



The SER-CAT SPECTRUM

Biannual Newsletter of the Southeast Regional Collaborative Access Team · Vol. 9, No. 1 · Summer 2011



Director's Message

Bi-Cheng Wang

Dear Colleagues,

Greetings! I hope your summer has been fun!

Since the formation of SER-CAT, one goal of its members was for the SER-CAT organization to someday become a "research support group" for sharing informational resources, in addition to just gaining access to the synchrotron. Thanks to all the members and staff involved, this aspiration is now a reality through SER-CAT's outward reaching activities. For example, SER-CAT's 8th Annual Symposium on the campus of North Carolina State University was another success. Congratulations to Professors Robert Rose and Carla Mattos for organizing such an outstanding gathering. Congratulations also to Dr. Fred Dyda and Mr. Stefan Gajewski for receiving this year's SER-CAT Scientific Awards (see page 2).

Plans for next year's Symposium are well underway, and the event will be hosted by Professor David Rodgers on March 16, 2012 at the University of Kentucky. Please mark your calendars.

Along with the philosophy of providing members with a "virtual home synchrotron source", I am pleased to report that more than 90% of our users access SER-CAT's beamlines from their home labs. With the successful implementation of our 16 hours/day and 7 days/week on-site user support, users may now request 8-hour slot assignments to better suit ones needs. As a successful example of how users may now gain access to SER-CAT from everywhere in the world, Professor Zhi-Jie (James) Liu was able to collect data at SER-CAT from his lab at the Institute of Biophysics in Beijing, China. Page 4 includes an exciting report of his real time experience for collecting data from half way around the world.

SER-CAT staff members have also made some significant advances at our facility. I hope that you will enjoy reading about these and other highlights in this issue of *The SER-CAT Spectrum*. If you have any comments or suggestion to improve SER-CAT's operations, please let us know. We appreciate you helpful input and interest.

Best wishes,
B.C.

9th SER-CAT Symposium & Board Meeting

March 16-17, 2012

University of Kentucky

Lexington, KY

Hosted by Professor DAVID RODGERS

Meeting details concerning registration and meeting information will be posted soon on SER-CAT's website



Nominations, Please

Nominations for the two annual SER-CAT Research Awards are requested. Please send your nominations for the 2012 SER-CAT Young Investigator Award and/or the Outstanding Science Award to Prof. John Rose before January 10, 2012. For additional information, please click on the links below:

[2012 SER-CAT Young Investigator Award](#)

[2012 SER-CAT Outstanding Science Award](#)

or

Send [John Rose](#) an email.

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8th SER-CAT Symposium by Gary Newton

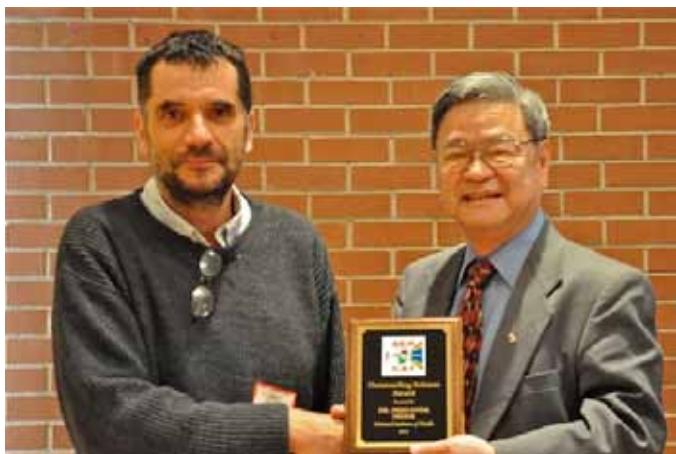
The symposium began on the morning of March 4, 2011 with welcoming remarks from the NCSU Vice Chancellor for Research Terri Lomax followed by a welcome message from the NCSU meeting host Prof. Robert Rose.

SESSION 1 - GTPases and Kinases: Chair **Prof. Carla Mattos** (NCSU). The speakers in this session were **Prof. John Sondek** (UNC-Chapel Hill), **Dr. Stephen White** (St. Jude Children's Research Hospital) and **Dr. Fred Dyda** (NIH-NIDDK), **2011's SER-CAT Outstanding Science Award winner**.

Prof. Sondek's title was "General Regulation of Phospholipase C Isozymes (PLCs)". He explained that there are thirteen PLCs in humans which are activated by various inputs and, despite the diverse array of controlling inputs, general mechanisms govern both the inhibition and activation of PLCs. For additional discussion, see **J. Lipid Res.**, 2009, 50, S243-248.

Dr. White's title was "The Structure and Mechanism of Pantothenate Kinase (PanK) and Modulation by Small Molecule Effectors". He and his group are studying the structure, mechanism, kinetics and control of PanKs in mammals and screening for small molecule effectors. Another target is bacterial PanKs as potential targets for new antibiotics. For further discussion, see **Chem. and Biol.**, 2010, 17, 892-902.

SER-CAT Outstanding Science Award Lecture: Dr. Fred Dyda, NIDDK, NIH --- Dr. Dyda's title was "When Dynamin meets Dynamin: a Catalytically Stimulating Encounter". He explained that dynamin is a large, atypical GTPase that catalyzes membrane fission. Using X-ray crystallography, they solved a high-resolution structure of the GG dimer at 2.0Å resolution complexed with the transition-state mimic GDP.AIF4-. Dimerization also causes a geometrical constraint in dynamin oligomers that in turn could contribute to membrane fission. For further discussion, Chappie, S. *et al.* (2010) **Nature**, V466, 435-440.



Prof. Wang presents the SER-CAT Outstanding Science Award to Dr. Fred Dyda (NIDDK, NIH)

SESSION 2 – Protein Complexes and Interactions: Chair **Antonella Longo**. Speakers in SESSION 2 were **Mr. Stefan Gajewski**, the winner of the 2011 Young Investigator Award, **Dr. Bob Reid**, GSK and **Dr. Bob Rose**, NCSU.

SER-CAT Young Investigator Award Lecture: Mr. Stefan Gajewski, St. Jude Children's Research Hospital.

Bacteriophage T4 is a well characterized model system for studying DNA metabolism in vivo and in vitro. They solved the crystal structure of the UvsX core domain (30- 350) and found it to align very well with RecA, despite the poor sequence homology. Finally, they were able to demonstrate that the Bacteriophage T4 Helicase UvsW is required to complete UvsX mediated branch migration reactions and is also capable of rescuing UvsX activity in disfavoring in-vitro conditions. This study provides novel functional and dynamical data on the central process of DNA metabolism in Bacteriophage T4. For further discussion, see Stefan Gajewski, **JMB** (2011) 405, 65-76

Dr. Reid's title was "AcrBIDARPin (chaperone) co-crystallography and phage display". Bacterial integral membrane efflux pumps are drug targets for potentiating antibiotic activity. Using Structural Biology approaches, they are working to understand drug molecular interactions for the E. coli efflux pump AcrB. Also described is the use of phage display to generate crystallization chaperones for other structural targets.

The next speaker was Bob Rose, NCSU. Dr. Rose's title was "The anatomy of a dual-specificity interface of the transcriptional coactivator DCoHz". They propose that the DCoHI homotetramer is kinetically, as opposed to thermodynamically, very stable. This may result in preferential stabilization of HNF-1 α homodimers by DCoHI, instead of HNF-1 α /HNF-1 β heterodimers.



Prof. Wang presents the SER-CAT Young Investigator Award to Mr. Stefan Gajewski

(St. Jude Children's Research Hospital)

...Continued on page 3...

SESSION 3 – DNA polymerases and DNA modifications: Chair Prof. Bob Rose, NCSU. The speakers in SESSION 3 were Dr. Sepideh Khorasanizadeh, Prof. Wei Yang, Dr. Scott Williams and Prof. Lorena Beese.

Dr. Khorasanizadeh's title was "Epigenetic Mechanisms Mediated by Lysine Methylation". Research in this group searches for the interaction of proteins with nucleic acids, peptides and small molecules associated with chromatin. In eukaryotes, genomic DNA and histones form a conserved unit particle known as the nucleosome. The recognition of the histone H4 tail monomethylated at lysine 20 by the MSL3 chromodomain was discussed.

Prof. Wei Yang's title was "Alternative Mechanisms for Translesion DNA Synthesis". DNA lesions due to the loss of bases or chemical modifications prevent normal Watson-Crick (WC) pairing and stall normal DNA polymerases. In this presentation, different mechanisms of trans lesion synthesis by a B-family member and the Y-family human DNA pol II was discussed.

Dr. Scott Williams (NIH NIEHS) title was "Cleaning up after DNA ligases in the DNA damage response". DNA ligases finalize DNA replication and repair through imperfect DNA nick-sealing reactions. Structures and mutagenesis support a HIT-Znf direct damage reversal repair mechanism, and illuminate protein folding, active site distortion, and FPK-pivot mutations underlying APTX dysfunction in neurodegenerative disease. Prof. Lorena S. Beese *et al.*, (Duke University) title was "Hu-

man exonuclease I complexes with DNA". Human exonuclease I (hExoI) plays important roles in DNA repair and recombination processes that maintain genomic integrity. The relative arrangement of binding sites in these enzymes provides a solution to a complex geometrical puzzle of substrate recognition and processing.

SESSION 4 – APS and SER-CAT Updates

The speakers in SESSION 4 were Dr. Dean Haeffner and Dr. John Chrzas.

Dr. Haeffner's title was "The Advanced Photon Source Upgrade Project". Dr. Haeffner discussed the ongoing \$350M APS-U project, continued over the next 6-7 years. There are two primary scientific themes and 6 beam line themes.

Dr. John Chrzas' title was "Status of SER-CAT Facility". Dr. Chrzas discussed the advances at SER-CAT: Bruker/Maa-tel MD2 diffractometer; the new software capabilities such as sample rastering and helical data collection and other developments.

EVENING SESSION

In the evening, we were bussed to the North Carolina Museum of Art for dinner. Before dinner, guests were invited to wander through the museum to see the various art pieces on display. After a delicious dinner in the museum dining room, we heard a talk by Roy Campbell (director of exhibits, North Carolina Museum of Natural Sciences, Raleigh) who told us about the new museum at the Nature Research Center, now under construction. The new building will feature a large globe of the earth in the front of the museum [<http://naturalsciences.org>].



Participants at the 8th Annual SER-CAT Symposium and Board Meeting

Remote Data Collection from China: Bringing SER-CAT to the WORLD

by Zhi-Jie (James) Liu

The Zhi-Jie Liu group at the Institute of Biophysics, Chinese Academy of Sciences, Beijing, uses a combination of X-ray crystallography and functional studies to gain mechanistic insights into the functions of human disease related proteins at the molecular level. Higher resolution data is routinely collected at synchrotrons. However, because of the limited access to the Shanghai Synchrotron Radiation Facility (SSRF), we must routinely make trips to different synchrotron beamlines outside China for data collection experiments. Recently, we completed two remote data collection sessions, 12 hrs each, at SER-CAT from our home lab in Beijing. The remote data collection generated tremendous interest and curiosity locally, which could be gathered from the fact that more than 10 people were present throughout the collection, starting at 12:00 AM Beijing time and continued all night until noon the next day. All the crystals were frozen and shipped to SER-CAT in a Dewar via overnight express mail 5 days prior to the beam time. A computer with NX client (<http://www.nomachine.com>) was installed for accessing the remote connection. A network speed of 100 Mbp/sec worked well for all the maneuvers during data collection. In two 12-hour data collection sessions, more than 50 crystals passed through the robotic diffraction screening and 16 complete data sets were collected.



Kick-off for the first remote data collection in the lab's history.

Data collection at synchrotrons in "remote mode" provides expanded access for truly remote laboratories and for reaching out to other critical non-crystallographic team members with previously limited exposure to certain aspects of structural biology. This greater access can be leveraged not only to increase interest and participation of more laboratories for structural studies, but also integrate, in many instances, people working on upstream processes – purification and crystallization – with the downstream processes like structure solution and analysis. This will translate into greater efficiency, more accurate biological interpretation of structures and deeper insights by bridging the gap between biochemists and crystallographers. Other obvious benefits of data collection in remote mode include savings in cost, time, and travel.

The help extended by the support staff at SER-CAT is highly appreciated. Our special thanks to Albert, Unmesh, Palani, Zhongmin, Beth and John Chrzas (SER-CAT staff) for making this data collection possible. We also thank Drs. Wang and Rose for sharing their valuable SER-CAT research beam time with us and thanks to Lirong Chen for coordinating the data collection time. We find remote data collection at SER-CAT extremely efficient and SER-CAT's data collection user interface is very versatile and friendly.



The remote data collection from China is in progress

New Training Videos by John Rose

John Chrzas and the SER-CAT staff have produced a number of narrated training videos that are available on the SER-CAT web site (www.ser-cat.org), see "Beamline Users Guide", fourth item in the leftmost menu. ==>[USE FIREFOX]

The videos include:

1. Getting Started
2. Beamline Optimization
3. Changing Energy
4. Robot Optimization
5. Setting up the Sample Page
6. Sample Alignment
7. Diffraction Centering - sample rastering
8. Helical Data Collection
9. General Data Collection

The videos are meant to serve as both an introduction to data collection at SER-CAT for new users and a refresher course for past users who visit less frequently.

The videos also address an important training aspect for the increasingly large number of users who are collecting data remotely and thus do not have the advantage of on-site user training or face-to-face interaction with beamline staff.

Note: The videos are best viewed using the Firefox web browser since the video format may not be compatible with other web browsers such as Safari. Several new videos, including an automated sample screening, are planned and should be available shortly. If you have not already done so, please checkout the videos and let us know how to improve them. Also, if you have any ideas for a new training video, please contact John Rose (Rose@BCL4.bmb.uga.edu).

SER-CAT Kappa Update

by John Chrzas

This is a good time to provide an update on the status of the MK3 minikappa for the ID MD2 microdiffractometer. After a lengthy customs clearance, we now hope to install and test the device during the September shutdown. The availability for the next run will be determined by the success of the following projects.

Installation:

When we purchased the MD2, it included the control systems for the MK3. The expectation is that we will be able to install the MK3 and change the configuration of the motor data base and the device should work out of the box. If this works as planned, the installation of the MK3 should be straight forward. If there is a problem with the MD2 control system, then this might prevent us from starting the commissioning of the device. SERgui and MX:

The original installation of the ID sample hardware included a Rosenbaum/Rock mini kappa. The modification of the MX motor data base should be straight forward to turn the “new” kappa motors back on. The SERgui modifications will originally be centered on the safety interlocks required for kappa operation – to prevent collisions and keep samples in the cold stream during kappa moves. The long term objective would be to fully integrate STAC into SERgui in a seamless manner.

KAPPA Strategy:

Strategy for Aligned Crystals (STAC) is an object orientated software package for determining the optimal strategy using a kappa goniometer. The software was developed through the EMBL as part of the European kappa work group. Fortunately for us, the MK3 installation on an MD2 is fully supported by this software and our neighbors at NE-CAT have been using this software for their MK3 on an MD2 for about a year. The initial plan will be to use STAC as an independent software package. As we become more familiar with the software, we plan to fully integrate it into the SERgui environment.

Precision:

The current single axis omega device in use on the MD2 has a circle of confusion of 1-2 micrometers. The MK3 specifications have this number increasing to about 5 micrometers. Until we have the device installed and tested, we will not have a solid number for the precision of the kappa goniometer. Conversations with Malcolm Capel of NE-CAT lead me to believe that we might not be able to use our 10 micron pin hole with 10-15 micron samples, but may be forced to use the 20 or 50 micron pin hole to ensure the sample stays in the beam. Once the device is operational and ready for data collection, we will have to determine how to optimally schedule this device. If the precision is a problem for some in the user community, then we might consider scheduling time during the run with the kappa or single axis operations to allow members to choose which hardware configuration is best for them. Member feedback will be crucial during this commissioning period to determine how to best use the MK3.

Commissioning:

The commissioning of this device has multiple components with most not requiring x-rays. The protocols for collision prevention and kappa rotations can be developed without x-rays

and integrated into SERgui. X-rays are required for the measurement of data quality and strategy confirmation.



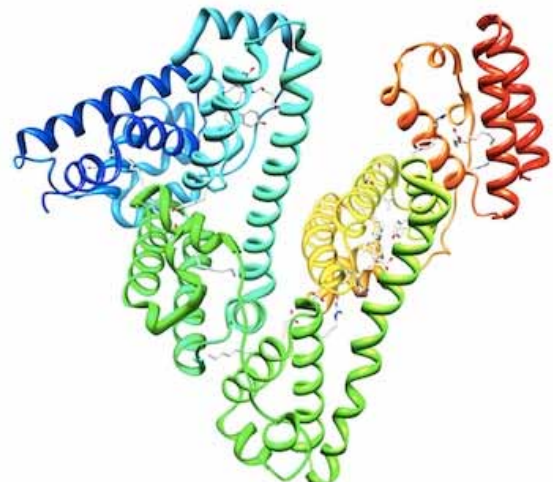
The MK3 minikappa goniometer

Summary:

We plan on installing and commissioning the MK3 on the ID MD2 during the September storage run shut down. If all of the different projects required for the experimental use of the kappa are completed by the beginning of the upcoming user run, then the kappa will be available for use for the next 22ID run. We will be asking the membership to report on their experience with the kappa so that we may determine if we will need to include the hardware configuration in our scheduling process.

If the kappa is not ready for use at the beginning of the run, then we will use beam time during the run to complete the commissioning of the device. Once the device is ready, we will expose the system to the users. If for any reason there are problems with the kappa, we can revert to the single axis system within 4 hours.

Can you identify the important protein shown below from its structure alone?



Answer in the next spectrum
or send email to newton@secsg.uga.edu

Fast Crystal Screening by John Rose

SER-CAT uses a highly modified ALS style robotic crystal mounter (See FIG. 1) with a capacity of 230 samples. The robot is extremely reliable and can change a sample (dismount-mount and center) in under 30 seconds!

SER-CAT has also developed a crystal screening system based on the ALS robot and the MD2 goniometer that can screen one sample every minute using optical loop centering.

In this mode, the crystal is mounted and two images are collected. The images are indexed, the crystal mosaicity is determined and a data collection strategy is generated. If diffraction centering is desired, screening time is increased to under 3 minutes per sample.

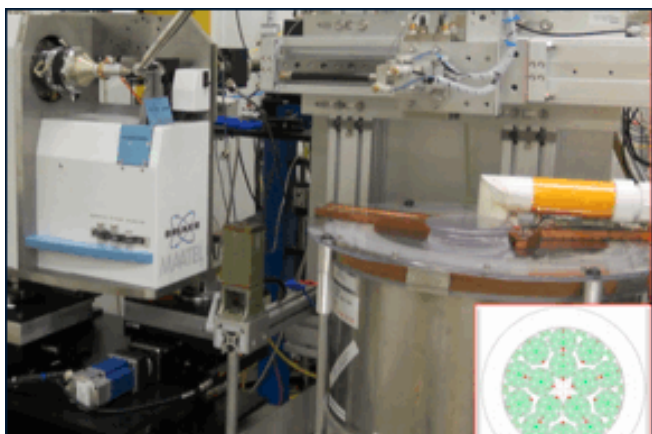


FIG. 1: ALS style robotic crystal mounter

All parameters, including an image of the crystal and the crystal's X, Y, Z centered coordinates are stored for future data collection and analysis.

The SERGUI Crystal Screening Page is shown in FIG. 2. A green background indicates successfully indexed crystals. The information panels also show diffraction resolution (based on the images collected), unit cell parameters and lattice type. A red background indicates screening failures.

The status column has a pull down menu that allows the user to: (1) skip the crystal, (2) mount and index the crystal or (3) collect data on the crystal.

A purple status background indicates crystals that have been screened, a yellow background indicates the status of the current crystal and the status of the remaining samples are indicated by a white background.

The user may select either enhanced loop centering based on the loop's diameter (size) or diffraction centering, if desired. The user may also upload an Excel spreadsheet (in cvs) format containing: (1) the sample description (puck ID, pin position, crystal/loop size and sample name); (2) status (skip, screen or collect data); (3) crystal centering method (loop or diffraction based); (4) desired data collection parameters (oscillation width, exposure time, number of images, starting phi, detector distance, energy, and attenuation). This information will be used as input to the screening process. If you would like more information on SER-CAT's automated crystal-screening system, please contact Dr. John Chrzas.

Note: a downloadable spreadsheet and a video tutorial for the automated screening process will be available shortly on the SER-CAT web site (www.ser-cat.org).

Sample	Status	Width	Time	#frames	Phi_0	Det_dist	Energy(ev)	%Transm	Resol	Lattice
9	Index Done	1.00	3.00	99	90.00	200	12658.00000	100	2.86	P1 5315 23
10	Index Done	1.00	3.00	154	175.00	201	12658.00000	100	2.29	P2 1859 31
11	Index Done	1.00	3.00	103	155.00	200	12658.00000	100	3.53	P3 3878 28
12	Index Done	1.00	3.00	157	35.00	199	12658.00000	100	1.86	P4 2906 33
13	Index Done	1.00	3.00	158	45.00	200	12658.00000	100	2.59	P5 8168 26
14	Skip	1.00	3.00	160	0.00	200	12658.00000	100	0.00	
15	Skip	1.00	3.00	160	0.00	200	12658.00000	100	0.00	
16	Skip	1.00	3.00	160	0.00	200	12658.00000	100	0.00	
17	Index Done	1.00	3.00	152	0.00	200	12658.00000	100	2.98	P6 2439 30
18	Index Done	1.00	3.00	149	50.00	201	12658.00000	100	2.33	P7 4323 87
19	Index Done	1.00	3.00	160	0.00	200	12658.00000	100	0.00	Failed
20	Mounting	1.00	3.00	160	0.00	200	12658.00000	100	0.00	
21	Index	1.00	3.00	160	0.00	200	12658.00000	100	0.00	
22	Index	1.00	3.00	160	0.00	200	12658.00000	100	0.00	
23	Index	1.00	3.00	160	0.00	200	12658.00000	100	0.00	
24	Index	1.00	3.00	160	0.00	200	12658.00000	100	0.00	
25	Index	1.00	3.00	160	0.00	200	12658.00000	100	0.00	
26	Index	1.00	3.00	160	0.00	200	12658.00000	100	0.00	

FIG. 2: SERGUI Screening Page

Inaugural ACA Fellows Announced

The new ACA fellows program recognizes a high level of excellence in scientific research, teaching, and professional duties, but also service, leadership, and personal engagement in the ACA and the broader world of crystallography and science. The ACA Fellows program celebrates the excellence of our members and promotes their recognition to world-wide constituencies such as employers, other scientific societies, and the government. ACA Fellows will serve as scientific ambassadors to the broader scientific community and the general public to advance science education, research, knowledge, interaction, and collaboration.

The Council proudly announced the inaugural class of ACA Fellows at the Awards Banquet held June 1, 2011 in New Orleans, La.

Inaugural ACA Fellows:

Helen Berman	Carroll Johnson
Phillip Coppens	Isabella Karle
Johann Deisenhofer	Jerome Karle
Bill Duax	Connie Rajnak
Judy Flippen Anderson	S. N. Rao
Jenny Glusker	Michael Rossman
Herbert Hauptman	George Sheldrick
Wayne Hendrickson	B. C. Wang

Congratulations to our SER-CAT Director, Prof. B. C. Wang, for this recognition of his high level of achievement in X-ray Crystallographic Science.

Selection of the MX300HS by Kathy Morris

In June of 2010, the NIH/NCRR awarded SER-CAT \$1.42 million for the purchase of a next generation, fast area detector for its 22ID beamline. Following news of the award, a committee was appointed by SER-CAT Director Dr. B. C. Wang to make a recommendation on the type of new detector most suitable for SER-CAT needs. The committee's members were Drs. Fred Dyda, CHAIRMAN (NIH), John Chrzas (SER-CAT), William Furey (University of Pittsburgh), John Rose (UGA), Gerold Rosenbaum (SER-CAT), and Ruslan (Nukri) Sanishvili (GM/CA).

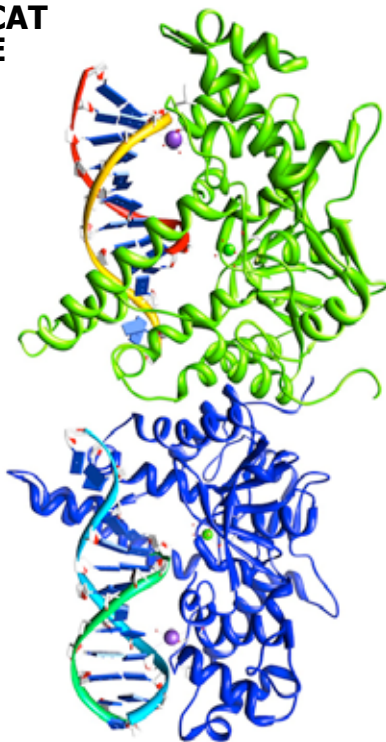
The Committee first began by establishing the necessary criteria to compare available detectors in the market. Two systems surfaced as the top candidates: the Rayonix MX300-HS CCD and the Dectris Pilatus 6M PAD. After further consideration of the variables available, the Committee's final recommendation was to follow the intent of the original proposal, i.e., purchase the Rayonix system. This decision further benefits SER-CAT by the fact that Rayonix is located very close to the Argonne National Laboratory.

RECENT SER-CAT STRUCTURE

PDB# 3QE9

BEESE GROUP
DUKE UNIVERSITY

Human exonuclease 1
DNA complexes



J. Orans, E.A. McSweeney, R.R. Iyer, M.A. Hast, H.W. Hellinga, P. Modrich, L.S. Beese
Reference: CELL, 145(2),212-23 (2011)

Rayonix MX300HS Ordered by John Chrzas

SER-CAT has ordered a Rayonix MX300HS detector for 22ID with an installation planned for late 2011. This purchase was funded by a \$1.42 million NIH Major Research Instrumentation grant and matching support of \$400,000. The new detector is based on a new split-frame transfer CCD (SFT-CCD) chip and offers data collection speeds up to 600 images per minute. This speed corresponds to a frame rate of 10 images per second in standard 2x2 binned mode (78 micron pixels). The new detector will also support both traditional and shutterless data collection.



The MX300HS is a faster version of the MAR MX300 detector currently installed on 22ID, and in addition to its speed, has a read noise one third (0.05 photons) that of the current MAR detector. Additionally, since the MX300HS is an integrating detector, it can, unlike current pixel array detectors, easily accommodate both traditional wide slice (0.5° - 1° images) or fine slice (0.05° - 0.3° images) data collection protocols. The wide slice data can be processed in a manner similar to that used for the current MAR detector. However, it is recommended that the fine slice data be processed using XDS or other data reduction programs that use 3D profile fitting.

The new system can provide a 180° data set with a modest 1° per second in 3 minutes (using current shuttered operation)! Once fully integrated into the SER-CAT beamline control environment, the new detector should allow the user to screen 240 crystals or 80 data sets (shuttered mode) in 4 hours of beam time! The screening mentioned above includes data indexing, determination of the crystal's mosaicity and generation of a data collection strategy.

The new detector should be in service in early 2012 and, once installed, the current 22ID MAR 300 detector will be moved to 22BM to increase its experimental envelope.

The SER-CAT Spectrum

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SER-CAT is governed by a 13 member executive board consisting of one representative from each membership state or organization. The representative states / organizations are Alabama, Florida, Georgia, Illinois, Kentucky, Missouri, North Carolina, Pennsylvania, South Carolina, Tennessee, Virginia, the NIH Intramural Research Program, and Industry.

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Florida State University

Georgia State University

Georgia Tech Research Corporation

Medical University of South Carolina

Monsanto Company

National Institutes of Health Intramural Research Program

North Carolina State University

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VISIT OUR WEBSITE AT
www.ser-cat.org

The *SER-CAT Spectrum* is the biannual newsletter of the SER-CAT group. Additional information about SER-CAT and the Advanced Photon Source at Argonne National Laboratory may be found at our website (www.ser-cat.org) or by contacting the SER-CAT Administrative Office at 706-542-3384.

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