

# National Research Program for Genomic Medicine

## Core Facility Project : D4

### Operation and Upgrade of the Synchrotron Radiation Protein Crystallography Facility (SPXF)

#### Progress Report (2008.09.01~2009.08.31)

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Institution: National Synchrotron Radiation Research Center

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#### ● Service Statistics:

Over the reporting period (2008/09/01~2009/08/31) the number of users accommodated at the SPXF has grown significantly up to 26 groups for BL13B1, and 20 groups for BL13C1. 173 PX experiments have been conducted and 1076 users have been trained. More than 85% BL13B1 beamtime and 80% BL13C1 beamtime are for user service which already exceeds the originally planned 65% service time and this number is expected to grow up in future. A table listed below documents this data by beamline, with the number of user groups, experiments, trained users, beamtime delivered hours, and percentage of beamtime used in user service.

Table1. Service Statistics

	# of Visit User Groups	# of Experiments	# of Trained Users	# of Beamtime Hours Delivered	% of Beamtime Used for Service
BL13B1 MAD-PX	84	119	606	3875	84.2% (35.9%)
BL13C1 Mono-PX	65	76	459	3682	80.5% (28.6%)
Sum	149	195	1065	7557	

( ) means the beamtime percentage used by NRPGM users.

#### ● Geographical Distribution of User Groups:

Because the beamline performance of BL13B1 is comparable to many famous PX beamlines in the world, many international users in Asia are interesting in using this facility. A table listed below shows the regional distribution of the user groups.

Table2. Regional Distribution of User Groups

	Domestic	International Users
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	Users				
	Taiwan	Japan	Singapore	Thailand	Hong Kong
# of User Groups	36	7	5	4	1

Nation	Affiliation
Taiwan, ROC	Academia Sinica Chung-Shan Medical University National Cheng-Kung University National Chiaji University National Chung-Hsing University National Defense Medical Center National Dong-Hwa University National Health Research Institutes National Ping Tung University of Education National Synchrotron Radiation Research Center National Taiwan University National Taipei University of Technology National Tsing-Hua University National Yang-Ming University Yuanpei University
Hong Kong	Hong Kong University
Japan	Hokkaido University Kyoto University Nagoya University Osaka University Photon Factory Yokohama City University
Singapore	Institute of Molecular and Cell Biology, A*STAR's Biomedical Sciences Institutes Nan-Yang Technological University National University of Singapore
Thailand	Mahidol University National Center for Genetic Engin. and Biotech. Suranaree University of Technology

● **Statistics of User Publications:**

The publication summary for reporting period (2008~2009/08/31) shows 64 SCI papers published (including Science, Proc Natl Acad Sci USA, J Am Chem Soc, Nucleic Acids Res, Plant Physiol, Mol Cell Biol, J Biol Chem, J Med Chem, J Mol Biol, J Virol., Biochem J, Proteins, PLoS One,...etc), 22 these completed and 73 conference abstract published for 159 total publications. All 159 publications represent service research where the SPXF provided facility and support only. Since SPXF is a 100% service-orientated core facility, there isn't any activity in R&D and Collaboration Research. Total 52 protein structures have been deposit to the Protein Data Bank (PDB) from SPXF beamlines. A table listed below provides a breakdown of the SCI papers in terms of total number and different Impact Factor (I.F.), conference abstracts, and PDB depositions.

Table3. Publications Statistics

	SCI Papers			Conference Abstracts	Theses	PDB Depositions
	Total	I.F. > 2	I.F. > 5.8			

2008	29	25	10	41	8	30
2009	35	32	13	32	14	22
Sum	64	57	23	73	22	52

- **Science Highlights**

#1 **A Cholesterol Biosynthesis Inhibitor Blocks *Staphylococcus aureus* Virulence:** Prof. Andrew H.-J. Wang, the vice-president of Academia Sinica, uses NSRRC protein crystallography facility to solve the structure of a key enzyme in synthesizing carotenoid pigment in golden yellow staphylococcus. This research clearly locates the position of ligand-binding pocket, which can accommodate two ligands at the same time. The development of more effective medicines aim at this enzyme can be expected soon. In addition, this protein structure provides important information toward designing new cholesterol-lowering medicines in the future. This publication is selected as the 2008 Research Highlight by US National Institutes of Health (NIH).

#2 **Crystal structure of the membrane-bound bifunctional transglycosylase PBP1b from *Escherichia coli*:** Dr. Che Alex Ma and Prof. Chi-Huey Wong, the president of Academia Sinica, presented the complete 3 dimensional model of the PBP1b protein on the surface of bacteria using NSRRC protein crystallography facility. This is the first time ever the mechanism of the enzyme that holds the key to bacterial cell wall formation is disclosed in details. They have successfully created an innovative approach which may very well be the platform to discover new antibiotics targeting "transglycosylase". The group has reported a perfect scheme for this rescue mission. This research has been reported in the Nature Review Drug Discovery as research highlight in 2008.

#3 **Human Tudor-SN in MicroRNA Binding and Degradation:** Human Tudor-SN is a multi-domain protein involved in many biological events, including transcription regulation, spliceosome assembly, translation repression, RNA interference and RNA editing. This interesting RNA-binding protein is also related to colon cancer and polycystic kidney disease. To investigate how Tudor-SN binds RNA and participates in RNA editing and interference, Professor H. S. Yuan determined the crystal structure of a truncated form of Tudor-SN, containing SN3, SN4, tudor and SN5 domains, at a resolution of 1.9 Å. The four domains of Tudor-SN are assembled into a crescent-shaped structure with a basic concave surface jointly formed by SN3 and SN4. The putative active sites of SN3 and SN4 are occupied by citrate ions. Professor Yuan also showed that the tandem repeats of SN domain are required for RNA binding and cleavage activities. In summary, this study suggests how Tudor-SN binds RNA and offers a testable model for further characterization of its diverse roles in RNA manipulation.

- **Performance Upgrade and Technological Development:**

Over the pass one year, many new functions have been developed and implemented to the core

facility, these include: (1) BL13B1 CCD detector has been upgraded from Q315 to Q315r, and BL13C1 CCD detector has been upgraded from Q210 to Q315r. The cost for this detector upgrade plan is about 1M USD and is funding by NSRRC. (2) The sample container of the Stanford Automated Mounting (SAM) system has been modified from SSRL cassette to Uni-puck which can be used in SAM system, Berkeley Automated Mounting (BAM) system and Photo Factory Automated Mounting (PAM) system. This change will allow users to have more flexibility in using different beamlines and different sample automounter. (3) An X-ray sensitive beam stopper has been developed and used to protect the expensive CCD area detector from exposure of the direct beam and to real time monitor the intensity fluctuation. (4) Implement the automatic XANES spectrum scanning and MAD energy determination function to the beamline control and data collection system. (5) Development an automatic sample lighting system. (6) Development an automatic sample annealing system.

- **User Training and Support:**

The best hardware and software are inefficient if well-trained support staffs are not available to assist the user, therefore, the training of the user support staff will be one of the most important issues to keep this facility running in the most efficient way. An 8 months training program provide by NSRRC seniors scientists is applied to every beginning support staff to ensure he/she has enough knowledge and skills to supervise different PX experiments. Currently, 7 well-trained support staffs are helping users conducting theirs experiments on the beamline daily. The support staffs provide 12 hours a day (8 hours for weekend), 6 days a week on-site user supports to help user collecting high-throughput data. All users have to take the beamline training before they can start their experiments at SPXF. A 2~4 hours on-site training tutorials is provided for each visiting research group to help how to condition the X-ray beam, get in/out the experimental hutch, mount and center the crystal, collect diffraction data, process diffraction data, and backup data. Over the reporting period (2008/09/01~2009/08/31) 1065 users have been trained and 195 experiments have been conducted. A facility web site (<http://bionsrrc.nsrcc.org.tw>) has been established. This web site is modified frequently and includes expanded user information. The user training and dissemination are also included on this web site. This web site is currently experiencing about 1000 hits per month and the user guide page is especially popular.

- **Facility Dissemination:**

The goal of facility dissemination is to disseminate experimental techniques of macromolecular crystallography to researchers or graduate student who has no previous experience but with an interest in using this specific method to further the scope of their research, and to introduce the latest developments in experimental methods to the user community to promote the usage of the facility. Dissemination activities include workshop for experienced users, training course for potential and novice users, and posters describing facility result and available services at conferences and workshops. In addition to posters, facility staff also manned a booth providing DM, and illustrating video film. Two workshops for experienced users and one training course for potential users were

held over the reporting period.

Activity Title	2009 Protein Crystallography Winter School
Sponsor/Venue	National Synchrotron Radiation Research Center
Date	2009/02/09-13
# of Tutors	11
# of Applicants	52 (restricted to novice user with a maximum of 14)
# of Participants	14 (1 Post Doc., 1 RA, 6 Ph.D. students, 6 MS students)
Web Page of Activity	<a href="http://bionsrrc.nsrcc.org.tw/training_open2.php?b_id=18&amp;bb_id=351">http://bionsrrc.nsrcc.org.tw/training_open2.php?b_id=18&amp;bb_id=351</a>

Activity Title	Mini-Lectures on Protein Crystallography
Sponsor	National Synchrotron Radiation Research Center and National Taiwan University
Venue	College of Life Science, National Taiwan University
Date	2009/03/5-6
# of Tutors	2
# of Applicants	91 (restricted with a maximum of 80)
# of Participants	80
Web Page of Activity	<a href="http://bionsrrc.nsrcc.org.tw/workshop1.php?b_id=3&amp;bb_id=74">http://bionsrrc.nsrcc.org.tw/workshop1.php?b_id=3&amp;bb_id=74</a>

Activity Title	2009 Software Practical Course on S-SAD Phasing
Sponsor/Venue	National Synchrotron Radiation Research Center
Date	2009/03/11~13
# of Tutors	10
# of Applicants	42 (restricted to novice user with a maximum of 42)
# of Participants	42 (1 Prof., 13 Post Doc., 9 RA, 7 Ph.D. students, 11 MS students, 1 BS student)
Web Page of Activity	<a href="http://bionsrrc.nsrcc.org.tw/training_open2.php?b_id=39&amp;bb_id=376">http://bionsrrc.nsrcc.org.tw/training_open2.php?b_id=39&amp;bb_id=376</a>

Activity Title	2009 Protein Crystallography Training Course
Sponsor/Venue	National Synchrotron Radiation Research Center
Date	2009/07/6-10
# of Tutors	11
# of Applicants	72 (restricted to potential user with a maximum of 18)
# of Participants	18 (4 Prof., 5 Post Doc., 1 RA, 8 Ph.D. students)
Web Page of Activity	<a href="http://bionsrrc.nsrcc.org.tw/training_open2.php?b_id=41&amp;bb_id=389">http://bionsrrc.nsrcc.org.tw/training_open2.php?b_id=41&amp;bb_id=389</a>

● **Committee Activities:**

The 2008-2<sup>nd</sup> User's Committee Meeting is held at National Synchrotron Radiation Research Center on Oct. 9<sup>th</sup> 2009, 11 of the 13-committee members attended this meeting. The committee recommended that since the responses of trainees of training course for potential user are very positive, the core staff should consider a training school for novice user(Activity web page: [http://bionsrrc.nsrcc.org.tw/research.php?s\\_id=9](http://bionsrrc.nsrcc.org.tw/research.php?s_id=9) ).

The 2009-1<sup>st</sup> User's Committee Meeting is held at Institute of Molecular Biology, Academia Sinica on April 3<sup>rd</sup> 2009, 11 of the 13-committee members attended this meeting. The committee recommended that since the user group growth so rapidly that result in the average beamtime for each user group is not enough for current research needs. D4 core should seek for new funding to expand the capacity of current facility (Activity web page: [http://bionsrrc.nsrcc.org.tw/research.php?s\\_id=9](http://bionsrrc.nsrcc.org.tw/research.php?s_id=9) ).