalline, amorphous, and highly disordered materials. Here, we discuss how recent advances in electron diffraction acquisition, data processing, and atomistic modeling have established ePDF as a robust tool for quantitative structural analysis in TEM. Rather than focusing on average crystal structures, ePDF provides direct access to short- and medium-range atomic order, opening new opportunities to investigate disorder, phase transformations, and complex structural evolution. Four representative case studies illustrate the breadth of this approach. First, we show how the combination of ePDF, high-resolution TEM, EXAFS, and first-principles calculations quantitatively revealed local structural rearrangements induced by ligand exchange in colloidal ZrO_2 nanocrystals. Second, we demonstrate how ePDF follows the structural evolution of highly disordered peroxide precursors during the synthesis of $BaSnO_3$ and $BaZrO_3$, revealing distinct crystallization mechanisms, including topotactic transformation and nucleation-and-growth pathways. Third, we present recent studies on glassy water, showing that cryogenic electron diffraction combined with real-space analysis enables quantitative characterization of frozen disordered matter. Finally, these examples collectively illustrate how ePDF complements high-resolution imaging and spectroscopy by providing structural information that cannot be obtained from Bragg diffraction alone. We propose that the next frontier of transmission electron microscopy is not simply higher spatial resolution, but richer quantitative structural information extracted from scattered electrons. Integrating imaging, spectroscopy, diffraction, and real-space structural analysis within a single experiment transforms TEM from a predominantly imaging technique into a comprehensive structural characterization platform. Extending this approach to cryogenic electron diffraction further expands the capabilities of Cryo-EM beyond imaging, enabling quantitative investigations of beam-sensitive, non-crystalline, and transient materials.

ATOMIC FORCE MICROSCOPY TO STUDY NEUROPROTECTION IN ALZHEIMER'S DISEASE.

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Alzheimer's disease (AD) is a neurodegenerative disorder characterized by progressive neuronal damage and memory loss, for which no cure currently exists. The amyloid-beta (1–42) peptide (Aβ) exerts neurotoxic effects primarily through non-specific interactions with neuronal cell membranes. Previously, we demonstrated that the changes in membrane composition play a crucial role in the mechanisms of amyloid toxicity (Drolle et al, PLOS ONE, 2017) which opens the possibility to protect them by altering membrane properties. In this work, we used model neuronal lipid membrane to study their interactions with Aβ using atomic force microscopy (AFM), Kelvin probe force microscopy (KPFM), nanopore electrophysiology and localized surface plasmon resonance (SPR). Our findings revealed that several membrane-active small molecules can alter lipid membrane properties and protect them from amyloid-induced damage. This includes melatonin—a neuroactive hormone (Mei et al., Nanotechnology, 2024), serotonin and trehalose (Xu et al., Biophys. J., 2024; ACS Omega, 2025; Membranes, 2025). Recently, we showed that psilocin, the active compound in "magic mushrooms," also exerts neuroprotective effects through membrane-level mechanisms. Collectively, these findings enhance our understanding of the critical role of lipid membranes in AD pathogenesis and may inform the development of novel preventive strategies centered on membrane protection.

EFFECT OF LI ISOTOPES ON POSNER MOLECULES AGGREGATION RESOLVED WITH ATOMIC FORCE MICROSCOPY AND CRYO- ELECTRON MICROSCOPY

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Posner molecules—calcium phosphate clusters (CaP) with the formula Ca9(PO4)6—have been theoretically proposed to act as biological qubits capable of sustaining long-lived quantum coherence. M. Fisher's hypothesis suggests these molecules facilitate quantum processing in the brain via the nuclear spins of phosphorus atoms (31P). Crucially, this model posits that lithium isotopes may modulate these effects, potentially explaining the element's profound impact on neurobiology and mood regulation. Recently, our team at the University of Waterloo discovered that two lithium isotopes, 6Li and 7Li, exert large and opposing effects on synaptic activity in rat brain tissues, supporting Fisher's hypothesis (Esmaeilpour, 2025). Despite their theoretical importance, the physical characterization of Posner molecules (PMs) remains a formidable challenge for microscopy. While larger amorphous calcium phosphate (ACP) capsules have been characterized using Atomic Force Microscopy (AFM), identifying the smallest PMs clusters is technically demanding. Previous work resolved the existence of tens of nm size ACP with cryo-EM (Straub 2023) and the NMR data indicated that their aggregation is affected differently by two Li isotopes (Straub, 2025). Building on previous work we used high-resolution AFM and cryo-EM to resolve the presence of smallest PMs and to evaluate if the ACP aggregation is affected by Li isotopes. Our data confirmed the presence of spherical clusters consistent with theoretically proposed PM dimensions (~1nm nm). Effect of Li isotopes on the size distribution has been analyzed and compared between the two isotopes and between the two microscopy methods. To move beyond topography and reveal their internal crystalline structure, integration of AFM with the chemical specificity of nano-spectroscopy is required. This combinatorial approach would provide definitive insights into the structural and compositional nuances of these putative biological qubits.

UNDERSTANDING THE DEGRADATION OF BATTERY ELECTRODES THROUGH IN SITU LIQUID-CELL SCANNING TRANSMISSION ELECTRON MICROSCOPY

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Understanding degradation processes at the electrode–electrolyte interface remains a major challenge for the development of next-generation lithium batteries. While post-mortem characterization has provided valuable information, many degradation pathways involve transient chemical and structural transformations that are difficult to capture ex situ. In situ liquid-cell scanning transmission electron microscopy (LC-STEM), combined with electron energy-loss spectroscopy (EELS) and electron diffraction, offers a unique opportunity to directly observe these dynamic processes in liquid environments with nanoscale spatial resolution. Here, we present recent advances in the development of LC-STEM methodologies for investigating battery materials in both aqueous and organic electrolytes. Using $LiCoO_2$ as a model cathode material, we directly visualized degradation processes in aqueous environments and correlated morphological evolution with chemical and structural transformations. Time-resolved LC-STEM reveals that chemical and electrochemical pathways simultaneously contribute for LiCoO2 degradation in aqueous electrolytes. Degradation primarily initiates at fracture sites and regions of increased surface roughness. An amorphous coating layer forms around LiCoO2 particles as they dissolve into the surrounding electrolyte. Spectroscopic data suggests that the loss of surface lithium weakens the cobalt-oxygen bonding within the layered lattice and unlocks a phase transition into a spinel cubic structure that precedes formation of surface cobalt oxide with a rocksalt structure. As cobalt oxide becomes mobilized into the aqueous medium, freshly exposed spinel lithium cobalt dioxide converts into cobalt oxide. This study demonstrates how combined in situ imaging, EELS, and diffraction can uncover transient degradation states that are inaccessible through conventional post-mortem approaches. Building on these insights, the LC-STEM methodology is being extended to commercially relevant high-voltage battery systems operating in organic electrolytes under air- and moisture-free conditions. We have adapted the experimental protocol for organic solvent environments and demonstrated the successful acquisition of real-time imaging, diffraction, and EELS chemical analysis during exposure and cycling of $LiMn_{1.5}Ni_{0.5}O_4$ (LNMO) cathodes to organic electrolytes. Ongoing work is focused on elucidating degradation pathways, interfacial reconstruction phenomena, and transition-metal redox evolution in LNMO/Li-metal cells. The latest results, including chemical and structural evolution under realistic battery conditions, will be presented and discussed. More broadly, this work establishes a versatile LC-STEM framework for investigating degradation mechanisms across a wide range of energy-storage materials and electrochemical environments.

THIOCYANATE ANIONS INDUCE HYPER-CONTRACTED CONFORMATIONS IN PRESTIN

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Prestin, a member of the SLC26A family of anion transporters is the membrane motor responsible for electromotility in outer hair cells. Recent CryoEM structures of prestin have suggested that its expansion and contraction within the membrane parallels the transporter cycle observed in other members of the family. CryoEM structures of prestin in the presence of SCN-, where large currents are observed in several of the SLC26A family members, including prestin, adopt an increasingly outward facing structure that contrasts with the inward facing and intermediate inward facing structures previously described in the presence of Cl-. Moreover, we find that these structures of prestin in SCN- to be more contracted than the hitherto described structures in Cl-. The contraction is in part brought about by an upward elevator like transporter movement of the core domain on the gate domain of its monomers that in turn causes a greater contraction of the transmembrane region closest to the cytoplasm. An additional contribution to the contracted state results from a reduction in the angle between each monomer of the dimeric structure with the monomers lying in closer approximation. These changes were reversed by salicylate that competes with SCN- at the internal binding site of prestin. The findings are discussed in the context of prestins gating charge movement (NLC) and SCN- associated currents, affinity for SCN-, and the sequence of movements in each transmembrane and cytoplasmic domain in the 7 distinct structures we identified.

IMAGING THE FORM AND COMPOSITION OF A BIOLOGICAL SPRING: STRUCTURAL AND MATERIAL CHARACTERIZATION OF THE CLICK BEETLE MESONOTUM

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Click beetles are among the many extraordinary animals that exhibit an actuation strategy known as latch-mediated spring actuation (LaMSA). The LaMSA components in click beetles are located within the hinge between their two primary body segments, enabling high accelerations and a powerful legless jump when the beetle is unconstrained. Among these components is the mesonotum, a saddle-shaped cuticle that acts as the primary spring. Although its geometry appears highly specialized for elastic energy storage, comparatively little is known about the microstructural and compositional specialization of the mesonotum cuticle itself. This study presents a process for characterizing the architecture and material composition of the click beetle mesonotum. Three species, Parallelostethus attenuatus, Melanotus similis, and Elater abruptus, are examined to investigate both individual and interspecific variation. Mesonotum are dissected, stained with osmium tetroxide, dehydrated through sequential ethanol treatment, critically point dried, and mounted in controlled orientations. Samples are first micro-CT scanned to capture three-dimensional geometry and internal structure. The mesonotum's are subsequently embedded in resin and sectioned across the sagittal plane using either a femtosecond laser or microtome. Femtosecond laser sectioning provides access to the internal cross-section while limiting the thermal and mechanical damage associated with conventional cutting techniques. A combination of environmental scanning electron microscopy (ESEM), energy-dispersive X-ray spectroscopy (EDS), confocal microscopy, Raman spectroscopy, and micro-CT is then used to examine cuticular architecture, internal layering, thickness, morphology, and chemical composition across the mesonotum. By combining three-dimensional imaging, targeted cross-sectioning, microscopy, and spectroscopy, this work establishes a framework for determining whether the mesonotum's exceptional spring function arises primarily from its geometry or from coupled specialization of geometry, microstructure, and material composition. Understanding this relationship provides insight into how biological systems engineer lightweight elastic structures and will inform the design of bioinspired energy-storage mechanisms, and ultimately bioinspired LaMSA robots and the study of all LaMSA organisms.

QUANTIFYING OPTICAL POWER DELIVERY AND PHOTOTHERMAL EFFECTS FOR IN SITU ENVIRONMENTAL TEM

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In situ transmission electron microscopy (TEM) enables direct observation of structural and chemical transformations under controlled stimuli and reaction environments. Extending these capabilities to light-sensitive materials is particularly relevant to photocatalysis, photovoltaics, and optoelectronics, where material behavior is strongly dependent on both the optical stimulus and local temperature. However, quantitative photoillumination within the TEM remains challenging. Optical losses along the illumination pathway and through the specimen environment can produce uncertainty in the power density delivered to the sample, while illumination-induced heating can obscure the distinction between photon-driven and thermally driven processes. Here, we introduce a methodology for quantitatively characterizing light delivery and photothermal effects within a closed-cell gas-phase TEM environment. Light spanning wavelengths from 350 to 2000 nm is introduced directly into a microelectromechanical systems (MEMS)-based reaction cell capable of maintaining controlled gas composition, pressures up to 1 bar, and elevated temperatures. Because optical power measured at the source does not necessarily represent the stimulus experienced by the specimen, a dedicated ex situ characterization workflow is used to quantify wavelength-dependent optical losses through the complete illumination pathway, including the holder and silicon nitride window. These measurements enable determination of the power density at the specimen position prior to in situ experimentation. A second challenge arises from photothermal heating of the MEMS device during illumination. The methodology therefore incorporates calibrated temperature measurements under illuminated and non-illuminated conditions to quantify the thermal contribution of the optical stimulus and compensate for illumination-induced temperature changes. This enables experiments to distinguish and independently control optical power density and specimen temperature rather than treating incident light intensity as the sole experimental variable. Together, these approaches establish a quantitative framework for controlling two critical parameters in photoinduced in situ TEM experiments: the optical power density reaching the specimen and the resulting photothermal response. By combining characterized illumination with controlled gas composition, pressure, and temperature, this methodology expands the range of experimentally relevant environments accessible for in situ and operando TEM studies of photocatalytic and other light-sensitive materials.

HIGH ENERGY-DENSITY GRAPHITIZATION OF BIOCARBON: COMPARISON OF SOLAR AND LASER PROCESSES

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Biomass graphitization allows both to valorize a waste, a non-edible and abundant resource, and to obtain high-value materials for a variety of applications. The main challenge resides in the high temperature (> 2500 °C) required to reach graphitization from resources classified as "hard carbons". The most common solution to this problem is the use of catalysts to provide a nucleation site for carbon to graphitize at lower temperature. In this solution, transition metals (Fe, Co, Ni), or more recently alkaline earth metals such as calcium (an earth abundant and non-toxic alternative) are used in an electrical furnace with low heating rate (< 60 °C/min). This process remains quite energy intensive and more rapid alternatives are thus proposed. High energy-density processes such as concentrated solar carbonization and laser graphitization are therefore explored. They both allow to rapidly reach high temperature, allowing a direct structuration of carbon from amorphous or turbostratic structure to graphitic. Here, the study focuses on spent coffee grounds valorization. The biomass is first pyrolyzed at 800 °C to obtain a carbon-rich biocarbon with low carbon structuration. The graphitization step is then carried out either using concentrated solar or scanning laser beam at around 1800 °C. The resulting carbon structure is investigated at local scale using Raman spectroscopy and Transmission Electron Microscopy (TEM). The TEM images show long graphene layers (> 30 nm) arranged in large stacks of around 20 graphene layers. This presence of graphitic structure and stacking quality are confirmed in Raman spectroscopy where the characteristic bands D (1345 cm-1) and G (1591 cm-1) appear. The ID/IG ratio is around 1.3 for laser graphitization, with a shoulder on the G band (presence of D' band at 1620 cm-1). For solar carbonization, this ratio is closer to 1.0 but with an important D'' band around 1495 cm-1 and no D' band. The different heating method therefore impacts the graphitization in itself but also the type of defects that appear or heal. Also noteworthy, the 2D band around 2700 cm-1 is more pronounced in the case of laser, emphasizing the important stacking through this process. Doping the biomass with calcium allowed a better graphitization (lower ID/IG) and the difference in defect types remained. This works highlights the effect of heating method on structure and characteristics of biocarbon during the graphitization using fast (solar) and flash (laser) localized heating.

AUTOMATED MICROSTRUCTURAL METROLOGY OF ADDITIVE MANUFACTURED AL2O3 USING DEEP LEARNING SEGMENTATION

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Automated microstructural analysis utilizing a supervised U-Net-based convolutional neural network (CNN) deep learning (DL) approach to the segmentation and analysis of stereolithography-produced green body Al2O3 is the focus of this study. It has the potential to provide insight into how changes to processing and production can be optimized by looking at microstructures and measuring outcomes of changes to the system. The techniques of analysis remain the same as historical methods, scraping information from microstructures and understanding phase differences, particle morphology, defects, crack propagation, and other microscopic differences in materials. The system of learning, segmenting, and measuring can be automated allowing the researcher to focus less on attaining the information and more on determining how to make changes to optimize systems. Building a supervised DL model provides a reproducible, non-human-biased means to gather data from multiple images and multiple regions of each image and look for observable patterns and thus processing decisions based on them. In this research, additive manufactured Al2O3 was studied, cure powers were modified, and the layers and interlayer areas were investigated for microstructural changes. The result is a series of measurements discussed later that demonstrates the data derived from these samples and the judgments that are possible because we have these data. Subsequently, decisions on changes in processing or material are possible because the model is reproducible, automated, and representative of changes or optimization of materials and processing methods.

CAPTURING DYNAMIC GATING INTERMEDIATES AND PHARMACOLOGICAL BLOCKADE OF THE KIR7.1 CHANNEL VIA SINGLE-PARTICLE CRYO-EM

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The inward rectifier potassium channel Kir7.1 (KCNJ13) plays an indispensable role in regulating membrane potential across diverse tissues, including the retinal pigment epithelium and uterine smooth muscle, and serves as a critical effector of G-protein-independent melanocortin-4 receptor (MC4R) signaling within hypothalamic circuits that regulate energy metabolism. Mutations in Kir7.1 cause severe ocular channelopathies, such as Leber congenital amaurosis and snowflake vitreoretinal degeneration, yet the structural mechanisms governing its dynamic gating cycle remain incompletely defined. Here, we utilize single-particle cryo-electron microscopy (cryo-EM) to resolve human Kir7.1 across multiple functional states. We reveal that a retinitis pigmentosa-associated R162Q variant stabilizes an open-conducting, docked architecture supported by defined lipid contacts. Conversely, binding of the inhibitor ML418 disrupts a coupled transmembrane network, driving cytosolic domain (CTD) translation and rotation while constricting the helix-bundle crossing. Furthermore, structural analysis of an E276A mutant captured a previously unseen transient gating intermediate featuring an open-like transmembrane domain decoupled from a detached CTD. By resolving the channel in inhibited, open, and decoupled states, this work delineates the specific conformational rearrangements that dictate Kir7.1 gating and its susceptibility to pharmacological blockade.

MULTICOLOR ELECTRON MICROSCOPY VIA CATHODOLUMINESCENCE

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Electron microscopy and optical microscopy provide complementary windows into biology and materials. But combining structural resolution with molecular specificity remains a central challenge. I will describe our efforts on developing cathodoluminescence electron microscopy, in which an electron beam locally excites light emission with nanometer-scale confinement. This approach enables far-field spectral readout while preserving the nanoscale resolving power of electron microscopy. Because cathodoluminescence and scattered electron signals are recorded simultaneously, cathodoluminescence electron microscopy eliminates the need for correlation and registration steps of conventional correlative light-electron microscopy and is inherently free of optical aberrations. I will discuss our work using cathodoluminescence to probe energy transfer in single lanthanide nanocrystals. I will also show that widely used small-molecule organic fluorophores exhibit cathodoluminescence with spectra closely matching their fluorescence, establishing them as practical "cathodophores" for multicolor biological imaging. Finally, I will outline our ongoing efforts to translate cathodoluminescence to 3D imaging of cryogenic and resin-embedded specimens, enabling multicolor electron microscopy reconstructions of cells and tissues with full ultrastructural context.

DYNAMICS OF THE HIV-1 ENVELOPE MEMBRANE-PROXIMAL EXTERNAL REGION ON VIRIONS

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Despite HIV-1 Env's immune evasion through high sequence variation and a dense glycan shield, some infected individuals develop broadly neutralizing antibodies (bnAbs) that target conserved elements of Env and neutralize diverse HIV-1 strains. Antibodies targeting the gp41 distal membrane-proximal external region (MPER) are among the most broadly reactive bnAbs. The MPER is a highly conserved linear segment of the gp41 ectodomain situated immediately N-terminal to the transmembrane domain. This location places the MPER epitope in close proximity to, or partially embedded in, the viral membrane, and MPER bnAbs often engage both the Env epitope and lipid components during recognition(16-20). This dual recognition feature has limited the clinical potential of certain MPER bnAbs, such as 2F5 and 4E10, due to increased polyreactivity and immune tolerance constraints. By contrast, DH511 and 10E8 recognize distal MPER bnAb epitopes overlapping that of 4E10 but exhibit lower polyreactivity and greater neutralization potency(20). These properties have renewed interest in the MPER as a vaccine target, and recent immunization studies in humans suggest that polyreactive MPER antibodies recognizing both lipids and gp41 polypeptide can be elicited. Structural studies using purified Env embedded in nanodiscs have provided snapshots of MPER antibody recognition. These studies suggested that a single antigen-binding fragment (Fab) can access the partially membrane-embedded epitope through a tilted Env conformation, and three Fabs can bind quasi-symmetrically by partially lifting the trimer out of the membrane. However, how MPER antibodies engage Env on native virions, and how this interaction interferes with receptor-triggered entry, remain incompletely understood. Here, we use cryo-electron tomography (cryo-ET) to define how the distal MPER bnAb DH511 recognizes Env on intact HIV-1 virions. We show that DH511 engages Env asymmetrically, with three Fabs binding one side of the trimer and stabilizing a highly tilted Env ectodomain. Density-guided modeling suggests that this one-sided three-Fab engagement requires asymmetric rearrangement of the Fab-bound MPER relative to the Env ectodomain and is associated with local viral membrane deformation. DH511-bound Env remains competent for CD4-induced opening and can adopt a quasi-symmetric Fab-bound state with reduced membrane deformation. At viral fusion interfaces, DH511 Fab and IgG increase viral–target membrane spacing and reduce Env–mCD4 accumulation at the interface center. Our study links MPER accessibility to the conformational dynamics of native Env on the viral membrane, reveals the structural plasticity of MPER bnAb recognition, and suggests a previously unrecognized mechanism by which MPER bnAbs alter viral–target membrane apposition and reshape Env spatial organization at the fusion interface during HIV-1 entry.

ORGANELLE-SCALE SPIRAL FILAMENTS OF A MALARIA PARASITE DYNAMIN-RELATED PROTEIN

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Plasmodium falciparum, the parasite responsible for over 200 million malaria infections and approximately 600,000 deaths annually, relies on rapid and highly coordinated cell division for its propagation and infectivity. Division of the parasite requires division of the parasite's single-copy endosymbiotic organelles, the mitochondrion and apicoplast, and their distribution to daughter parasites. While the division is essential for *Plasmodium* replication and infection, the molecular mechanism(s) governing it remains poorly understood. Here, we employ single-particle cryo-electron microscopy (cryo-EM) and cryo-electron tomography (cryo-ET) to characterize structural states of a key organelle fission protein. This protein assembles as filamentous spiral scaffolds on membranes and catalyzes membrane fission. We also tested the in vivo relevance of these conformational states: parasites carrying mutations at key residues showed disrupted protein-lipid interactions and impaired segmentation and fitness. These findings contribute to our understanding of the molecular machinery underpinning endosymbiotic organelle division in malaria parasites.

LEVERAGING COMPLEMENTARY ELECTRON MICROSCOPY AND MICROANALYSIS TECHNIQUES TO REVEAL LOCALIZED STRUCTURE, CHEMISTRY, AND FUNCTIONALITY IN COMPOUND SEMICONDUCTORS

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Electron–specimen interactions generate a rich variety of signals that provide complementary insight into a material's structure, chemistry, and functionality. As a result, modern electron microscopy has evolved beyond imaging alone into a multimodal characterization platform capable of probing atomic-scale structure, local composition, optical behavior, and electronic functionality across multiple length scales. When combined with advanced microanalytical techniques and computational data analytics, these approaches enable a more complete understanding of complex semiconductor systems than can be achieved through any individual method alone. This work highlights correlative characterization workflows developed for compound semiconductor materials and devices. Functional- and defect-sensitive imaging modalities are first employed to identify localized regions of interest associated with optical, electrical, and structural anomalies. Site-specific specimen preparation via focused ion beam (FIB) techniques subsequently enables targeted nanoscale and atomic-scale characterization, while complementary microanalytical methods, such as APT, provide quantitative compositional information and three-dimensional chemical mapping. Advanced computational analysis of electron microscopy datasets further expands these capabilities by revealing subtle structural features, improving measurement repeatability, and enabling statistically robust evaluation of complex microstructures. Through the correlation of these datasets, direct relationships can be established among local functionality, crystal structure, defects, bonding, dopant distributions, and impurity incorporation. Representative examples include investigations of impurity segregation in FACELO AlGaN/GaN epitaxial structures, dopant diffusion along individual dislocations, polarity inversion in N-polar AlN associated with oxygen incorporation, failure analysis of GaAs-based VCSEL devices, and structure–function correlations in light-emitting device architectures. Together, these studies demonstrate how targeted, multimodal characterization workflows spanning multiple length scales and sensitivity regimes can uncover materials insights that are inaccessible through individual characterization techniques alone. Beyond advancing scientific understanding, these approaches provide practical solutions to real-world characterization challenges encountered in semiconductor research, process development, failure analysis, and manufacturing.

BIO-ENGINEERED LAMOTRIGINE MICROPARTICLE: GREEN SYNTHESIS, PHYSICOCHEMICAL CHARACTERIZATION AND IN-VIVO ANTISEIZURE EVALUATION

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Poor aqueous solubility, limited drug loading, and uncontrolled release of lamotrigine (Lamo) remain major challenges in antiepileptic therapy, often resulting in suboptimal seizure control and dose-related neurotoxicity. In this study, these limitations were addressed by developing polymer-engineered calcium carbonate ($CaCO_3$) microparticles with layer-by-layer (LbL) surface modification to enhance drug encapsulation and regulate release behavior. Gelatin-based (GC) and Soluplus-based (SC) $CaCO_3$ microparticles were prepared by controlled co-precipitation, followed by sequential deposition of sodium alginate and cationic guar gum layers. Morphological and surface characterization using SEM revealed discrete, uniformly shaped uncoated microparticles, while LbL coating produced increased particle dimensions and roughened surfaces, confirming multilayer formation. Nitrogen adsorption–desorption analysis showed formulation-dependent modulation of porosity: GC microparticles exhibited a BET surface area of 22.99 m^2/g and pore volume of 0.115 cm^3/g, which were retained after coating, whereas SC microparticles displayed a substantial increase in surface area from 0.403 to 8.797 m^2/g and pore volume from 0.0028 to 0.0917 cm^3/g following LbL assembly. Zeta-potential measurements demonstrated alternating surface charge oscillation between approximately −26 and −4 mV, confirming controlled electrostatic self-assembly. LbL coating significantly enhanced lamotrigine loading, with LBL-GC achieving the highest drug content (7.8 ± 0.8%). In vitro release studies demonstrated sustained, diffusion-dominated lamotrigine release (Higuchi model, $R^2 = 0.77–0.85$), while in vivo evaluation showed that Lamo-LbL-SCproduced dose-dependent suppression of seizure activity without impairment of motor coordination or locomotor behavior. These findings demonstrate that LbL-engineered $CaCO_3$ microparticles effectively improve lamotrigine loading, control release kinetics, and enhance anticonvulsant efficacy through rational carrier design.

CRYOGENIC IR NANOSCOPY FOR MAPPING SMALL-MOLECULE METABOLISM WITHIN CELLS

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The native localization of small molecules within cells is central to metabolism, signaling, and regulation, yet it remains difficult to measure directly at the nanoscale. Existing imaging methods cannot yet provide the necessary combination of molecular specificity, near-native preservation, and nanometer-scale spatial resolution. Molecular labels are available for only a limited set of molecular targets and can perturb the cellular environment, while high-resolution structural methods generally do not directly report the chemical identity of small metabolites in their native environment. This project addresses this gap by developing cryogenic atomic force microscopy-infrared spectroscopy (cryo-AFM-IR) for label-free nanoscale chemical imaging of small molecules in cryopreserved cells in their native state. Cryo-AFM-IR combines the high spatial resolution of AFM with the chemical sensitivity of infrared absorption spectroscopy. In this approach, rapidly frozen cells are cryo-sectioned to preserve near-native molecular distributions while exposing the cell interior to the AFM probe. A tunable mid-infrared laser is focused on the sample near the AFM tip, and local absorption is detected via the sample's photothermal response beneath the cantilever. Because the signal is generated locally at the tip-sample contact, cryo-AFM-IR can map infrared absorption well below the diffraction limit of conventional IR microscopy while keeping the sample in a preserved cryogenic state. The current work focuses on establishing a cryo-AFM-IR capability as a functional microscope for biological sections. This includes integrating cryogenic AFM imaging with infrared excitation, developing cryogenic sample-preparation and section-transfer workflows, and implementing resonance-enhanced photothermal detection for small-molecule detection. Achieving reliable cryo-AFM-IR imaging would provide a unique route to link nanoscale structure with native local chemical information in cryopreserved cells. Our proof-of-concept system is Chlamydomonas reinhardtii, a model green alga with highly organized carbon metabolism. These algae account for one-third of global carbon fixation by concentrating CO2 in an organelle called the pyrenoid. We are particularly interested in mapping CO2 and related carbon-fixation metabolites within and around this structure. These measurements could reveal how algae organize CO2 uptake at the nanoscale and may inform future strategies to improve photosynthetic efficiency. By establishing cryo-AFM-IR for cellular sections, this work aims to provide a practical first step toward direct nanoscale imaging of small-molecule metabolism in near-native biological environments. Once established, this platform could also enable more advanced infrared approaches, such as broadband dual-comb near-field spectroscopy and quantum-based molecular discrimination.

KRIOS 5 CRYO-TEM REVEAL THE INTRICACIES OF LIFE WITH UNPRECEDENTED FIDELITY AND THROUGHPUT

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In recent years, cryo-electron microscopy (cryo-EM) single-particle analysis (SPA) has benefited from a multitude of technological advances, culminating in the ability to resolve biological macromolecules at atomic resolution. These advances continue to drive rational structure-based vaccine design and therapeutic development by enabling rapid, high-throughput screening and data collection.

More recently, cryo-electron tomography (cryo-ET) has become an indispensable tool for studying macromolecular structures in their native cellular context. As cryo-ET applications continue to expand, improving workflow connectivity, automation, and data quality is becoming increasingly important. Our ongoing efforts focus on further streamlining the workflow between cryo-FIB sample preparation and cryo-TEM data collection, making tomography workflows safer, faster and cleaner. To address bottlenecks associated with data acquisition and processing, the integrated **Cryo Tomo Software Suite**, combining Thermo Scientific Tomo 5, Tomo Multigrid and Tomo Live, provides automation capabilities for pre-acquisition tasks, enhances productivity through predictive tracking and enables rapid quality assessment. Robust and optimized motion correction helps produce crisp tomograms with the image quality needed for subtomogram averaging. Results are presented with new quality metrics on a web-based platform, providing an intuitive user experience and facilitating advances in structural biology.

In this poster, we highlight the latest Thermo Fisher Scientific innovations in **300 kV cryo-electron microscopy with Krios 5**, demonstrating advances in both SPA and cryo-ET workflows designed to improve efficiency and performance.

ELUCIDATING ORGANIC THIN-FILM CRYSTALS VIA SCANNING PROBE MICROSCOPY

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The electronic properties of crystalline organic thin films depend critically on the solid-state packing and nanoscale morphology. Atomic Force Microscopy (AFM) offers the distinct advantage of enabling direct, real-space, three-dimensional visualization of these nanoscale features without destroying the delicate organic layers. We employ AFM to explore the complex crystallization behaviors and morphological evolution across three distinct organic thin-film systems: molecularly templated chiral organic small molecules, solid-phase epitaxy (SPE) films, and doped crystalline NPB layers. In the chiral BINAP system, we utilized molecular epitaxy to investigate the crystal structure of the platelet-forming scalemic BINAPs. High-resolution AFM topography reveals the epitaxial relationship established at the organic-organic interface. This allows us to understand their crystal properties with detailed crystallographic precision, leading us to the discovery of a "kinetic trapping" crystallization of specific enantiomeric packing configurations. Solid-phase epitaxy (SPE) is an advanced technique used to transform an amorphous organic precursor film into a highly crystalline state via rapid, mild thermal annealing, guided by an underlying crystalline template. AFM provides a direct visualization of this amorphous-to-crystalline phase transition at the nanoscale, capturing the growth of crystalline domains and the propagation of crystallization fronts to extract valuable information regarding roughness evolution and the overall crystallization process. Scanning probe investigations are further extended to assess the morphological impact of molecular doping in the hole-transporting material NPB. By analyzing the nanoscale topography of doped NPB films, we evaluate dopant dispersion, potential dopant-induced aggregation, and structural perturbations within the host matrix. This allows us to map the microstructural homogeneity of the doped films at the nanoscale. Collectively, these studies highlight the indispensable role of scanning probe microscopy in advancing our fundamental understanding of organic thin-film engineering. By resolving the specific nanoscale features of molecularly templated BINAP, SPE-grown crystals, and doped NPB layers, we demonstrate how local structural elucidation directly informs the precise design of organic semiconductor interfaces for next-generation optoelectronics.

CRYO-AFM SUBTRACTIVE NANOTOMOGRAPHY OF VITRIFIED BIOLOGICAL SAMPLES FOR STUDYING INTRACELLULAR MORPHOLOGY

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Correlative cryogenic microscopy has transformed our ability to locate molecular targets and visualize intracellular structures. Cryo-CLEM and integrated light-ion-electron microscopes guide three-dimensional targeting and lamella preparation for tomography, while serial cryo-FIB-SEM extends structural imaging into cellular volumes. Yet the structural information from a freshly exposed, unstained vitrified biological block face remains predominantly electron-derived, which is intrinsically challenged by weak biological contrast, charging, and milling-related artefacts. Here we introduce atomic force microscopy as an active block-face imaging technique and establish a route towards cryo-AFM-enabled subtractive nanotomography. A tip-scanning AFM using custom self-sensing polymer cantilevers integrated directly within a cryo-FIB-SEM interrogates freshly milled surfaces of vitrified biological cells. AFM measurements reveal intracellular organelles, membrane boundaries, and associated structures at an organelle-scale morphology that are weakly represented in corresponding SEM images. Beyond providing quantitative nanoscale topography, AFM introduces a complementary surface-sensitive contrast on the vitrified block face, allowing label-free imaging of intracellular morphology. Combining AFM imaging with successive FIB milling extends this information into three dimensions. FIB provides controlled axial progression through the cellular volume, complemented by AFM imaging of nanoscale height information from each newly exposed surface. Registration across successive block faces imaged with FIB/SEM/AFM provides coarse-to-fine subtractive nanotomography. Cryo-AFM-FIB/SEM thus provides a route from correlative cryogenic volume imaging towards multimodal nanotomography of native biological matter.

CLEAM: CORRELATIVE LIGHT, ELECTRON, AND ATOMIC FORCE MICROSCOPY FOR MULTIMODAL ULTRASTRUCTURAL VOLUME IMAGING OF TISSUE

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Understanding tissue architecture requires connecting molecular identity with ultrastructural context and nanoscale morphology. However, no single imaging modality can provide all three with sufficient resolution and contextual integrity. Fluorescence microscopy (FM) provides molecular identity and nanoscale localization of proteins within complex tissue environments, albeit at poor resolution. Back-scattered electron imaging inside a scanning electron microscope (SEM) provides ultrastructural context; however, it lacks specificity and has limited contrast. Atomic force microscopy (AFM) has been extensively used to study proteins and molecules in-vitro, at nanometric resolution; however, it is limited to surfaces. Here, we present CLEAM (Correlative Light, Electron and Atomic Force Microscopy), a correlative workflow that integrates the three microscopy modalities to image Tokuyasu cryo-sections for multimodal and volumetric tissue imaging. Tokuyasu cryo-sectioning is known to preserve antigenicity in tissues while maintaining their ultrastructural integrity. This enables high-specificity labeling for fluorescence imaging prior to light fixation for SEM imaging and force-based topographic mapping. Following fluorescence imaging, individual sections are correlated with backscattered-electron SEM to establish ultrastructural context and subsequently interrogated by an AFM integrated within the SEM chamber. The large field of view of SEM enables reproducible navigation to regions of interest, while AFM provides quantitative nanoscale topography from the corresponding section surface. We use this correlative workflow to study mitochondrial organization in the outer plexiform layer of zebrafish larval retina tissue. Through accurate registration across the three modalities and serial Tokuyasu cryo-sections, CLEAM generates aligned datasets that integrate protein localization, ultrastructure, and topographical contrast within the same volumetric framework. Thus, CLEAM establishes a scalable platform for correlative nanoscale analysis of biological tissues, with broad applicability in structural cell biology and pathology.

MULTISCALE MICROSCOPY OF MONOLAYER TWO-DIMENSIONAL SEMICONDUCTORS

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Layered two-dimensional (2D) semiconductors such as transition metal dichalcogenides (TMDs) are promising building blocks for next-generation electronics and optoelectronics. Because the electronic properties of TMDs depend strongly on their layer number, precise thickness control is essential. Despite substantial progress in growing uniform TMD films, achieving self-limiting monolayer growth has remained an unsolved challenge. Here we present the self-limiting synthesis of monolayer TMDs, including WS_2 and WSe_2, in which multilayer formation is suppressed while high monolayer crystallinity is maintained. Electron microscopy and optical spectroscopy techniques confirm the high quality of the synthesized monolayer across seven orders of magnitude in length scales, from atomic scale to 2-inch scale. At the atomic scale, integrated differential phase contrast (iDPC) imaging resolves the monolayer crystal structure, while high-angle annular dark-field scanning transmission electron microscopy (HAADF-STEM) reveals crystalline domains with well-stitched grain boundaries. Scanning electron microscopy (SEM) characterizations directly tracks the growth evolution and provide evidence for self-limiting behavior. Widefield Raman and photoluminescence imaging reveal spatially uniform monolayer signatures across 2-inch substrates. Field-effect transistor measurements further demonstrate the high electrical performance of the synthesized monolayer films. Together, these multiscale measurements connects atomic structure, macroscopic uniformity, and device performance, establishing a self-limiting growth approach for high-quality monolayer semiconductors.

SIMULTANEOUS ATOMIC FLAT HYDROPHOBIC AND HYDROPHILIC NANOTERRACES FOR BIOMOLECULES SPM OBSERVATION, ANALYSIS, AND TESTING

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The knowledge at the single-molecule level of biomolecules nanomorphology and physico-chemical properties is of paramount importance in a wide variety of fields, including biology, medicine, nanobiotechnology, environment and material science. Here, we report on an innovative technology and material to produce SPM atomic flat substrates to be used for biomolecules deposition and the successive investigation of biomolecules morphological, physical and biochemical properties at the nanoscale. The technology we present allows to produce atomic flat substrates, for SPM analysis, that can show simultaneously atomic flat hydrophobic and hydrophilic nanoterraces. The innovative technology is based on the cleavage of a specific Mg-Al-hydroxysilicate [1]. This hydroxysilicate is a very particular phyllosilicate. After cleavage, it presents both positively charged hydrophobic areas, and negatively charged hydrophilic ones, related to its particular and specific crystal structure. Both areas are atomic flat (~ 0.2 Å rms) and can be extended from few nanometres to millimetres. These areas can be used for biomolecules deposition, that can be performed without any need of additives or substrate surface modification. SPM characterization of biomolecules physical properties such as, dimensions and sizes, average polarity, hydrophobicity/hydrophilicity character as a function of physico-chemical conditions (pH, ionic strength, temperature, pressure), down to the nanoscale, has been performed. In more details, the present technology has been efficiently used to study and characterize by SPM, at the nanoscale, single nucleotides, DNA and RNA filaments, amino acids, red blood cells, proteins molecular changes induced by oxidation and oxidative stress, and also DNA stretching and DNA-protein interaction at the single filament level [2, 3]. In addition, to understand the interaction between the specific surfaces and the biomolecules, quantum mechanics simulations at the DFT/B3LYP level of theory have been performed. [1] D. Moro, G. Ulian, G. Valdrè, 3D meso-nanostructures in cleaved and nanolithographed Mg-Al-hydroxysilicate (clinochlore): Topology, crystal-chemistry, and surface properties. Applied Clay Science 169, 74-80 (2019). [2] M. Pignataro, G. Di Rocco, L. Lancellotti, F. Bernini, K. Subramanian, E. Castellini, C. A. Bortolotti, D. Malferrari, D. Moro, G. Valdrè, M. Borsari, F. del Monte, Phosphorylated cofilin-2 is more prone to oxidative modifications on Cys39 and favors amyloid fibril formation. Redox Biology 37, 101691 (2020). [3] D. Moro, G. Ulian, G. Valdrè, SPM of layer silicates and interaction with single biomolecules at the nano-atomic scale. Conference of the Italian Society of Mineralogy and Petrography (SIMP). 3-5 September 2024, Bari, Italy. Page 586, doi.org/10.3301/ABSGI.2024.02.

THE WAFFLE METHOD ON THE EM ICE HIGH PRESSURE FREEZER

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The Waffle Method, when integrated with the Leica EM ICE high-pressure freezer, provides a physically robust strategy for vitrifying thick or structurally complex biological specimens for cryo-electron tomography (cryo-ET). By incorporating an electron microscopy (EM) grid directly into the high-pressure freezing (HPF) assembly, this approach hybridizes grid-based sample handling with the advantages of HPF for thicker samples. In this workflow, biological material is placed onto a carbon-coated EM grid and confined between modified carriers that define a controlled sample thickness. Under high pressure, conductive cooling is significantly enhanced while high pressure suppresses ice nucleation, enabling rapid and uniform vitrification of samples otherwise inaccessible to plunge freezing. The resulting vitrified "waffle" is supported by the grid squares, where grid bars serve not only as spatial constraints but also as reinforcing elements during downstream processing. Following vitrification, cryo-focused ion beam (cryo-FIB) milling benefits from this engineered geometry. The introduction of trenches and stress-relief notches enables controlled material removal, producing lamellae of 100–200 nm thickness with improved structural integrity. The mechanical stabilization imparted by the grid significantly reduces lamella deformation and failure, increasing both survivability and throughput across a wide range of challenging specimens, including microsporidian spores, yeast, bacteria, mammalian cells, and macromolecular assemblies. Overall, this integration of the Waffle Method with the Leica EM ICE supports HPF grids as both vitrification substrates and mechanical stabilizers, enabling reproducible preparation of thick specimens and expanding the practical limits of cryo-FIB-SEM lamella fabrication for cryo-ET.

ADVANCES IN AUTOMATION AND IMAGING DURING ULTRAMICROTOMY

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Ultramicrotomy remains a core component in many transmission electron microscopy (TEM), volume electron microscopy (volume EM), and correlative light and electron microscopy (CLEM) workflows, despite also remaining a step requiring significant user skill. Challenges associated with instrument setup, precise localization of subsurface regions of interest, sectioning-induced deformation, and inconsistent serial section production can limit imaging efficiency, data quality, and reproducibility. Leica Microsystem's Enuity ultramicrotome supports workflows frustrated by these problems through the integration of digital knife control, automation during knife alignment and trimming, multimodal imaging, and automated coordinate registration. Existing workflows are improved through better use of preliminary structural data, targeting material removal during trimming and sectioning by fluorescence, and streamlined serial sectioning collection. Micro-computed tomography (micro-CT)-guided trimming exemplifies a shift toward data-driven specimen preparation by incorporating non-destructive volumetric imaging prior to sectioning. Three-dimensional X-ray datasets provide spatially resolved information that can be registered to the embedded specimen and translated into trimming coordinates, allowing targeted material removal based on internal sample architecture rather than external visual cues alone. By reducing iterative trimming and improving block-face alignment with the desired cutting plane, micro-CT integration enhances targeting accuracy and preserves valuable specimen volume for downstream EM acquisition. Similarly, fluorescence-guided ultramicrotomy extends the principles of correlative imaging into the sectioning process itself. Real-time optical imaging of labeled features during trimming and ultrathin sectioning provides continuous feedback on specimen position, enabling dynamic adjustment of block orientation and cutting depth. Integration of fluorescence signals with sectioning workflows increases confidence in targeting rare or spatially restricted structures, reduces operator-dependent decision making, and improves workflow efficiency in CLEM applications by facilitating accurate correlation between light microscopy and subsequent ultrastructural imaging. Advances in serial section production also result from this automation and closed-loop control strategies designed to improve section quality and consistency through the mitigation of mechanical and environmental variability that contribute to compression, folding, section loss, and registration errors. These innovations support the generation of longer, more uniform serial-section datasets while reducing the need for manual intervention, thereby improving the reliability and throughput of three-dimensional EM reconstructions. By combining multimodal imaging, coordinate-aware targeting, environmental control, and automation, next-generation ultramicrotomy platforms are becoming increasingly capable of delivering reproducible, high-fidelity sections that support scalable volume EM and CLEM studies. Such integration not only improves sectioning precision and data quality but also enhances workflow efficiency, reproducibility, and the effective utilization of downstream electron microscopy resources.

CRYO-EM STRUCTURES OF BRAIN-ISOLATED G PROTEIN-COUPLED RECEPTORS

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Glutamate is the main excitatory neurotransmitter in the brain and mediates its actions by both ionotropic (e.g. NMDA and AMPA) and metabotropic glutamate receptors (mGluRs). The Groups II and III mGluRs, which pre-synaptically inhibit glutamate release, are important for synaptic plasticity, modulating neuronal excitation, learning and memory. Our current understanding of the structural organization and dynamics of these and other mGluRs, as well as most other GPCRs, relies mainly on studies using recombinant and highly engineered systems in vitro. Here, we combine CRISPR-mediated protein tagging, proteomics and a rapid immunoaffinity purification method to isolate endogenous mGluR2-containing assemblies from mouse brain and visualize them via cryo-EM. Analysis of the particle sets reveals the molecular structures of at least 11 distinct endogenous receptor assemblies that span active and inactive states, homomeric and heteromeric dimers, and G protein-coupled and uncoupled species. We find that mGluR2 homodimers and mGluR2/3 heterodimers are the major endogenous mGluR2-containing species present in the brain, with the mGluR2/3 heterodimers detected only in active state complexes, potentially reflecting basal activation of mGluR3 containing dimers by chloride. Reconstructing a comprehensive conformational equilibrium for the brain-isolated receptors in detergent reveals endogenous ternary complexes comprising mGluR2 homodimers and mGluR2/3 heterodimers with a single GαoA heterotrimer which exhibit significant differences from prior studies with recombinant systems. Our work illuminates the endogenous conformational, proteomic and compositional landscape of the heterogenous mGluR2 complexes in the brain, thereby providing a structural framework for the pathophysiology of psychiatric disorders. This information has the potential to be leveraged for therapeutic targeting of endogenous glutamatergic signaling complexes.

ACCELERATING CROSS-PLATFORM ELECTRON MICROSCOPY AUTOMATION WITH AUTOSCRIPT

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Electron microscopy increasingly depends on reproducible automation that can be adapted across instruments. Thermo Scientific AutoScript is a Python-based API for microscope control on SEM, DualBeam FIB-SEM, and TEM platforms. Its consistent programming framework allows users to develop customized recipes for image acquisition, electron and ion beam control, stage positioning, detector operation, and patterning. Because these workflows are implemented in Python, microscope control can be combined directly with image processing, data analysis, and machine learning.

AutoScript supports applications ranging from repetitive procedures and parameter sweeps to feature tracking, high-throughput acquisition, and adaptive patterning. We highlight several examples, including deep-learning-based particle segmentation and quantitative feature analysis in SEM images, as well as neural-network-assisted atom detection in HAADF-STEM images acquired on a TEM. We also discuss EMCoPilot, a Python-based proof-of-concept application that explores how multimodal large language models, image segmentation, and AutoScript commands could support natural-language-guided and adaptive FIB-SEM workflows.

AI coding assistants such as Codex and Claude may reduce the time required to develop, understand, test, and revise AutoScript scripts when used with development environments such as VS Code and PyCharm. AutoScript can also be used alongside established applications such as Maps. Through MapsBridge, Python analysis can identify regions of interest, initiate additional acquisitions based on current results, or connect to AutoScript when more advanced instrument control is needed. This combination offers a practical path from guided workflows to customized, closed-loop automation and, ultimately, more autonomous electron microscopy.

IONIC STRUCTURES AND BONDING AT CHARGED SOLID-LIQUID INTERFACES UNDER ELECTRIC FIELD

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Charged solid-liquid interface is central to confined fluidic transport such as in biological channels, and to electrochemical energy systems at which the electron-ion transfer occurs, yet their microscopic structure and bonding has remained elusive. Owing to the subtle interplay of electrostatic forces and thermal dissipation, complex ion arrangements form at interfaces known as the Electrical Double Layer (EDL). Despite the century-long evolution, current understanding of EDL is still primarily based on classic mean-field theory represented by the Gouy-Chapman-Stern (GCS) model, while direct experimental observations of ion arrangements at interfaces have been scarce. Here we utilize cryo-EM to visualize ion arrangements at operando-frozen charged solid-liquid interfaces at the angstrom level, in conjunction with vibrational EELS to probe hydration and bonding environments. We reveal how do ions, water and interphases reorganize under applied electric field.

MULTISCALE 4D-STEM DIAGNOSTICS OF DYNAMIC STRUCTURAL HETEROGENEITY IN BATTERY CATHODES

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Dynamic structural deformation in layered materials, including Ni-rich $LiNixMnyCo1-x-yO_2$ (NMC) and MoS_2, arises from the coupled evolution of interlayer displacement, lattice strain, and domain misorientation during phase transformation and interfacial interactions. Understanding how these structural heterogeneities evolve across multiple length scales is essential for revealing the mechanisms governing structural accommodation and degradation. Here, atomic-resolution and in situ STEM are integrated with multiscale 4D-STEM to correlate local structural motifs with strain fields, domain configurations, and diffraction signatures, providing a comprehensive view of dynamic structural evolution. In Ni-rich NMC cathodes recovered after long-term cycling, 4D-STEM maps the spatial distribution of interlayer-gliding domains and distinguishes deformation pathways in glide- and crack-dominant particles. Glide-dominant particles retain comparatively coherent domains and moderate lattice distortion, whereas crack-dominant particles exhibit localized strain, rotational discontinuities, and extensive domain fragmentation. Gliding-induced Bragg-peak splitting, supported by multislice simulations, further identifies long-period stacking modulation and directly links local interlayer displacement to structural degradation. At dynamic solid–liquid interfaces, in situ atomic-resolution STEM captures exfoliation, transient amorphization, nanodomain migration, and recrystallization in MoS_2 nanosheets within the same region. Complementary in situ 4D-STEM reveals localized strain, rippling, lattice rotation, and folded domain boundaries while preserving the parent layered structure, demonstrating the capability of multiscale electron microscopy to quantify transient structural heterogeneity under realistic environments. Together, these studies establish multiscale 4D-STEM as a versatile approach for bridging atomic-scale structural evolution with mesoscale deformation, providing new mechanistic insight into layered materials and a broadly applicable microscopy framework for investigating dynamic functional materials.

MULTIMODAL SCANNING PROBE MICROSCOPY REVEALS LOWER-STEP-SELECTIVE ZN CAPTURE ON PT(111) AND ITS UNDERLYING MECHANISM

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Pt–Zn bimetallic catalysts have attracted considerable attention for applications in the propylene production and the electrochemical methanol oxidation reaction. Designing an efficient Pt-Zn catalyst often involves depositing Zn on a Pt catalyst to change the electronic structure of Pt active sites. These functions depend critically on the atomic distribution of Zn, yet how Zn atoms are initially captured on Pt surfaces remain poorly understood. Over the past two decades, scanning probe microscopy (SPM) has pushed the resolution limit to atomic scale. Combined with built-in spectroscopy, it has been used to understand intermolecular interactions, charge distributions, and electronic orbital signatures, making it an ideal platform to study the initial growth during bimetallic deposition. Here, we combine atom-resolved SPM, atomic force spectroscopy (AFS), and first principle calculations to investigate the early stage of Zn deposition on stepped Pt(111). We observe that, at low coverage of 0.02 ML, the Zn adatoms preferentially decorate the Pt(111) steps. Moreover, using both STM and AFM with Pt and O-functionalized tips to image the edge absorbates, we are able to distinguish whether Zn stays at lower or upper step edge. Statistical analysis of the Zn distribution at different absorption sites shows an order-of-magnitude preference for the lower step edge over the upper step edge and terrace, showing a step-flow-like growth behavior in the Zn/Pt system. The absorption preference Of Zn at Pt(111) lower step edge can be explained by a lower adsorption energy than other equilibrium absorption sites given by DFT calculations. Furthermore, to understand what kind of interaction contributes to the lower-step-edge stabilization, we performed LCPD measurements on Pt(111) steps and calculated the adsorption energy, charge transfer and orbital hybridization along various approaching trajectories, and we find that the stabilization mainly comes from enhanced Zn 4s/4p orbital coupling with neighboring Pt rather than electrostatic interaction between Zn and Smoluchowski dipole moments along the step. This study reveals an atomic-scale mechanism for the initial capture of Zn atoms at Pt surface defects, providing insight into the earliest stages of Zn/Pt bimetallic catalyst formation.

RESOLVING THE DYNAMIC STRUCTURAL PATHWAY OF FE-CATALYZED GRAPHITIZATION OF BIOCHAR VIA HIGH TEMPERATURE IN SITU ELECTRON MICROSCOPY, X-RAY DIFFRACTION AND SYNCHROTRON TOTAL SCATTERING

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The conversion of renewable carbon resources into graphene-like structures has gained momentum in engineering and manufacturing of advanced carbon materials for energy storage. Yet, the structural pathways involved in metal-catalyzed graphitization remain difficult to unlock since they occur dynamically, over multiple length scales, and at elevated temperatures. Here, we investigate the Fe-catalyzed transformation of cellulose, the most abundant biomass polymer, using a multi-scale high-temperature technique, with an emphasis on directly resolving the evolution of carbon nanostructure. In situ transmission electron microscopy (TEM) up to ~ 1200 °C enables real-space observation of the carbon structural evolution during heating, revealing the progressive reorganization of initially disordered carbon into increasingly ordered graphitic domains. It shows that the mobile Fe nanoparticles serve as nucleation sites, initially surrounded by carbon rings that further grow into tail-like graphitic layers and eventually detach from the nanoparticles. These nanoscale observations are complemented by in situ X-ray diffraction up to 1500 °C, which tracks the evolution of crystalline ordering and carbon structure at the crystallite scale. In situ Raman spectroscopy carried out at 1400 °C enables probing the defects in the carbon structures. In situ synchrotron X-ray total-scattering and pair distribution function (PDF) analysis at temperatures above 1400 °C further reveal changes in short- and medium-range atomic correlations during graphitization. By correlating real-space electron microscopy with reciprocal-space diffraction and atomic pair correlations, this multimodal dynamic approach provides multiscale insights of the transition from disordered biomass toward graphitic structures catalyzed by Fe. These results highlight how complementary high-temperature in situ imaging and scattering methods can reveal dynamic structural transformations that are inaccessible from conventional ex situ characterization alone.

EXPLORING THE COMPLEXITY OF TRIC'S INGENIOUS ROUTE TO RING CLOSURE

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Eukaryotic chaperonin TRiC folds ~10% of the cytosolic proteome through an ATP-driven cycle in which the ring closes to form a protected chamber. How this closure is orchestrated across TRiC's eight distinct subunits has remained unclear. Here, combining high-resolution cryo-EM with deep-learning–enabled analysis of conformational heterogeneity, we reconstruct a near-atomic, time-ordered structural trajectory of human TRiC ring closure triggered by ATP binding. We identify an ingenious, multi-step mechanism in which closure proceeds via stepwise extension of an apical helical interaction network around the ring, coordinated with sequential rotations of the equatorial domains. Because each subunit exhibits a distinct ATP response, the global motion is asymmetric, ordered rather than concerted. Together, these results map the conformational landscape underlying ATP-binding–induced (rather than hydrolysis-driven) closure and provide a mechanistic framework connecting allosteric propagation, chamber formation, and substrate handling.

DIFFUSION MODEL 3D RECONSTRUCTION WITH EMBEDDED PHYSICAL PRIORS FOR SPARSE-VIEW ELECTRON TOMOGRAPHY

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Electron tomography (ET) serves as a pivotal technique for deciphering the 3D microstructure of material and biological specimens. Traditional workflows require 60 to 120 projection views, leading to prolonged imaging cycles and elevated risk of radiation damage to samples. Existing sparse-view reconstruction methods based on convolutional neural networks tend to produce structural artifacts and detail loss under extremely sparse viewing conditions, and the credibility of their reconstructed structures remains widely debated in the community. In this work, we present a physically constrained diffusion model framework for 3D ET reconstruction. We incorporate electron projection geometry, contrast transfer function (CTF) and Poisson noise model into both the diffusion sampling process and the loss function, establishing a dual-driven paradigm of data learning and physical priors to fundamentally ensure the authenticity of reconstructed structures. For in-situ imaging and radiation-sensitive samples, our method delivers high-precision reconstruction with merely 10–20 extremely sparse views. Furthermore, we adopt a hybrid scheme of 2D projection-domain diffusion denoising combined with 3D iterative back-projection, which markedly boosts reconstruction efficiency. Extensive validation on public material science and cryo-electron tomography datasets demonstrates that our method surpasses state-of-the-art approaches in reconstruction resolution, structural similarity and artifact suppression under extremely sparse views. It accommodates diverse imaging requirements across physical and life sciences, and offers a novel technical route for developing next-generation low-damage, high-throughput electron tomography tools.

Speaker Abstracts

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Poster Abstracts

POSTER SESSION: Tuesday, October 20, 5:00 – 6:30 p.m. Princeton Friend Center

1. Posterboards will be numbered, and thumbtacks will be provided.
2. You can begin hanging up your poster on Tuesday, October 20 beginning at 3:00 p.m. Please do not bring your poster on Monday, as we will not be able to store it.
3. Please ensure that your poster dimensions are **30"W x 40"H**.
4. **Poster numbers are subject to change.** Updated lists will be sent out via the Whova app and the final list will be at the Registration Desk for you to double check.
5. You must take your poster down after the Poster Session concludes. Posters not taken will be discarded.

Assignments below are in ALPHA order by Last Name.

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Cheng	Guangming	4
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