

Life Sciences at the APS

April 14, 2014

Bob Fischetti

XSD, Associate Division Director

GM/CA, Group Leader



Life Sciences Grand Challenge

- Relating structure to function
- How does the structure of protein, DNA, RNA, complexes ... impart function
- What role does structure play in the hierarchy of organelles, cells, organs and species
 - How do integral membrane proteins perform their very diverse, biologically important functions
 - How complex molecular machines embedded in cell membranes work
 - How drugs interact with their targets
 - Improve specificity
 - Reduce side effects



Tool of Life Scientists

- Crystallography
 - High resolution probe of molecular structure
 - Single molecular domains to large macromolecular complexes
 - Requires crystals!
- SAXS/WAXS
 - Protein shape or folding
 - Protein-protein interactions
 - Drug/ligand binding
 - Solution or solid state
- X-ray fluorescence microscopy
 - Probe the role of metallo-protein function in organisms
 - Study factors such as the local chemical / trace elements
- X-ray (micro)-diffraction
 - Visualize local reciprocal space
 - Plays an important role in studies relating to biomineralization



Life Sciences Today at the APS - BTRs for FY2013

Over 40% of users self declare as Biological or Life Sciences

Beamline	Total BTRs for Biological and Life Sciences	Total BTRs All Sciences	% Biological and Life Sciences	MX	X-ray Absorption and Emission Spectroscopies	SAXs, WAXS, USAXS and GSAXS	Diffraction Other	Scattering (inelastic, nuclear resonant, magnetic)	Tomography	Time Resolved Scattering, Crystallography and XAS	Imaging and Microscopy	Fiber diffraction	Laue Crystallography	Other
1-ID-B,C,E	17	219	7.8		1	4	10		1					1
2-BM-A,B	28	87	32.2						20		7			1
2-ID-B	9	30	30.0		4		3				1			1
2-ID-D	63	169	37.3		46	3	11				3			
2-ID-E	64	81	79.0		54		2		5		1			2
3-ID-B,C,D	29	154	18.8				3	26						
4-ID-C	12	190	6.3		5									7
7-ID-B,C,D	9	140	6.4				6			3				
8-BM-B	20	32	62.5		19	1								
9-ID-B,C	20	110	18.2		1		11	6						2
12-ID-B	37	135	27.4			34	2			1				
13-ID-E	10	114	8.8		9				1					
14-BM-C	19	26	73.1	10		2				1		5		1
14-ID-B	67	86	77.9	9	4	15		2		25		1	10	1
15-ID-B,C,D	30	261	11.5			5	15	9						1
17-ID-B	28	30	93.3	28										
18-ID-D	97	118	82.2	5	3	61				5	1	21		1
19-BM-D	16	16	100.0	11			4							1
19-ID-D	139	139	100.0	127		3	7				1			1
20-BM-B	11	149	7.4		11									
20-ID-B,C	31	148	20.9		30			1						
21-ID-D	53	62	85.5	41	6		1	1			3			1
21-ID-F	51	53	96.2	50			1							
21-ID-G	58	59	98.3	57						1				
22-BM-D	32	158	20.3	32										
22-ID-D	94	251	37.5	94										
23-ID-B	162	162	100.0	154	2	1	4							1
23-ID-D	179	180	99.4	176	1		2							
24-ID-C	181	181	100.0	173			8							
24-ID-E	126	131	96.2	125			1							
31-ID-D	28	32	87.5	28										
32-ID-B,C	25	99	25.3	2					8		15			
34-ID-C	17	90	18.9	2			15							



Diversity of LS CAT Funding

Bio CARS

NIH (NIGMS)
(APS for non-biological TR studies)

BioCAT

NIH (NIGMS)

SBC

DOE, BER

GM/CA-CAT

NIH (NIGMS, NCI)

LS-CAT

University of Michigan
Michigan State University
Van Andel Institute
Wayne State University
Vanderbilt University
University of Illinois
University of Wisconsin/Madison
Northwestern University
GlaxoSmithKline
Emerald Biostructures
Cayman Chemical
Grand Valley State University
Rice University
Medical College of Wisconsin

IMCA-CAT

Abbot
Bristol-Myers Squibb
Johnson & Johnson (Subscriber)
Merck Research Laboratories
Novartis
Pfizer
HWI

LRL-CAT

Eli Lilly & Company

NE-CAT

NIH (NIGMS)
Columbia University
Cornell University
Harvard University
Memorial Sloan-Kettering Cancer Center
Massachusetts Institute of Technology
Rockefeller University
Yale University

SER-CAT

Duke University
Emory University
Florida State University
Georgia State University
Georgia Tech Research Corporation
Medical University of South Carolina
Monsanto Company
NIH Intramural Research Program
North Carolina State University
Rosalind Franklin U. of Med. and Sci.
Scripps Florida
St. Jude Children's Research Hospital
University of Alabama at Birmingham
University of South Florida
University of Georgia
University of Kentucky
University of Missouri at Kansas City
University of North Carolina at Chapel Hill
University of Pittsburgh
University of South Carolina
University of Virginia
Vanderbilt University

Aggregate MX CAT Investment

- Cap. Eq. since 2010 \$ 23M
- Total capital equipment \$ 144M
- Annual operating costs \$ 19M

DOE-BES does not fund MX at APS



Life Sciences Council - Formed in 2008

Provides a forum for discussion of common issues related to life sciences

The LSC is a subcommittee of the Partner User Council

Meet quarterly to discuss current issues and develop long range strategic plans

Partner User Council representative from life sciences CATS

Denis Keane, DND-CAT

Keith Moffat, Vukica Srajer, BioCARs

Lisa Keefe, IMCA-CAT

Tom Irving, Bio-CAT

Andrzej Joachimiak, SBC-CAT

John Chrzas, Gerd Rosenbaum, Bi-Cheng Wang, SER-CAT

Wayne Anderson, **Keith Brister (Chair)**, LS-CAT

Bob Fischetti, Janet Smith, GM/CA-CAT

Malcolm Capel, Steve Ealick, NE-CAT

Steve Wasserman, SGX-CAT

Other Community Members

Chris Jacobsen

Stefan Vogt

Lydia Finney

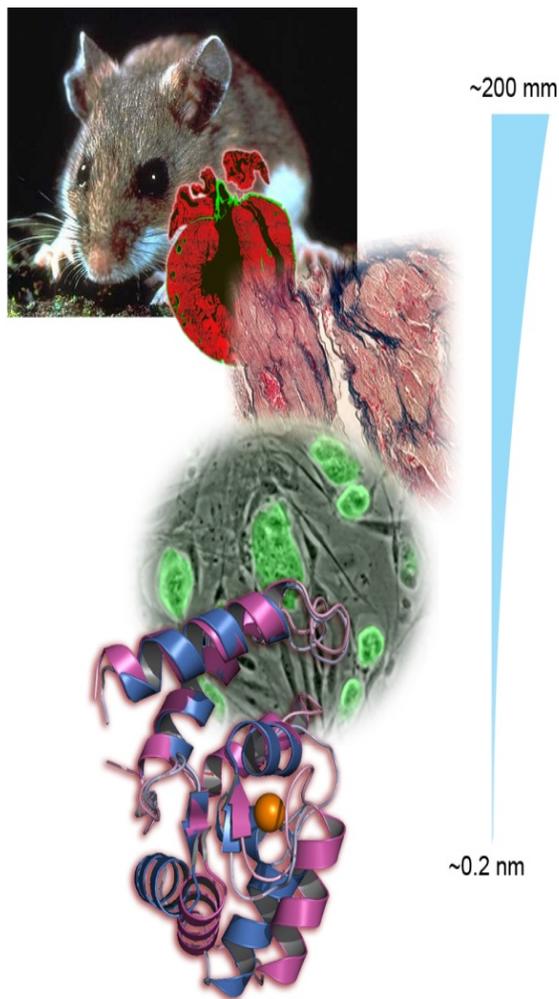
Gayle Woloschak

Randy Winans

Xiaobing Zuo



Proteins to Organisms



Grand Challenge

- identify how macromolecules determine organism function
- understand how function is influenced by trace metals and other factors

APS-U, and the high-energy, high-brilliance x-rays at APS will critically extend our ability to

- probe proteins and protein dynamics
- directly (and indirectly) image non-periodic, hierarchical soft structures from the mm to the 10 nm scale.

Support NIH mission

- Biomedical research

Support DOE/BER mission

- biofuels / environmental stewardship / new tools to probe interface of biology and physics

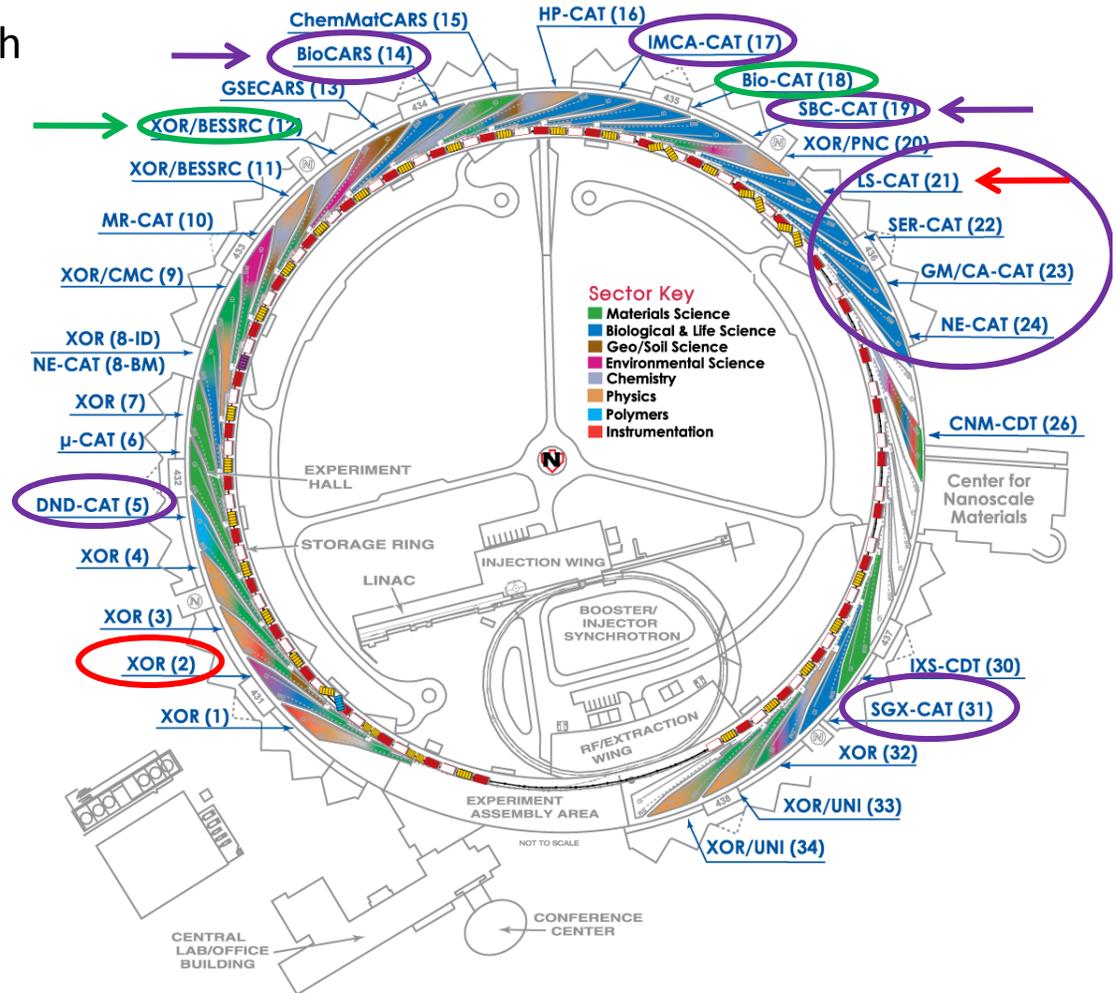
Proteins to Organisms Beamlines at the APS

Many APS beamlines carry out research in the area of Proteins to Organisms

Macromolecular Crystallography
 14-ID, 17-ID, 19-ID, 21-ID, 22-ID, 23-ID, 24-ID, 31-ID

SAXS/WAXS/Fiber diffraction
 5-ID, 12-ID, 18-ID

Microprobes
 2-ID, 18-ID, 21-ID-D



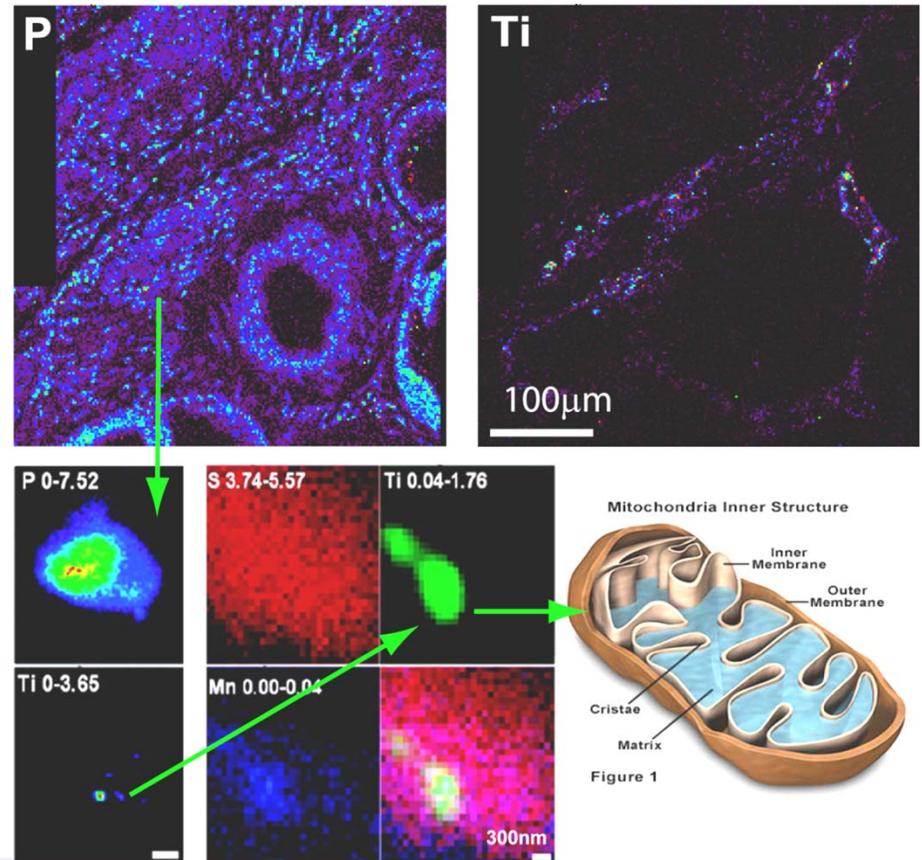
Overview of Proteins to Organisms Beamlines

- **BioNanoprobe upgrade (21-ID):**
 - Upgrade beamline and endstation for Nanoprobe optimised for trace element mapping of frozen-hydrated biological and biomedical samples down to the 10 nm spatial resolution
- **Enhanced SAXS/WAXS:**
 - Dedicated sample environment for macromolecular solution scattering including FPLC, Flow cell, and UV-Vis.
 - Optimised in-vacuum chamber to simultaneously record SAXS and WAXS, and easily change WAXS q-range
- **Microfocus MX Beamline (19-ID, 23-ID):**
 - Upgrade 19-ID to provide multiple 1 micron line focused beams
 - Upgrade 23-ID to provide a 1 micron circular beam
 - To study microcrystals and minimize radiation damage.
- **Enhanced Time-Resolved MX Beamline (14-ID):**
 - Develop new analog-integrating Pixel Array Detector matched to fast data acquisition
 - Improve beamline with fast mechanical shutter, and x-ray focusing (CRL) .
- **Long Wavelength National Resource**
 - B.C. Wang submitted a LOI to the APS
 - SAD phasing



BioNanoprobe upgrade (21-ID)

- Understand the fundamental, intrinsic role trace elements play in biological systems, specifically as integral components with regulatory or catalytic functions
 - eg, role of Zn in regulation of Meiosis in Mammalian Oocytes (Kim *et al*, Nature Chem Biol 2010)
- Understand and address the links between metals and disease:
 - endogenous dysregulation (eg, Alzheimer's; Wilson's disease – Ralle *et al*, J Chem Biol 2010)
 - exogenous uptake (eg, As contaminated wells, Se deficiency).
- Develop **functional** nanocomposites for nanomedicine. Combine
 - targeting (e.g., cancer cells)
 - diagnostics (e.g. Gd as contrast agent for MRI)
 - therapy (e.g., TiO₂ with photo-induced cleavage of DNA)
 - targeting (e.g., sequence specific DNA)
- Questions
 - Do the nanocomposites enter the cells ?
 - Do they 'find' the right target ?
 - Do they ONLY interact with the right target (-> toxicity) ?
 - Do different components remain joined ?
 - What are their intracellular distributions

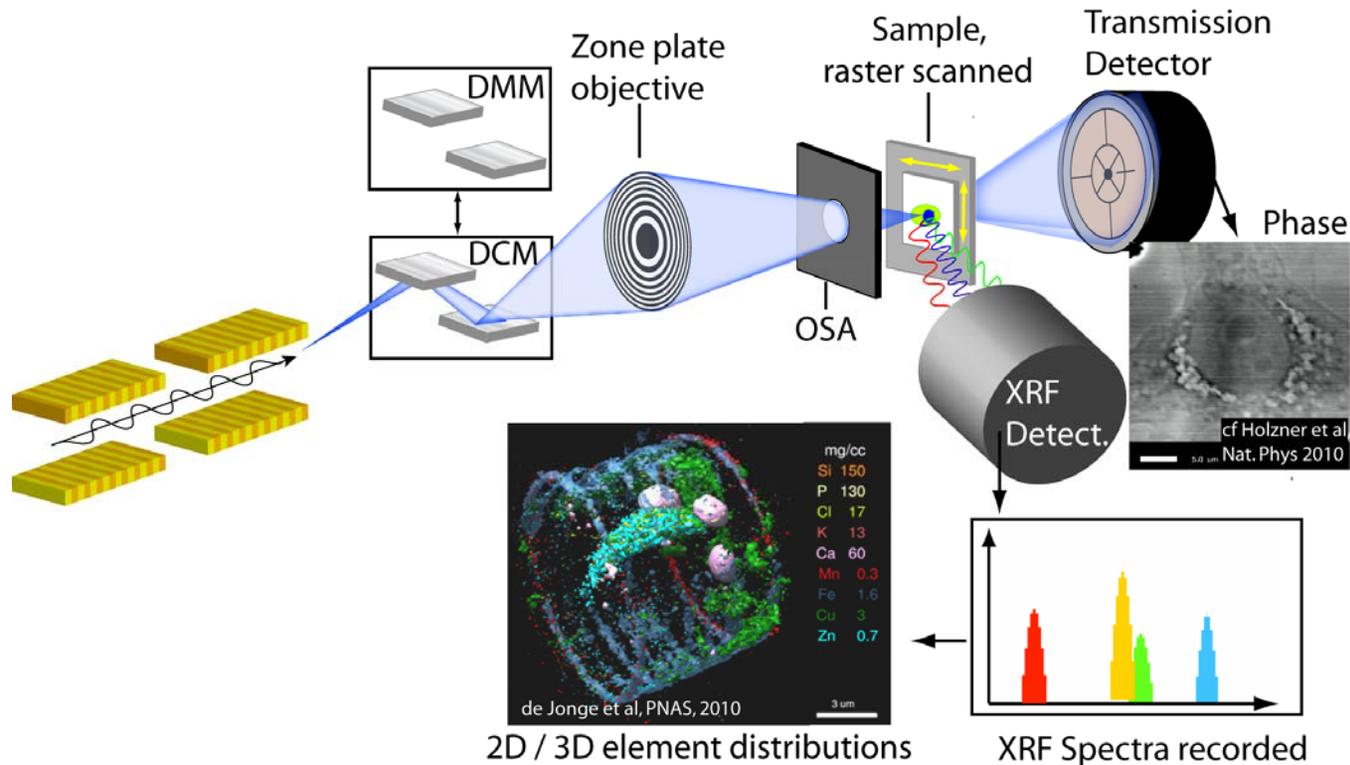


Bionanoprobe @ APS: status and plans

- First-of-its-kind cryo-transfer X-ray Fluorescence Microscope (XFM), studying role of nanoparticles, ions, metalloproteins in cells.
 - 2-3x higher spatial resolution (<50 nm), best biological fidelity (frozen hydrated) compared to previous APS XFMs.
 - Publications now appearing: *ACS Nano*, *J. Synch. Rad.*
 - With MBA upgrade and optimized beamline, can image 1000x more samples for better statistics, or dramatically increase resolution and sensitivity. **Game changer!**
 - Instrument purchased via S10 RR029272 (Woloschak *et al.*), operated at LS-CAT (21-ID-D; Brister *et al.*) with APS assistance (Vogt, Chen, Lai *et al.*). Cryo sample prep and ptychography supported via R01 GM104530 (Jacobsen & Woloschak).
 - Enthusiastic NIH-supported user community growing rapidly. NIH-supported workshop at Northwestern in August 2013 (R13 EB017630) drew 70+.
- Planning for P41 proposal submission (Woloschak and Jacobsen, plus Brister, O'Halloran, Vogt, Jin, Chen...):
 - Technical developments: fluorescence nanotomography, cryo prep including FIB, improved sensitivity and quantitation, improved resolution using new generation optics.
 - Operations support for NIH-supported researchers.



BioNanoprobe (21-ID) - future need



- Dedicated beamline (currently only 25% of 12-ID)
- Energy range: 5 – 30 keV, but optimized for >10 keV
- Two interchangeable monochromators (crystals) to trade flux vs res
- Resolution: $\delta \leq 20$ nm
- Sensitivity: <10 atoms in thin samples
- In vacuum, cryo-system
- MBA-lattice: higher sensitivity and throughput



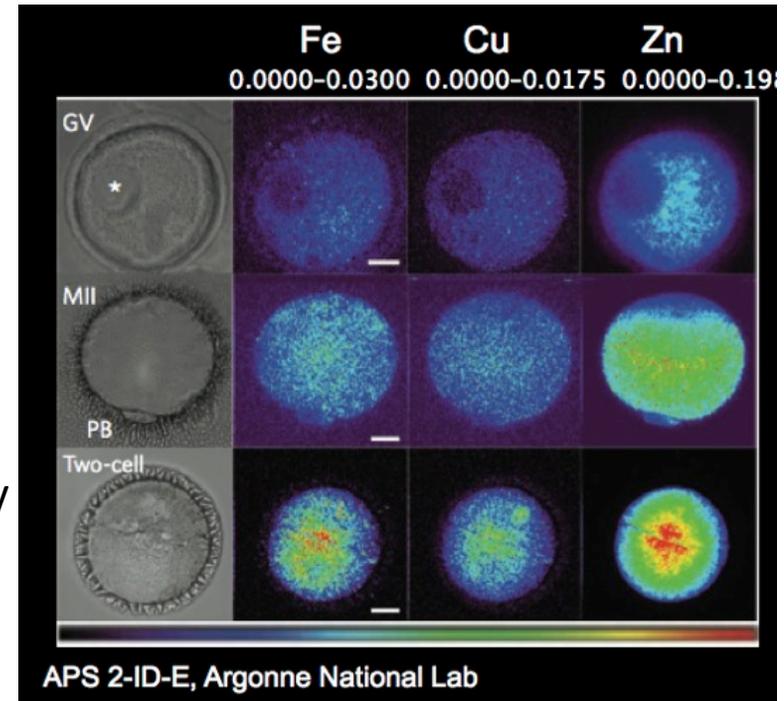
Tracking metals' influence in disease, infertility

Opportunity

- View metals key to cancer treatment
- Track metal interactions in individual cells (proteins, membranes, vesicles, organelles...)
- Study statistically significant populations

Gains from APS MBA Lattice

- Improve spatial resolution of metals at ~ 20 keV and higher
- Improve speed of data collection
 - 3D imaging a single cell now requires 36 hours, could be reduced to 15 minutes
- Improve sensitivity



Elemental images of immature oocyte (GV), a mature egg (MII), and a two-cell embryo.

Now: X-ray microscopy with $\sim 100 \times 100$ nm spatial resolution and 1,000s of atoms sensitivity

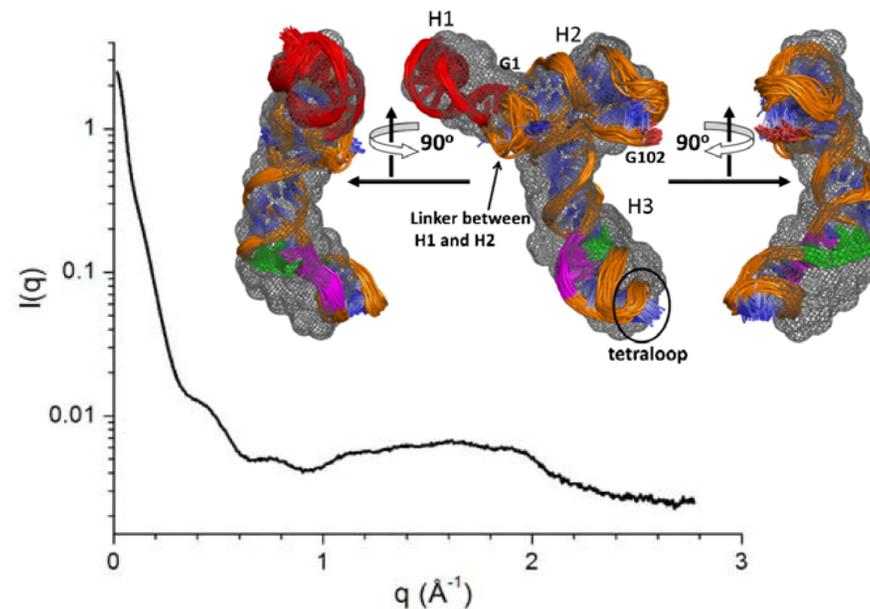
APS MBA upgrade: $\sim 5 \times 5$ nm resolution, < 5 atoms sensitivity: **factor of > 200 improvement**



Enhanced SAXS/WAXS

Why SAXS/WAXS ?

- SAXS provides a low resolution electron density map, but requires precise background-scattering (from buffer) subtraction.
- Water scattering measured by WAXS is an ideal reference to determine the background-scattering contribution to SAXS.
- Determine the orientation of individual protein domains within a complex



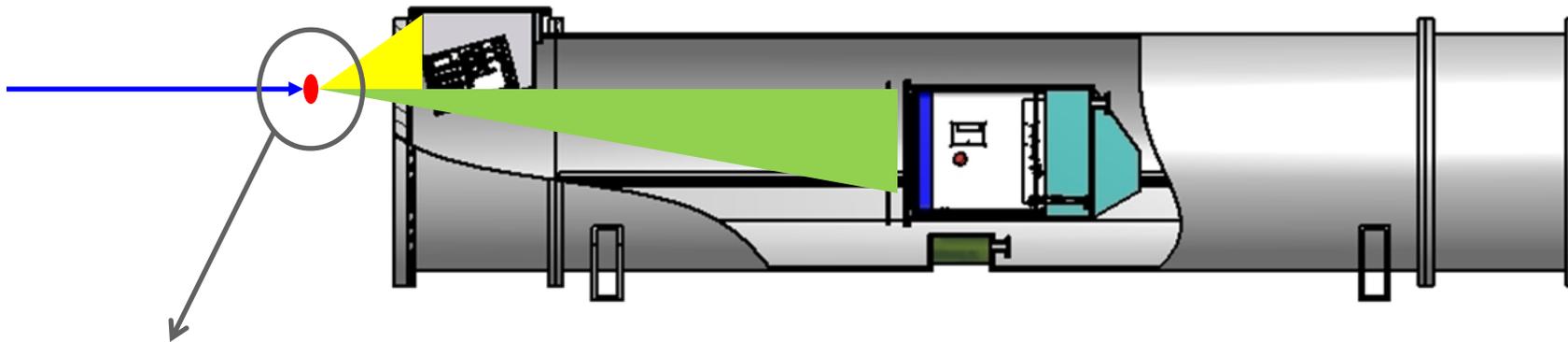
The ensemble of global structures of the ribosome-binding structural element determined by SAXS

Zuo X et al. PNAS 2010;107:1385-1390



Enhanced SAXS/WAXS beamline:

Dedicated beamline

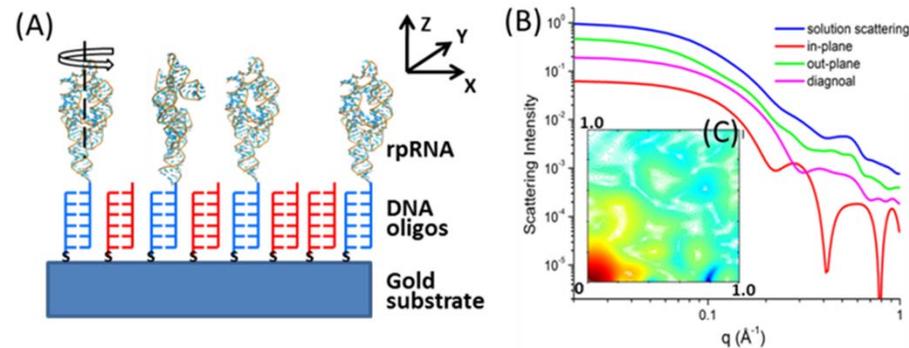


- Dedicated sample environment for macromolecular solution scattering
 - Fast protein liquid chromatography, for sample purification (distinguish dimer and monomer versions of a protein)
 - SAXS sample cell (quartz capillary) equipped with liquid flow system
 - UV/Vis, for simultaneous measurements, and automatic sample loading system



SAXS/WAXS Capabilities

Beamline	Full Flux (Ph/s)	Nominal Beam size (μm)	Energy range (keV)	Q-range	Simultaneous SAXS/WAXS	Quick detector distance change	Anomalous SAXS	Temperature controlled flow cell
Operational								
APS 12-ID-B	1×10^{13}	400 x 9	7.6 - 14	0.001 - 2.6	Y	Y	Y	Y
APS 12-ID-C	2×10^{13}	500 x 300	5.5 - 35	0.001 - 2.8	N	Y	Y	Y
APS 18-ID-D	2×10^{13}	160 x 60	3.5 - 39	0.0071 - 2.0	N	N	Y	Y
Petra-III SAXS	1×10^{13}	200 x 60	4 - 20		Y	Y	Y	Y
Under Construction								
NLSL-II LIX	2×10^{13}	1 or 5 x 10	2.1 - 18	0.003 - 3.0	Y	Y	Y	Y



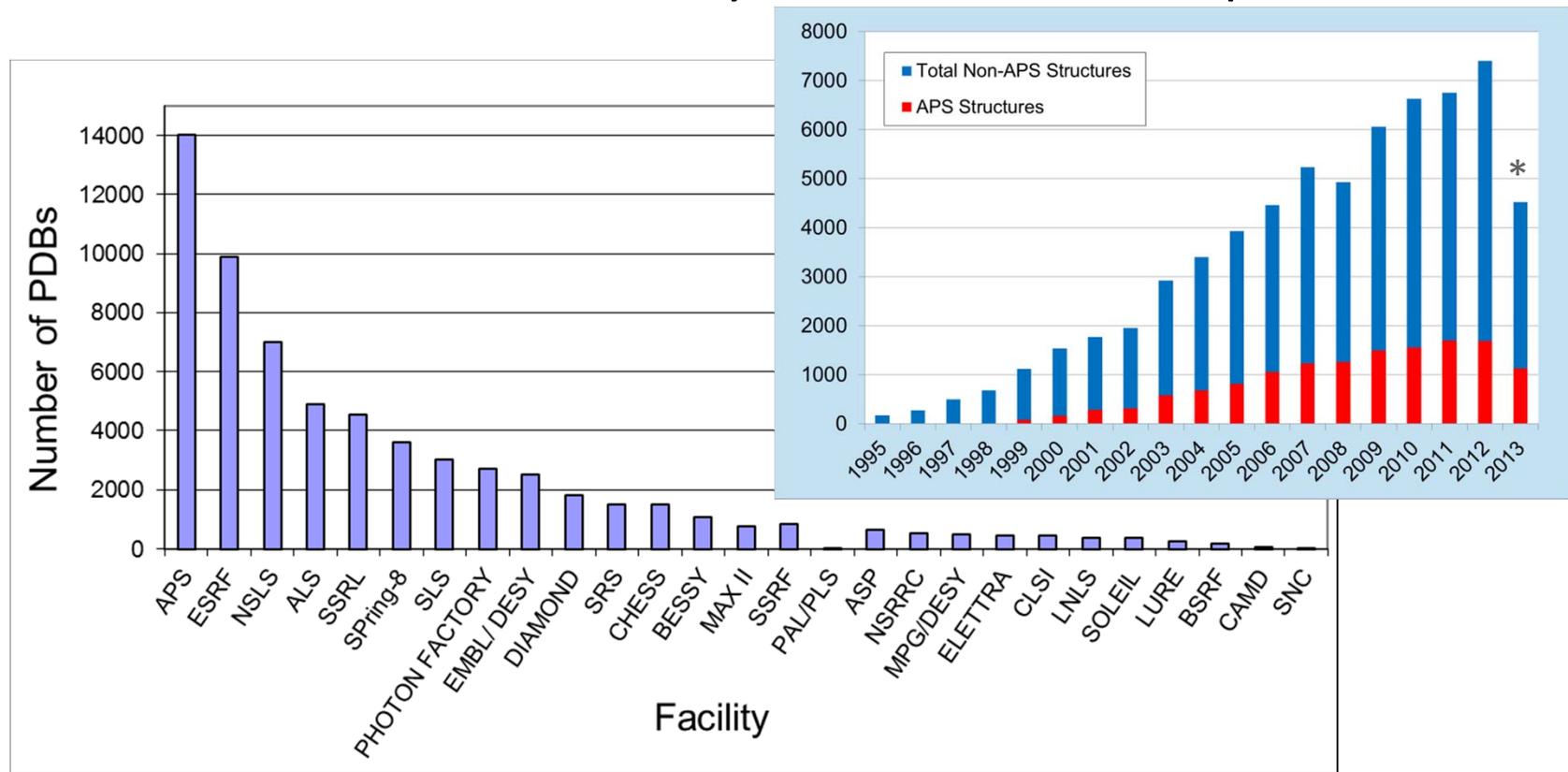
(A). Schematic drawing of surface alignment of rpRNA on DNA-gold substrate
 (B) Simulated solution x-ray scattering (blue) and line cut profiles (in-plane, red; out-of-plane, green; diagonal, magenta) of simulated 2D GIXS pattern (C, inset) of rpRNA with displayed uniaxial alignment.



Macromolecular Crystallography Protein Data Bank Deposits (as of January 2014)

APS MX beamlines

- contribute more than any other facility world wide
- contributed 20% of all synchrotron based deposits



* 2013 numbers are still changing rapidly



Improving human health

Scientific Opportunity

Membrane proteins and proteins complexes mediate cellular responses, act as cellular gateways, have been implicated in many diseases, and are the target of many drugs.

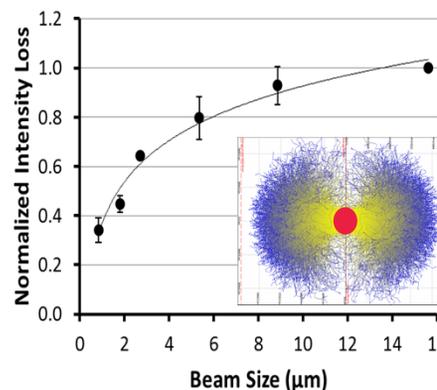
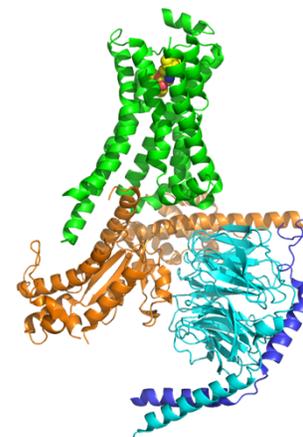
Breakthrough Techniques

Microcrystallography has recently enabled the determination of high impact, 3D structures of these complexes. Crystals tend to be small (<10 μm), inhomogeneous and weakly scattering. Recording full data sets requires merging partial data sets from many crystals.

MBA-lattice Enabler

Will allow one to obtain critical data from nano-crystals (>500 nm), and to mitigate the effects of radiation damage by exploiting both the high brightness and high X-Ray energy of the new MBA-lattice.

β_2 adrenergic receptor-Gs protein complex – the first structure of a human GPCR



Submicron beam size reduces radiation damage

Brian Kobilka shared the 2012 Nobel Prize in Chemistry for studies of G-Protein-Coupled Receptors



Understanding protein synthesis and better antibiotics

Scientific Opportunity

Ribosomes are large, complex molecular machines that translate the genetic code of mRNA to synthesize proteins. Antibiotics block protein synthesis killing bacterial infections. Polysomes (ribosome clusters) simultaneously read mRNA to synthesize proteins.

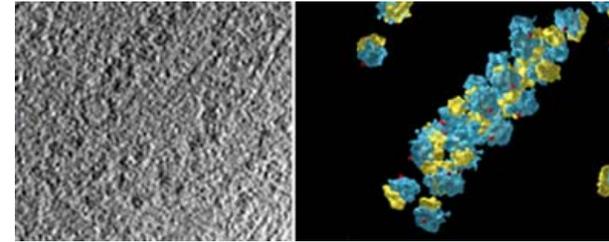
Breakthrough Techniques

Highly collimated beams were required to solve the ribosome structure. Weak scattering and extreme unit cell dimension (2700 Å) from polysome crystals require enhance microcrystallography techniques.

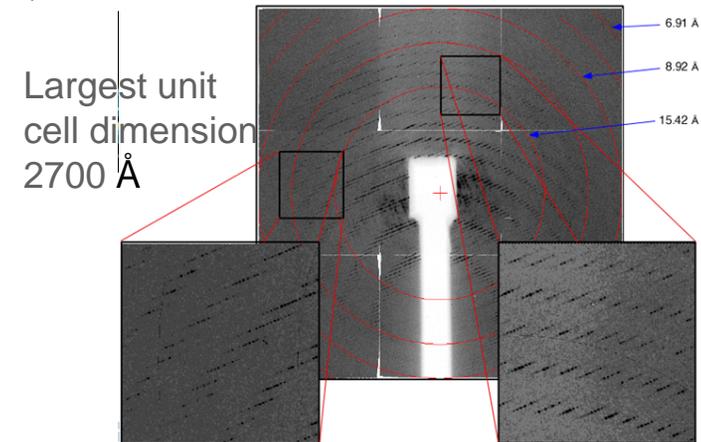
MBA-lattice Enabler

The large unit cell and weak scattering can be combated by the intense, extremely collimated beam to probe micro-crystals or well-ordered regions of larger crystals.

EM reconstruction of polysome



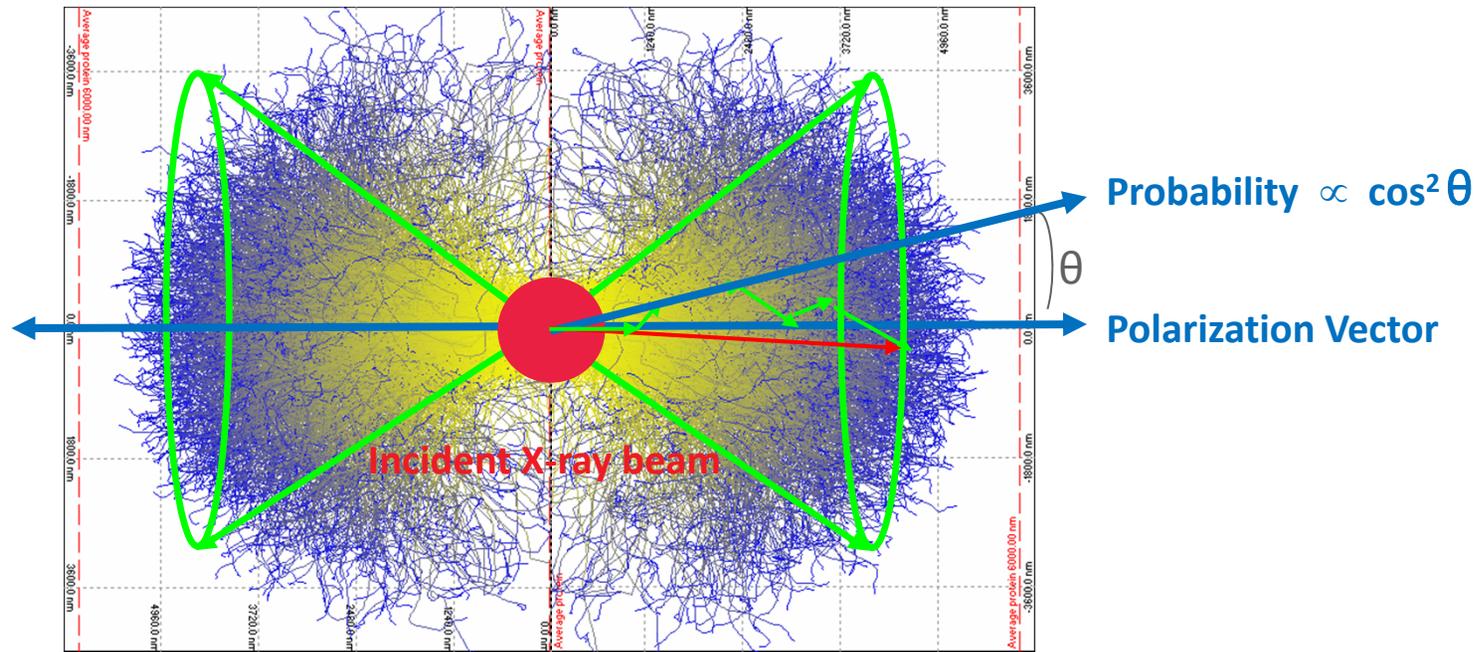
Diffraction pattern from a polysome (5 ribosomes)
(Courtesy of Dr. Jaime Cate, U. of California Berkeley)



Ada Yonath shared the 2009 Nobel Prize in Chemistry for studies of the structure and function of the ribosome.



Photoelectron emission and Monte-Carlo simulations



Casino Monte Carlo Simulation (by Gerd Rosenbaum)

Known facts:

1. Photoelectrons are preferentially ejected along the polarization vector.
2. Along their path they interact with atoms, lose energy and create radicals by ionizations, and are scattered.

Monte Carlo Simulations (Casino) by Nave and Hill, *J. Synchrotron Rad.* (2005) 12, 299–303

1. For photon energies used for biological crystallography, PE path lengths are a few microns.
2. A significant fraction of damaging energy may escape the illuminated volume.

Note: the reach of electrons (red arrow) is shorter than the path length (green arrows) calculated from the Constant Slow Down Approximation.

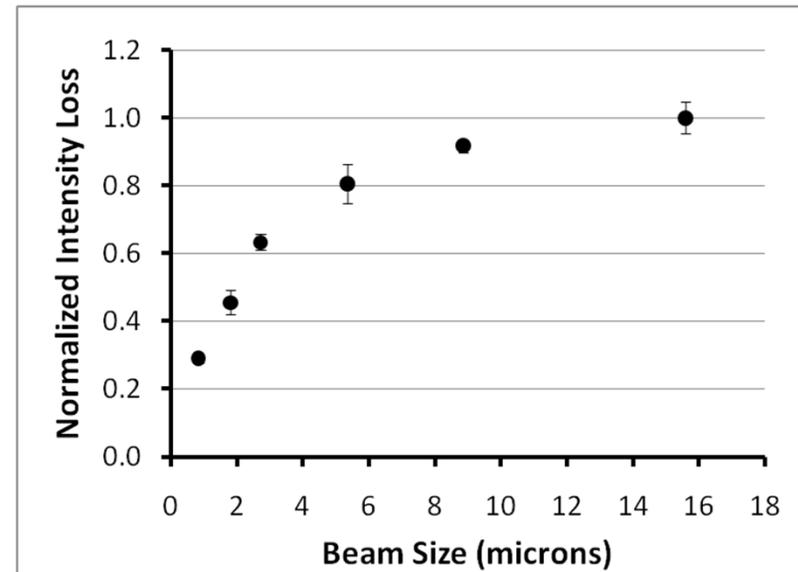
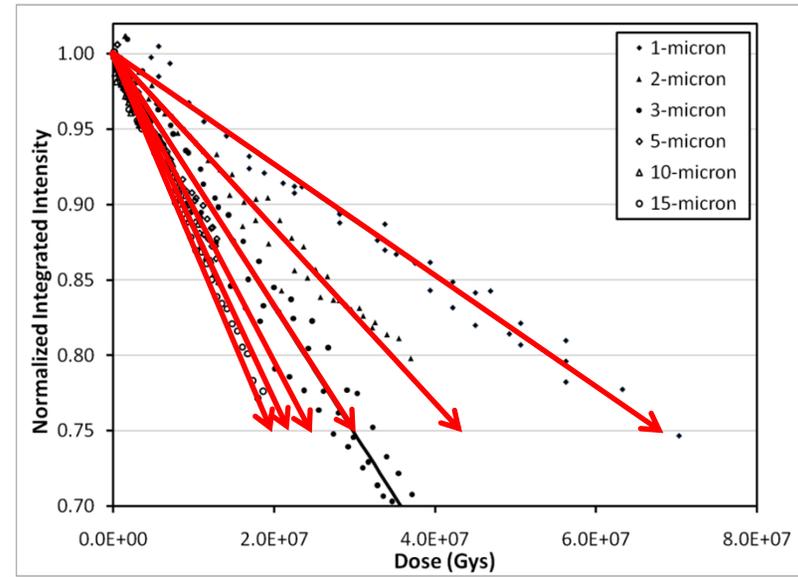


Intensity loss as a function of beam size and dose

Experimental protocol

- X-ray energy = 18.5 keV
- Collect 1° diffraction images at identical omega angle (same frame)
- Integrate all diffraction spots
- Sum all intensities (both full and partial)
- Normalize total intensity of each frame to the 1st frame
- Plot intensity vs. dose for each beam size
- Plot slope(i)/slope(15) vs. beam size

Damage decreases 3-fold with beam size



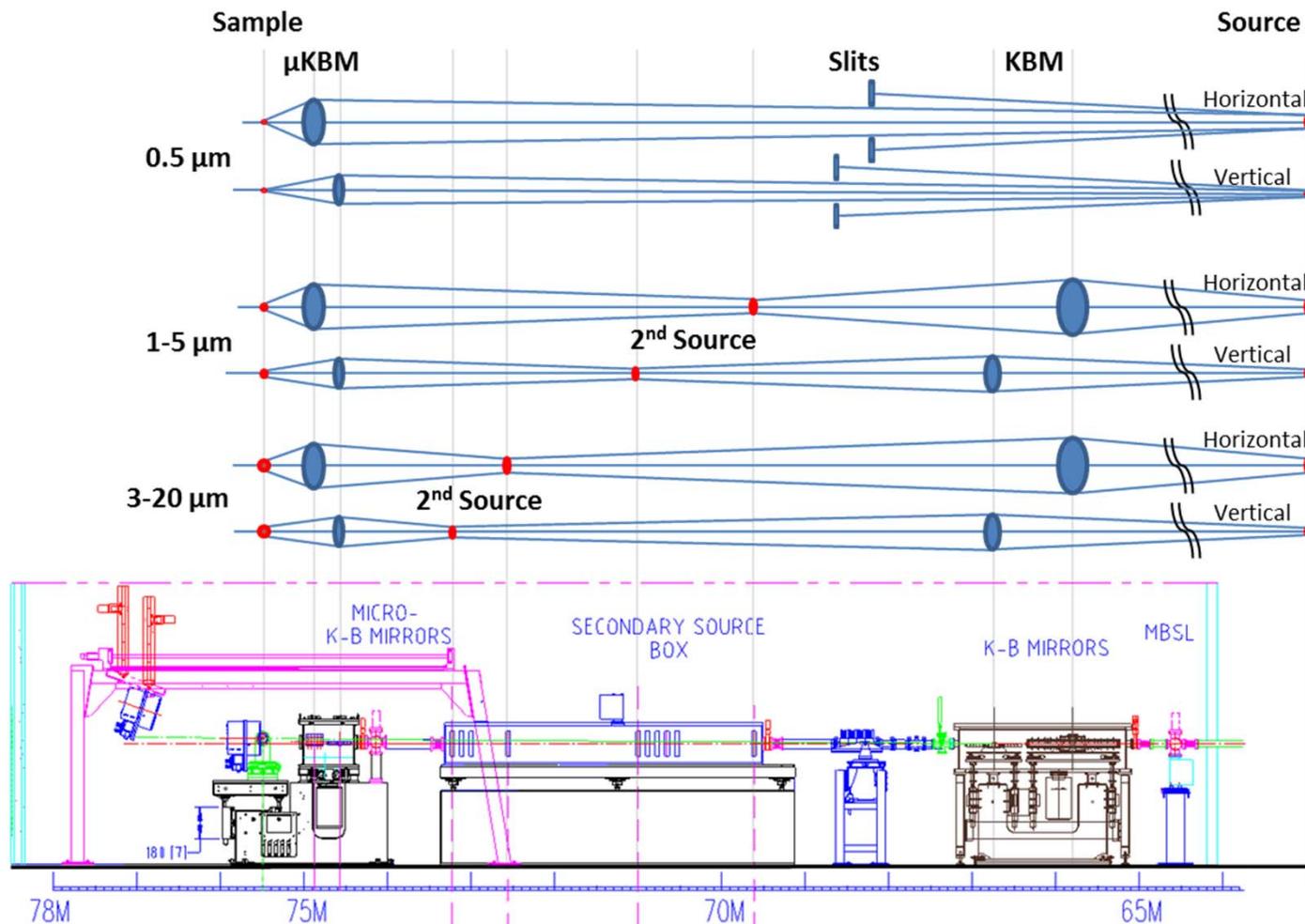
GM/CA Microfocus Upgrade - Now and with MBA-lattice

Variable focus from 1 – 20 μm

Intensity in 1 μm beam at sample position: 5×10^{10} photons/s, $< 1000 \mu\text{rad}^2$ @ 12.0 and 18.5 keV

MBA Lattice would enable world leading capability

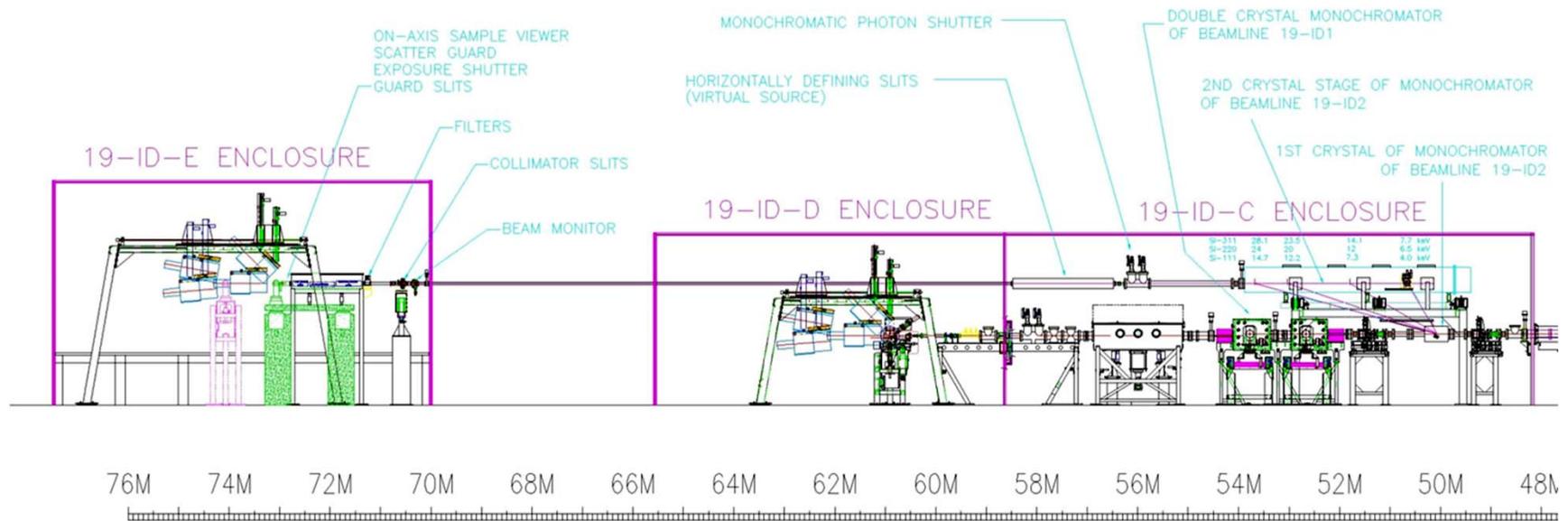
\rightarrow 0.5 μm beam with $\sim 1.2 \times 10^{13}$ photons/sec, $\sim 500 \times 300 \mu\text{rad}^2$ @ 12.0 keV



Proposed Micro-focus Beamline options:

1: Structural Biology Center

2: move SBC to new sector – wide field imaging on S19



1- μ m line focus, 1×10^{12} photons/sec



Time to the Garman Limit (cryo-cooled)

Garman limit¹ ~ 3.0 x 10⁷ Gray (35% intensity loss)

Deposited energy in sample – not incident energy!

E ~ 12.68 keV

Beamline	Divergence (μrad, FWHM)	Smallest beam width (μm)	Smallest beam height (μm)	Flux (ph/sec)	Flux density (ph/s/μm ²)	Dose rate (Gy/s)	Time to Garman limit (msec)
APS-U MBA 23-ID-D	3200 x 1200	0.40	0.50	5.3E+13	3.4E+14	1.0E+11	0.29
NLSL-II FMX‡	1700 x 700	1.00	0.50	5.0E+12	1.3E+13	3.9E+09	7.60
DLS VMX¥		0.50	0.50	1.0E+12	5.1E+12	1.0E+09	29.90
APS-U MBA 23-ID-D	270 x 180	6.10	5.20	6.1E+13	2.4E+12	7.6E+08	39.52
Petra3 MX2	500 x 300	4.00	1.00	5.0E+12	1.6E+12	4.9E+08	60.81
SPring8 BL32XU§	1520 x 980	0.90	0.90	6.2E+10	9.7E+10	3.0E+07	992.99
Petra3 MX1	200 x 150	28.00	13.00	1.0E+13	3.5E+10	1.1E+07	2766.63
APS 23-ID-D*	400 x 100	5.00	5.00	5.4E+11	2.8E+10	8.5E+06	3518.81
DLS I24	2000 x 50	8.00	8.00	1.0E+12	2.0E+10	6.2E+06	4864.40
ESRF ID23-2†	550 x 360	7.50	7.50	4.0E+11	9.1E+09	2.8E+06	10688.38
APS 23-ID-D*	400 x 100	70.00	25.00	2.00E+13	1.46E+10	4.50E+06	6650.55

*APS 23-ID intensities are for 12.0 keV except where noted

§SPring8 BL32XU intensities area at 12.398 keV

†ESRF Upgrade may have changed these numbers

‡NLSL-II AMX/FMX intensities are at 12.7 keV

¹ Owen, R.L., Rudino-Pinera, E. & Garman, E.F. *Proc Natl Acad Sci U S A* **103**, 4912-7 (2006)

² RADDOS http://biop.ox.ac.uk/www/garman/lab_tools.html



In situ Data Collection

Scientific Opportunity

In Situ screening will provide important diffraction feedback on limited quantities of biological material at an early stage in crystallization trials.

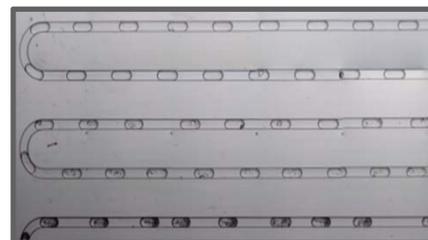
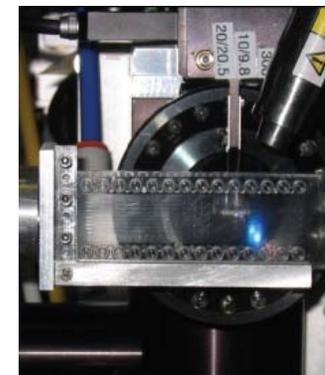
Breakthrough Techniques

- Samples introduced by novel delivery systems (e.g. acoustic drop or microfluidics)
- Data collection on large number of micro-crystals complexed to a variety of compounds.
- Data collection on high symmetry space groups (e.g. viruses)

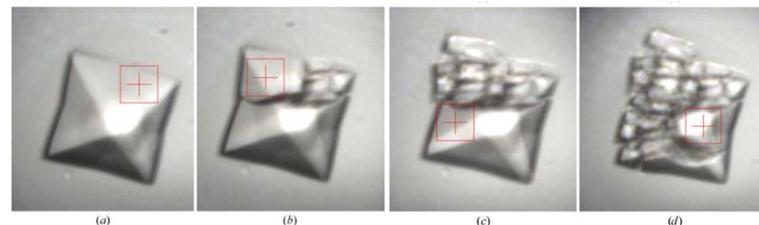
MBA-lattice Enabler

Higher brightness and faster detectors employed in the search for every shrinking crystals of increasing complexity and biological importance.

Microfluidic crystallization card on a goniometer with a mini-beam collimator

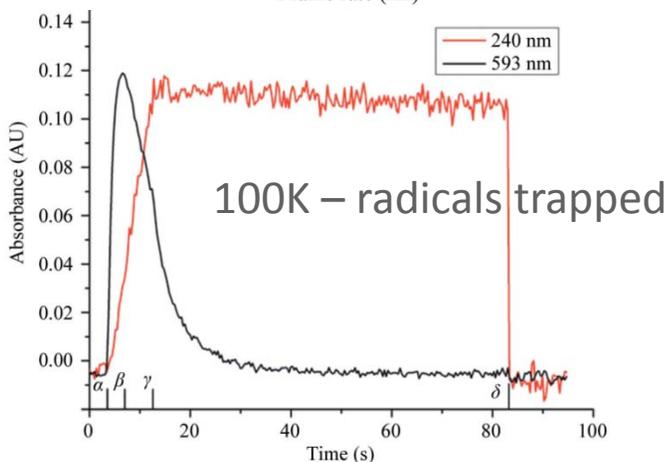
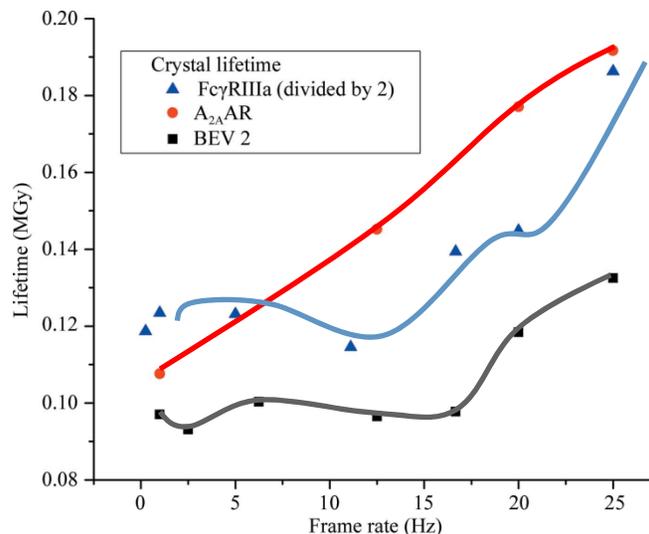
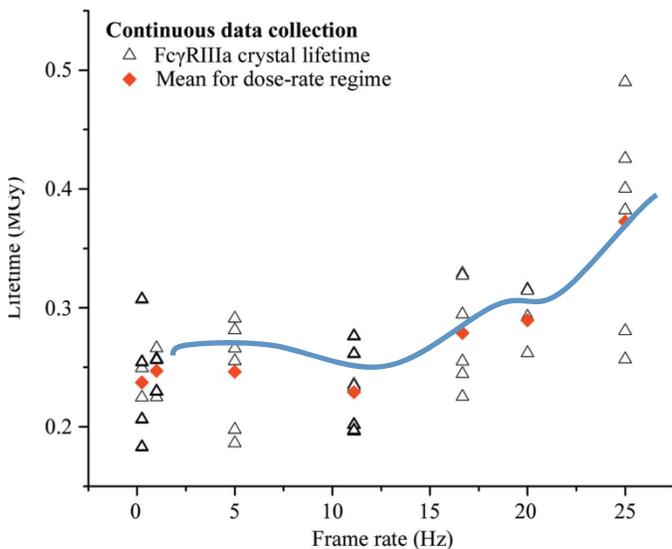


Many crystals in micro-channels for rapid data collection



Crystallization tray - containment for virus crystals. Radiation damage occurs quickly (<100 ms) requiring many crystals.

Outrunning 2nd Rad Dam at Room Temp - Beware!



Crystal lifetime
Start-stop
 40 msec exp; 4 sec dead time → 0.186 Mgy
Shutterless
 >90 msec exp → 0.257 MGy
 40 msec exp → 0.373 Mgy
Compare cryo-cooled → 30 MGy

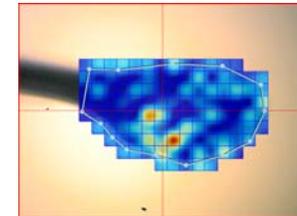
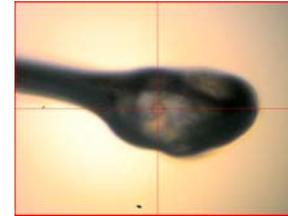
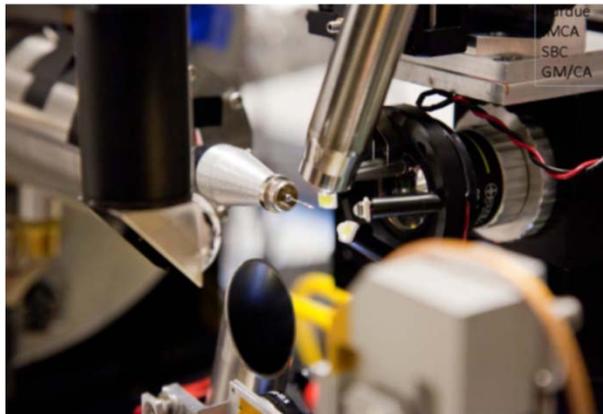
Black – generation of aqueous or solvated e⁻, followed by 1st order decay
 Red – postulated to be hydroxyl radical

Owen *et. al.* Acta Cryst. (2012) D68, 810-818



Second Order Non-linear Imaging of Chiral Crystals (SONICC) / Two-Photon Excitation UV Fluorescence

- High-sensitivity technique to detect sub-micron sized crystals, even in turbid media such as lipidic cubic phase (LCP).
- Measures Second Harmonic Generation (SHG) signal that arises from interaction of high-field laser with anharmonic polarizability tensor of chemical bonds.
- Multi-CAT collaboration
GM/CA, IMCA, SBC
- Pioneered by Garth Simpson's group at Purdue University.



Sample containing human opioid receptor in lipidic cubic phase from Vadim Cherezov (TSRI).
Top: sample in bright field
Middle: SONICC image in laser focal plane
Bottom: Diffraction in JBluice .

MICHAEL BECKER, ANL



Time resolved MX: probing macromolecules in action

Allosteric action in real time:

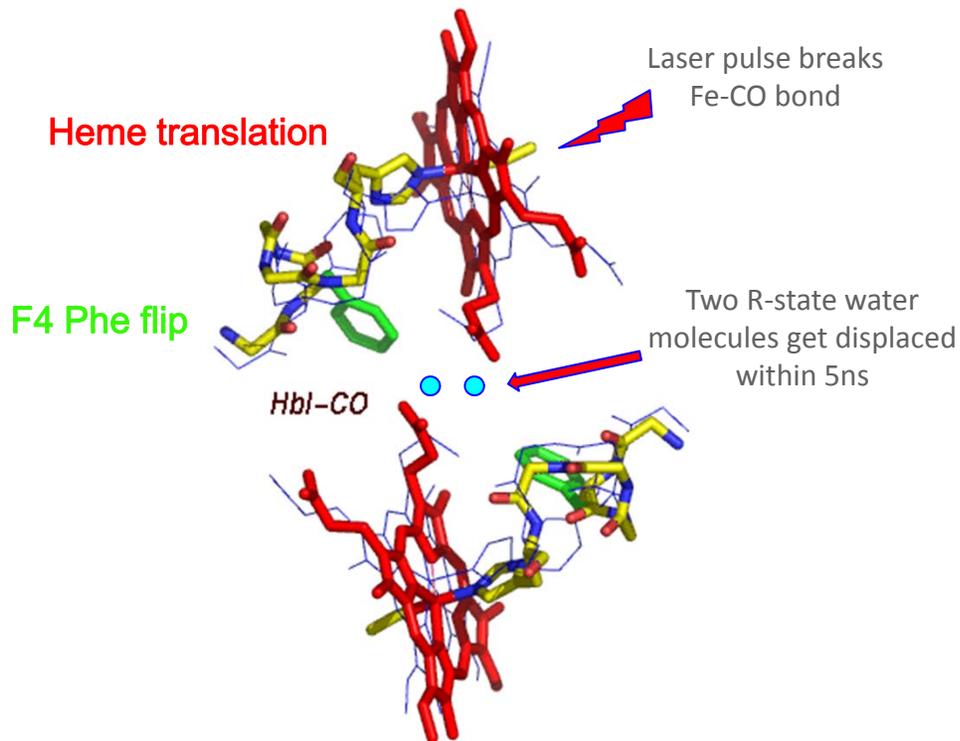
Time-resolved crystallographic studies of a cooperative dimeric hemoglobin. (Hbl)

W.E. Royer, Univ. of Mass. Medical School and V. Srajer, BioCARS

Knapp et al, Structure 17, 1504 (2009)

Srajer and Royer, Methods Enzymol. 437, 379 (2008)

Knapp et al, PNAS 103, 7649 (2006)



Models derived from difference refinement for the following individual time points: dark, 5ns, 200ns, 700ns, 2 μ s, 9 μ s, 80 μ s

Key structural transitions in dimeric Hbl

What is the cascade of structural events?

An intermediate is formed rapidly (100ps) upon CO ligand release, relaxing to Hbl T-like structure in the μ s time domain.

Are these transitions concerted or sequential?

Key allosteric changes appear to be tightly coupled.

What is the triggering event?

Rapid disordering of two water molecules H-bonded to heme propionates.

Do structural intermediates facilitate R to T transition?

Disordering of water molecules appears to lay the foundation for subsequent heme movement.



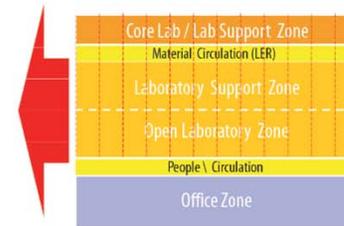
Time Resolve MX: future needs

<p>Large-area analog pixel array large-area detector (APAD) and associated X-ray chopper</p>	<p>Main challenge for time-resolved studies of macromolecules: Studies of irreversible reactions</p> <p>Solution: Single laser pump, multiple X-ray probe measurement. Requires fast read-out, large-area detector to allow recording of multiple consecutive frames following single reaction initiation. Need analog not photon counting PAD. A mechanical chopper is needed to protect sample from radiation damage.</p>
<p>Enlarging the 14-ID-B experimental station</p>	<p>A larger end station is needed to house a large-area detector and accommodate additional customized instrumentation for expanding time-resolved user community.</p>

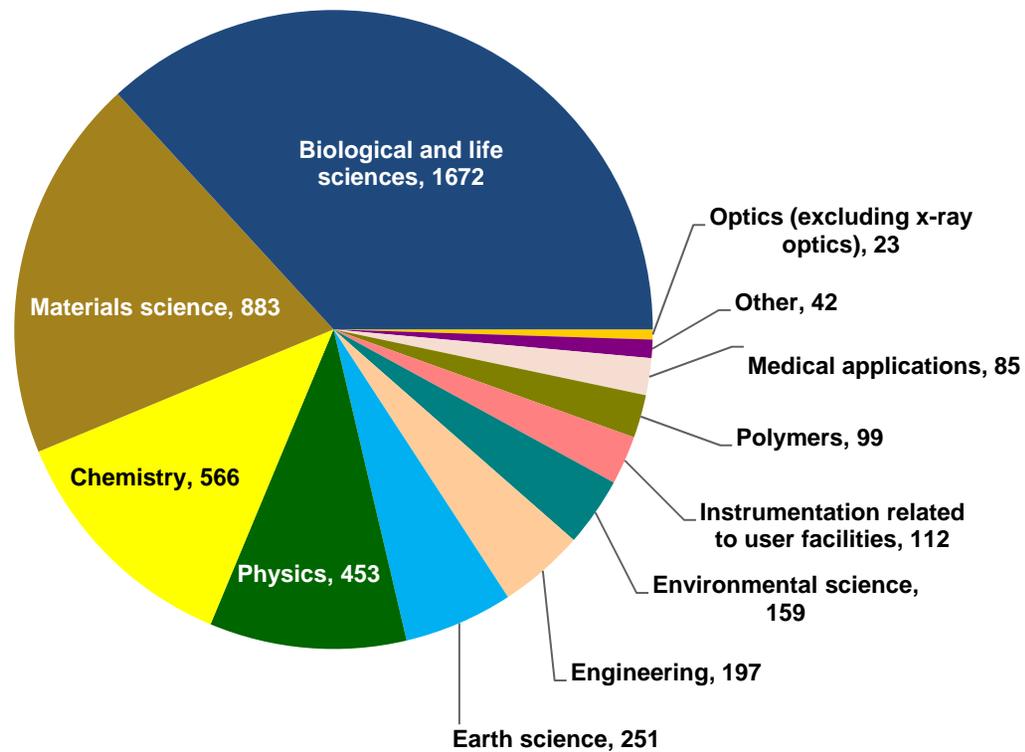


Advanced Protein Crystallization Facility (APCF)

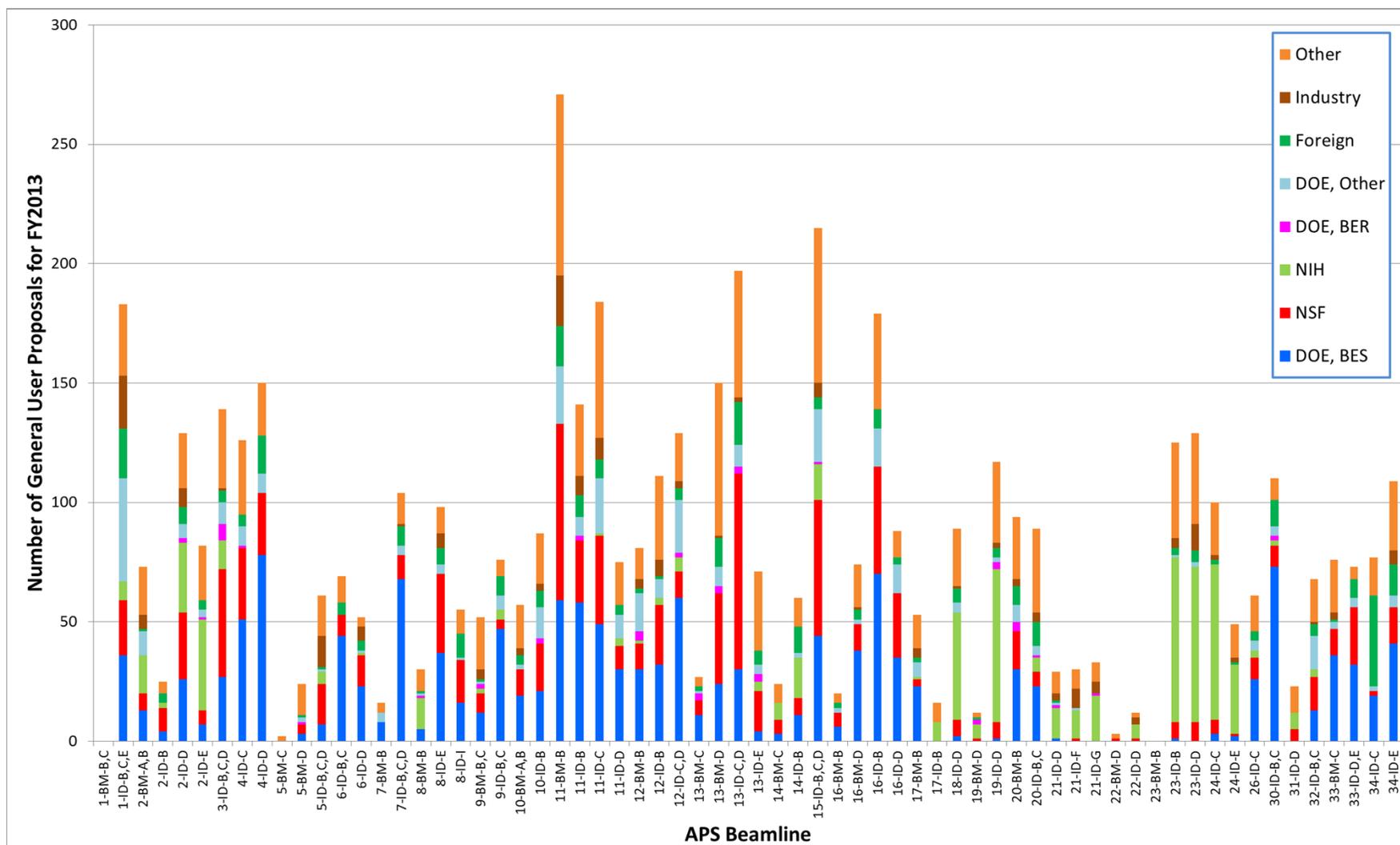
- A state-of-the-art crystallization laboratory integrated with a collaboration facility in a 60,000 GSF building next to APS MX beamlines
- Includes laboratories with high-throughput robot-assisted technologies for the production and structural and functional characterization of proteins and macromolecular complexes
- Will host structural genomics, structural biology and biological systems science programs
- Will take full advantage of the APS and Argonne's computing resources to determine structures and characterize functions of macromolecules
- Will enhance APS capabilities and be a resource for the national life sciences community
- Will enable collaborative research by providing research facilities for community projects, by hosting visiting scientists and by developing informatics gateways



APS Users by Experiment Subject (Fiscal Year 2013)



Number of General User Proposal vs. Beamline and Funding Agency (Fiscal Year 2013)



Seek External Funding

BES funding for life sciences beamlines is highly unlikely

We have engaged NIH in discussions

August 2010

February 2014

Need more frequent conversations

Need active support from the user community

Submitted a S10 High End Shared Instrumentation Grant

Anticipate funding for a Pilatus3 6M or Eiger 16M

Other potential sources of funding

BER for environmentally relevant science

HHMI occasionally invests in beamline development

Industrial



Acknowledgements



Chris Jacobsen
Stefan Vogt
Randy Winans
Xiaobing Zuo
Keith Brister
Keith Moffat



Conclusions

We plan to provide a significantly enhanced suite of instruments

- to better understand how proteins, and macromolecules function and interact
- probe membrane proteins and other difficult to crystallize proteins
- explore opportunities for reduced radiation damage
- solve protein structures for dynamic , irreversible processes
- determine larger ensembles and molecule interactions using enhanced SAXS/WAXS
- to probe trace element content at the subcellular level, and investigate their role in cell biology, environmental stewardship, disease, and nanomedicine.

“make a connection from proteins to organisms”

