

Studying structural dynamics in biology at SLAC

Sandra Mous

LCLS Biological Sciences department and
the Center for Structural Dynamics in Biology

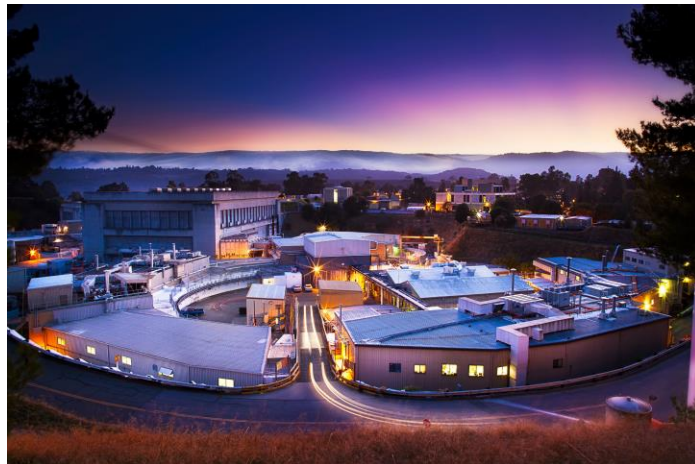
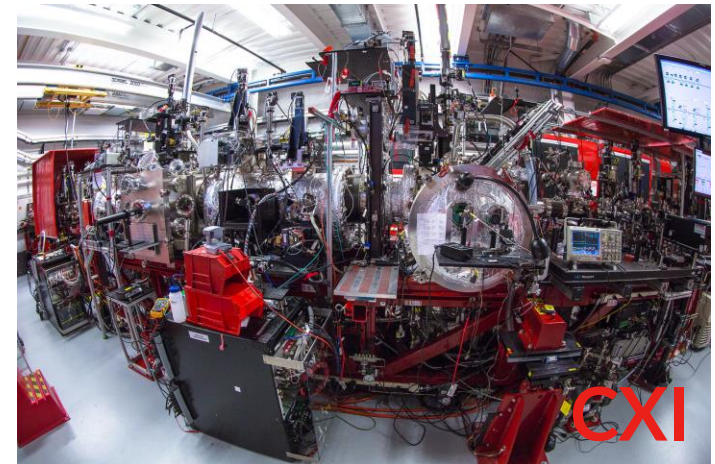
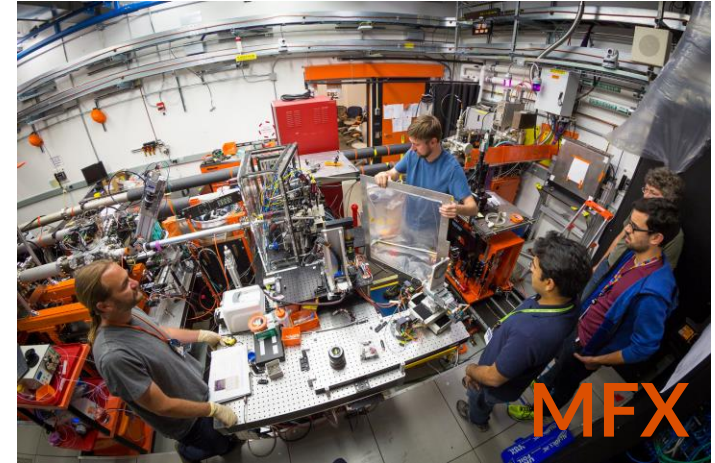
LEAPS meets Life Sciences conference - May 18th, 2023



X-ray science at SLAC National Accelerator Laboratory



Linac Coherent
Light Source (LCLS)



Stanford Synchrotron
Radiation Lightsource (SSRL)

X-ray science at SLAC National Accelerator Laboratory

Sample Prep Lab (Far Experimental Hall)



Arrillaga Science Center Biolabs

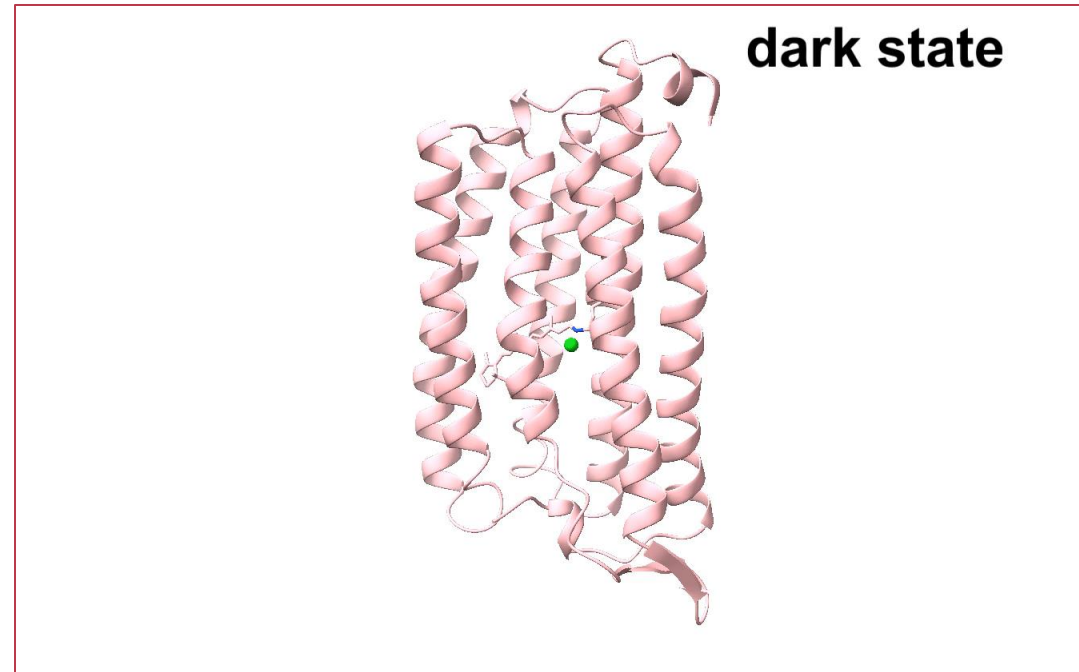
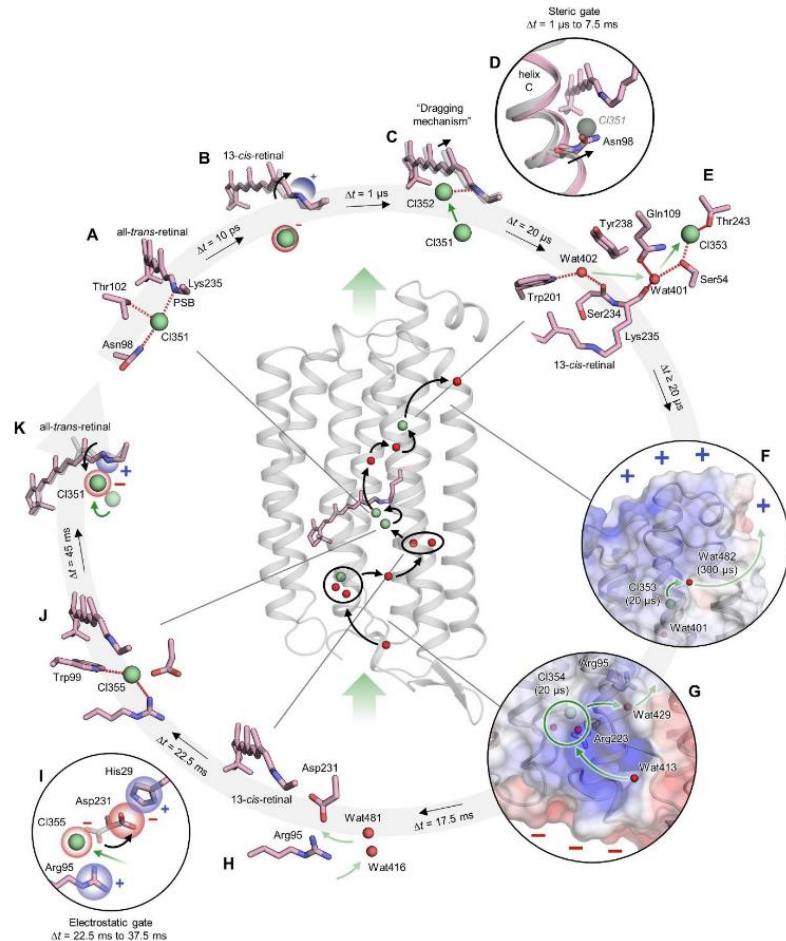


Injector Characterization Lab (with FEL simulator)



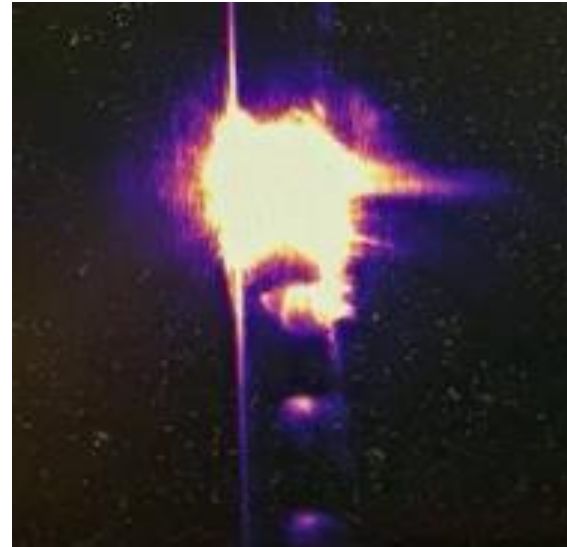
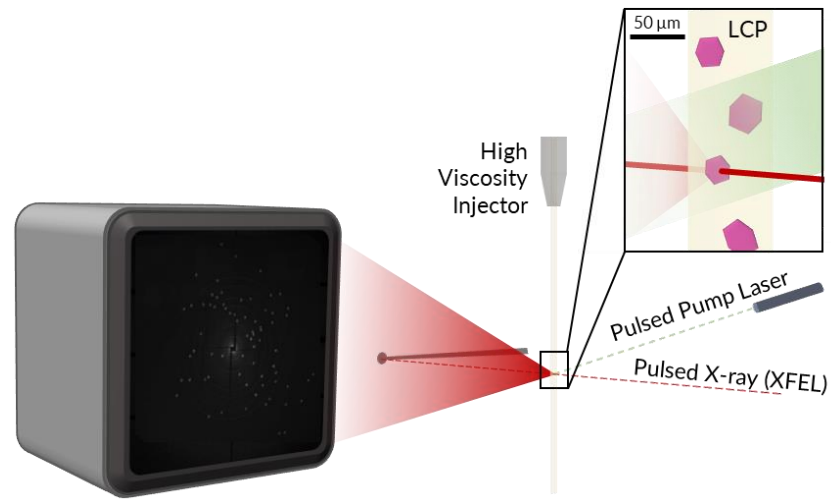
Serial crystallography is very rewarding

- X-ray free electron lasers such as LCLS have enabled a structural and temporal understanding of biology

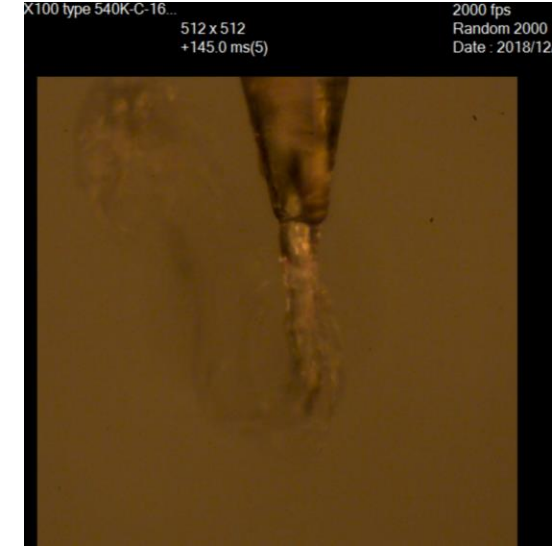


Mous *et al.* (2022) *Science* 375:6583.
Nogly group - ETHZ

Serial crystallography is very challenging



European XFEL, exp. 2480, July 2019



Paul Scherrer Institute, December 2018

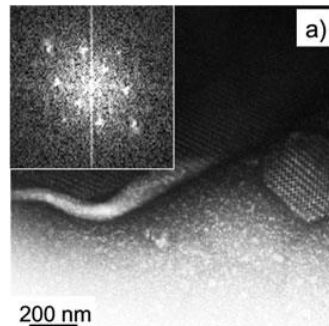
- Unstable jetting
- Clogged injector
- Crystal size heterogeneity
- Low crystal density and hit rate
- Low resolution
- Low indexing rate
- Anisotropy
- Inactive protein

The many components of a serial crystallography experiment

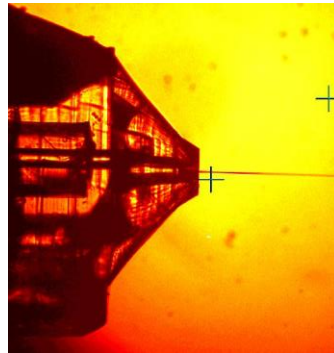
Sample preparation



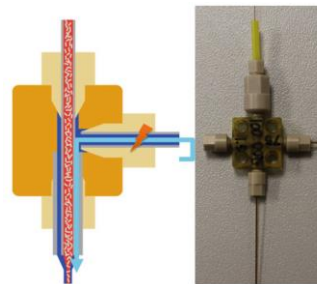
Sample characterization



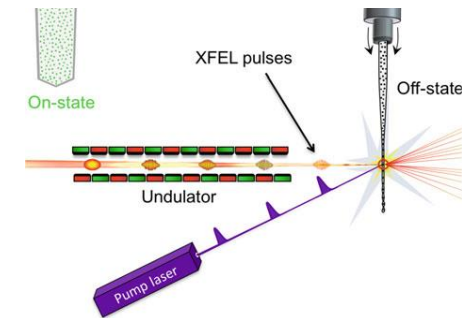
Sample delivery



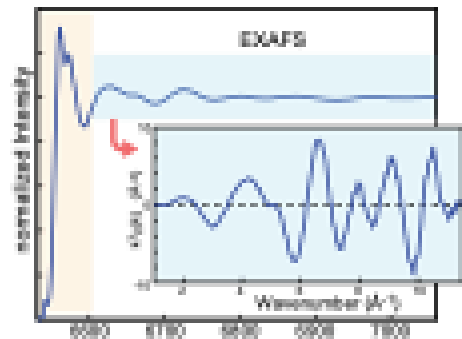
Sample triggering



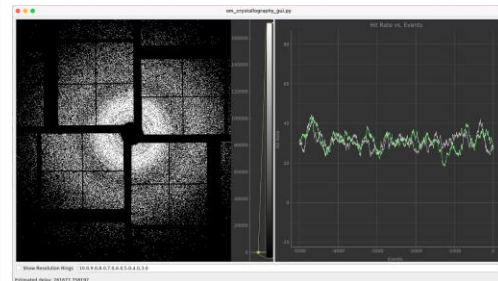
Diffraction data collection



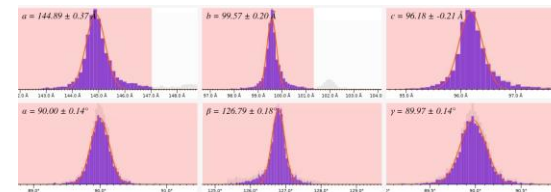
Spectroscopies



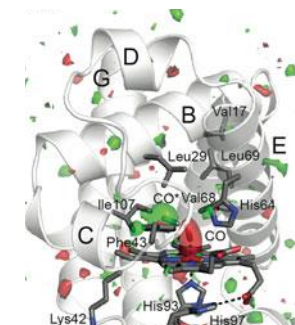
Online feedback



Data analysis



Interpretation





The LCLS Biological Sciences department partnered with the NIH to create the Center for Structural Dynamics in Biology

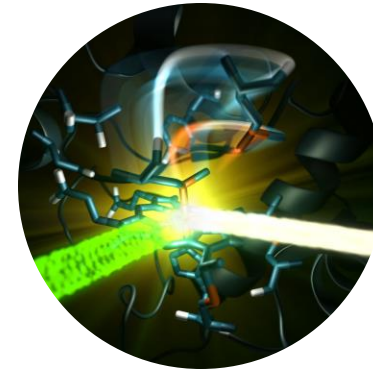
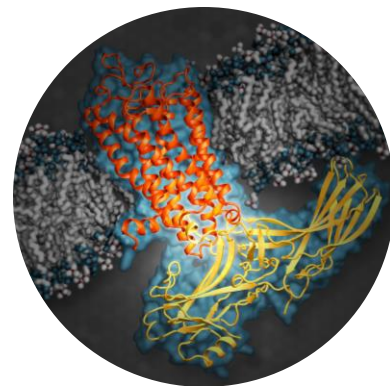
Driving Biomedical Projects

GPCR activation

Vadim Cherezov (USC)
Brian Kobilka (Stanford)

Riboswitch dynamics

Yun-Xing Wang (NCI)



Metalloenzymes

Jan Kern (LBL)
Denis Rousseau (Einstein M)

RNA transcription

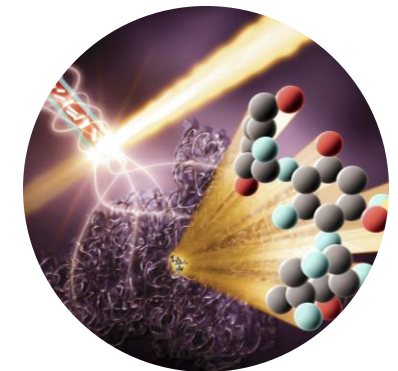
Guillermo Calero (U Pitt)

Antibiotic resistance

Edward Yu (CWRU)
Marius Schmidt (UWM)

Thermal dynamics

James Fraser (UCSF)
Michael Thompson (UCM)





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Technology Research & Development

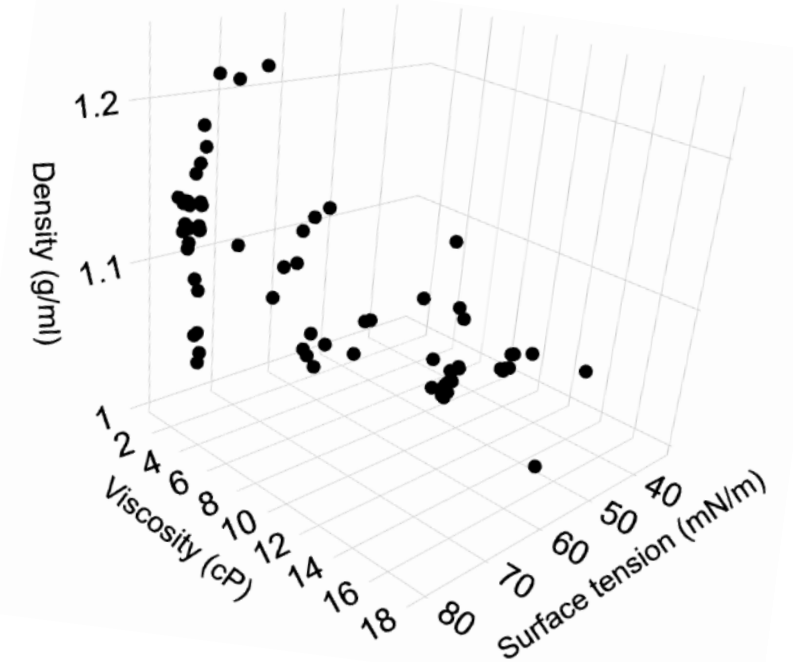
- Making the serial crystallography experiment easier, allowing the broad biomedical research community to study dynamics of macromolecules
 - Improve our understanding of samples and sample delivery systems
 - Ease access to LCLS
 - Provide automation to increase the efficiency
- Pushing the boundaries by expanding our capabilities
 - Support the science case of our Driving Biomedical Projects
 - Develop new technologies in our labs and at the beamline

Understanding crystalline sample delivery

- Physicochemical characterization of common injection fluids and crystalline samples
 - Density
 - Rheology (viscosity)
 - Surface tension
- High-speed jet testing station
 - Wide range of injectors and sample environments supported

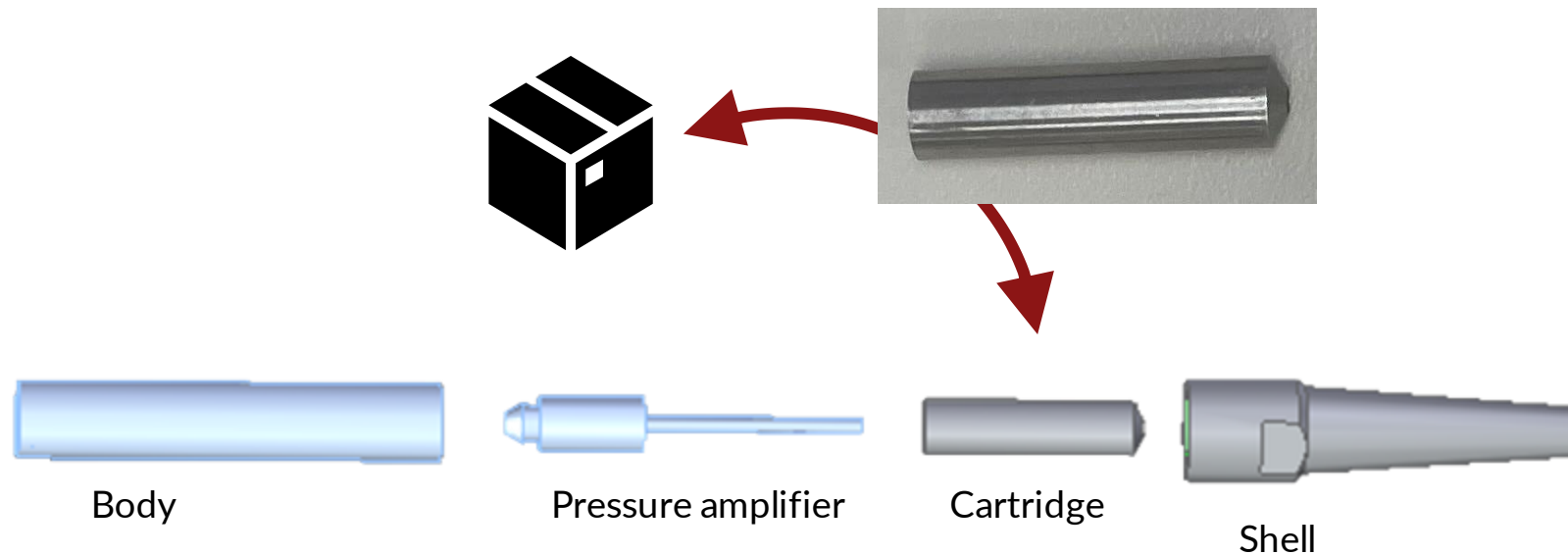


Hampton Research Index
crystallization screen



Providing remote access to LCLS

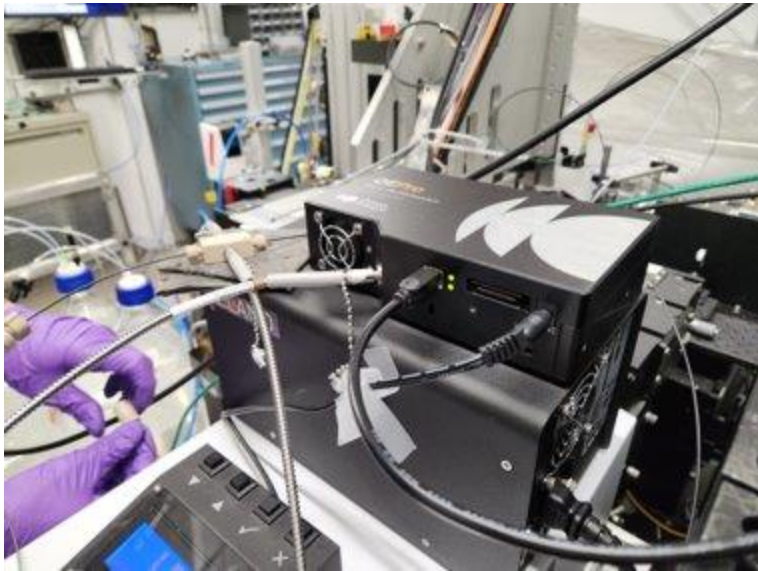
- Cartridges allow mail-in of protein crystals in viscous media
- Taking the first step to remote-access protein crystal screening



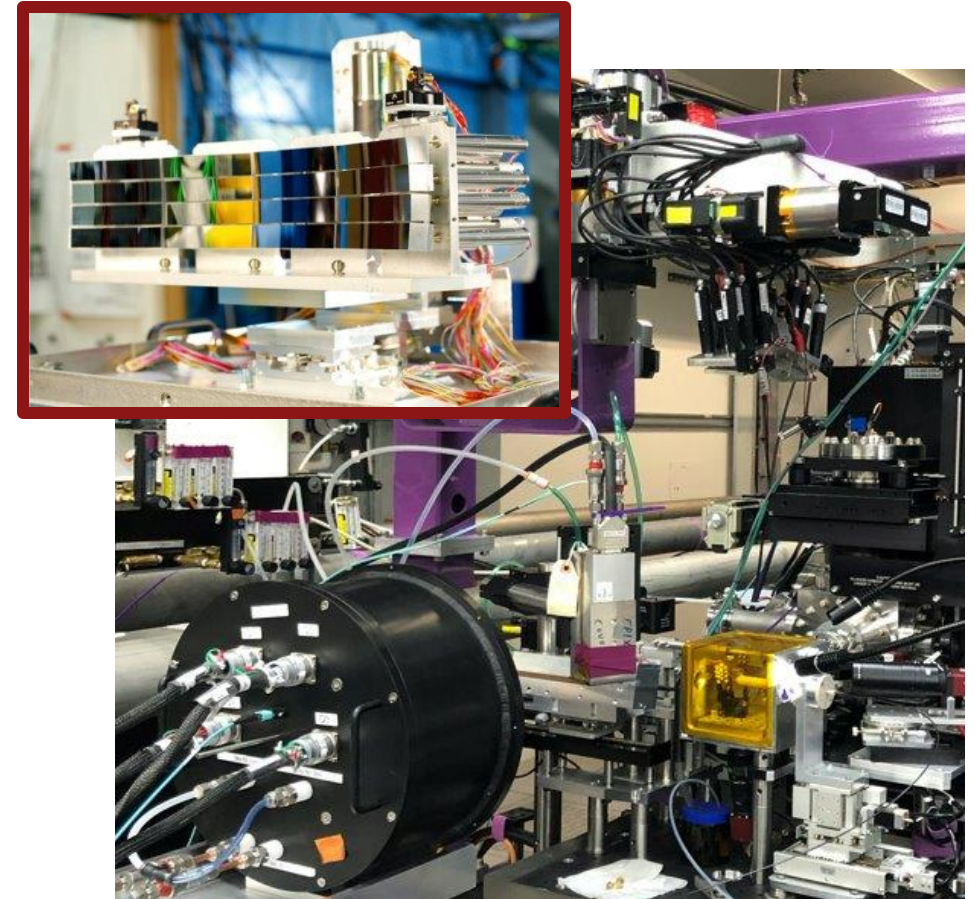
- Currently investigating other mail-in opportunities using different sample delivery systems

On- and offline spectroscopic characterization

- UV-Vis and X-ray emission spectrometer at LCLS XCS/MFX and SSRL BL15-2 for online monitoring
- UV-Vis and Raman spectroscopy in the lab for offline sample characterization



Microspectrometer setup at SSRL BL15-2 for monitoring sample integrity



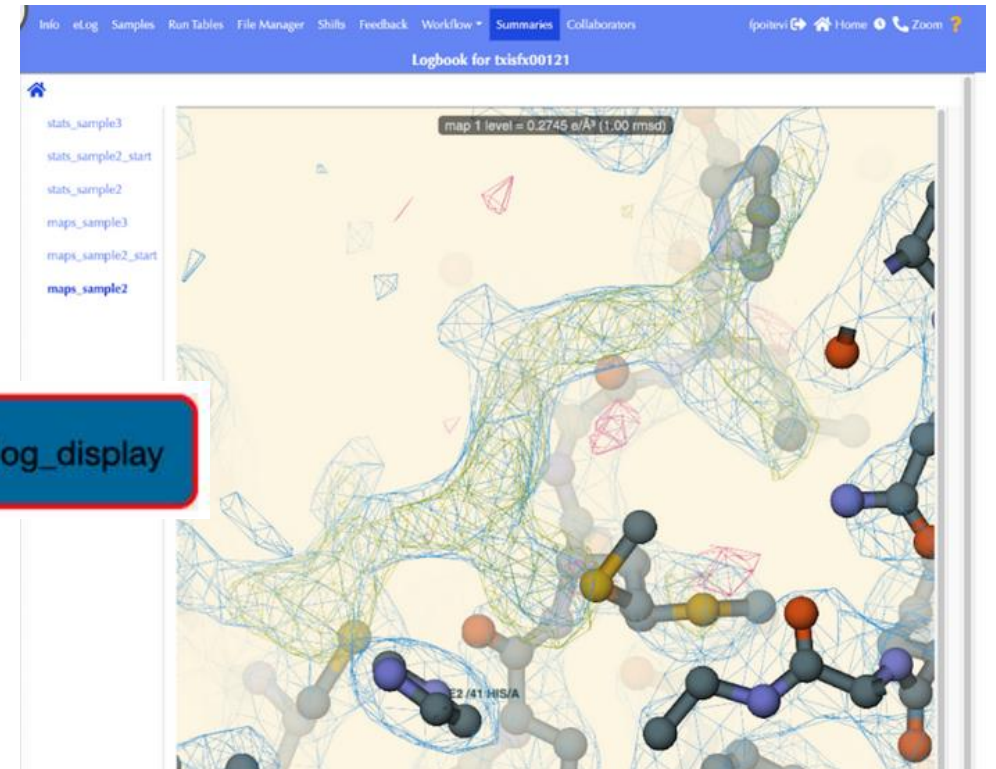
X-ray diffraction + X-ray emission spectroscopy setup at XCS

Increasing beamtime efficiency through automation

- Simplified and automated workflow for LCLS data analysis will reduce complexity
 - Processing output and statistics visualized in eLog during the experiment to inform users during data collection

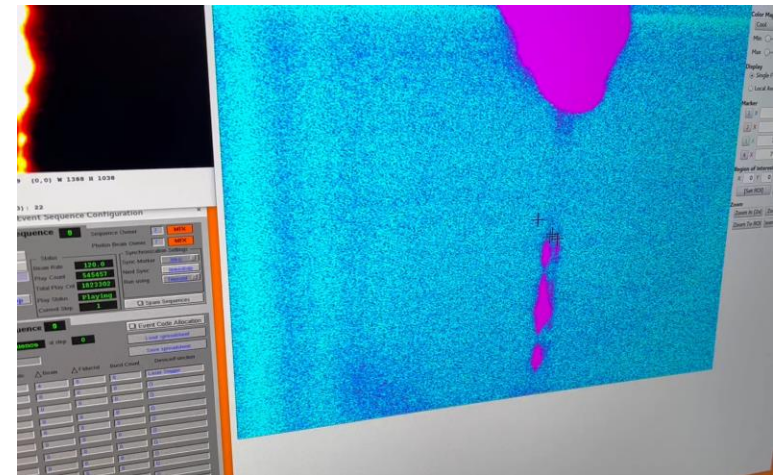
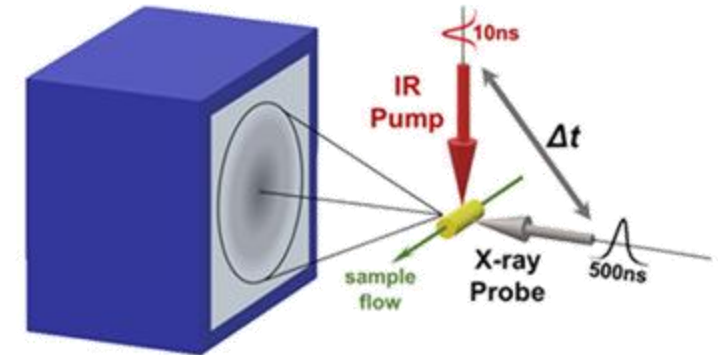


- Hardware automation will reduce repetitive, time-consuming tasks while improving reliability
 - Jet tracking (injector position and adjusting flow rate)
 - Automated sample exchange
 - Equipment protection



Expanding the range of bioscience experiments possible at LCLS

- Estimated that only ~0.5% of proteins are naturally photoactive
- Successful T-jump experiments at MFX thanks to a new infrared pump laser and IR imaging
- Ultrafast far-IR and THz laser system for MFX under development
- Mixing characterization and further development of mixing injectors (e.g., DFFN and coMESH)



Driving Biomedical Project: M. Thompson, UC Merced

Reaching out to the life sciences research community

- Outreach opportunities
 - New Bio@LCLS website
 - Conferences and workshops (hands-on SFX tutorials in September 2023)
 - Internships and student projects
- Become a partner of the Center for Structural Dynamics in Biology
- Please let us know what your needs are. How can we do better?
 - Expanded capabilities for on-site characterization
 - Mail-in program for SFX sample screening
 - Post-beamtime data analysis support
 - Fast feedback for SAXS/WAXS experiments
 - What should biology at LCLS-II-HE look like?



Explore Biology at LCLS online!

A screenshot of the 'Short Proposal Form Biology' web interface. The header features the SLAC and LCLS logos on the left and the title 'Short Proposal Form Biology' on the right. The main content area includes a 'Proposal information' section with a 'Proposal type' dropdown menu and a link to 'https://cls.slac.stanford.edu/proposals/modes-of-access'. Below this is a 'Descriptive experiment title' text input field with a character count note: 'This title will be made public if you are awarded beamtime. Maximum of 150 characters.' The next section asks 'Is this experiment related to one or more LCLS proposals that have been previously submitted or accepted?' with a dropdown menu and a note: '→ If previously accepted, list experiment number(s):'. The 'Spokesperson' field is partially visible at the bottom.

Thank you

Please reach out to us!

Center for Structural Dynamics in Biology

Roberto Alonso Mori

Sebastien Boutet

Sergio Carbajo (UCLA)

Aina Cohen

Gabriel Dorlhiac

Christopher Robin Frank

Leland Gee

Brandon Hayes

Mark Hunter

Maithri Kashipathy

Christopher Kupitz

Mason Landrum

Stella Lisova

Darya Marchany-Rivera

Sandra Mous

Victoria Nguyen

Jose Ortiz

Raymond Sierra

& LCLS Biological Sciences team and affiliates

Andy Aquila

Alex Batyuk

Sebastian Dehe

Dan DePonte

Tim van Driel

Christina Hampton

Valerio Mariani

Ariana Peck

Fred Poitevin

Omar Quijano

Ryan Ribson

Bob Sublett

