Data acquisition system for macromolecular crystallography (MX) at SPring-8

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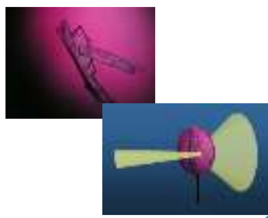
September 25th, 2014 at 10th NOBUGS

Target of SPring-8 MX beamlines

Micro crystallography (BL32XU)

Sample size < 10um

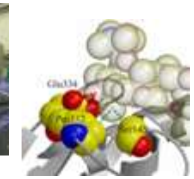
- Micro-focus beam for micro-Crystals
- Support for Micro-Crystal handling



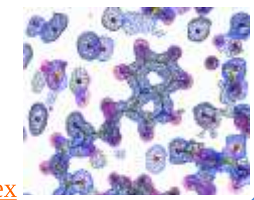
High-precision data collection (BL41XU)

Sample size > 10um

- High-flux beam
- Sub-atomic resolution



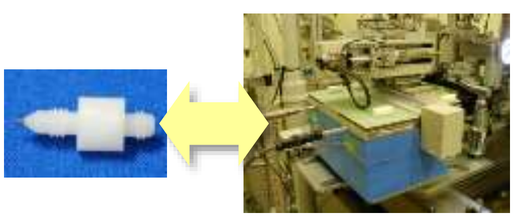
Substrate Complex



Visualization of H-atom

High-throughput & Routine MX (BL26s, BL38B1 & BL12B2)

- Automatic data collection
- Mail-in & Remote Data Collection



Sample size > 50um

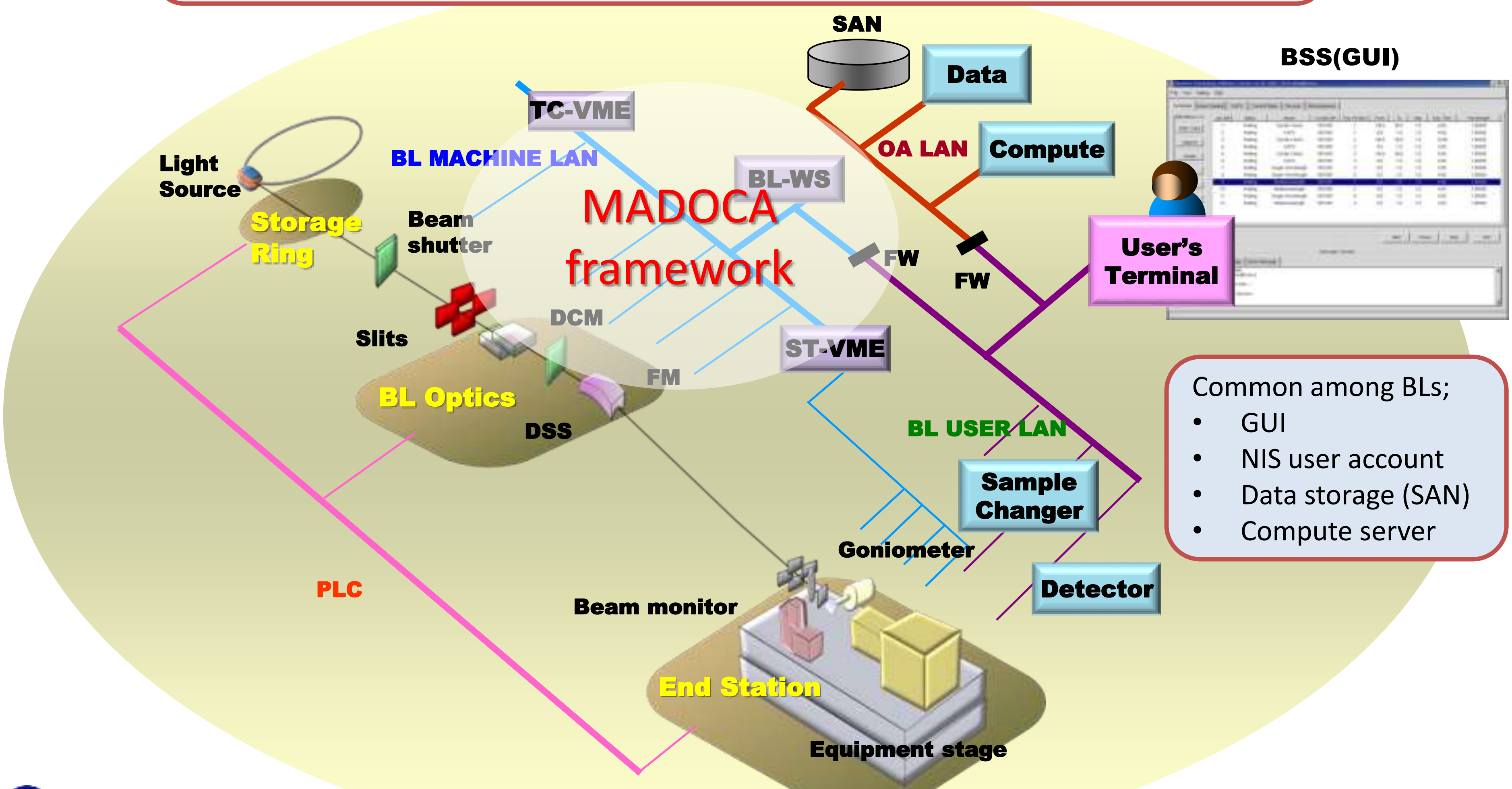
Large Molecular Complex (BL44XU)

- Parallel Beam for Large Unit Cell (>500Å)

P2 station for Virus

Control system for SPring-8 MX beamlines

- **Unified beamline control with a GUI under C/S architecture**
- **Common storage & compute server on the network**
- **SR, BL, end station are seamlessly controlled via MADOCA**

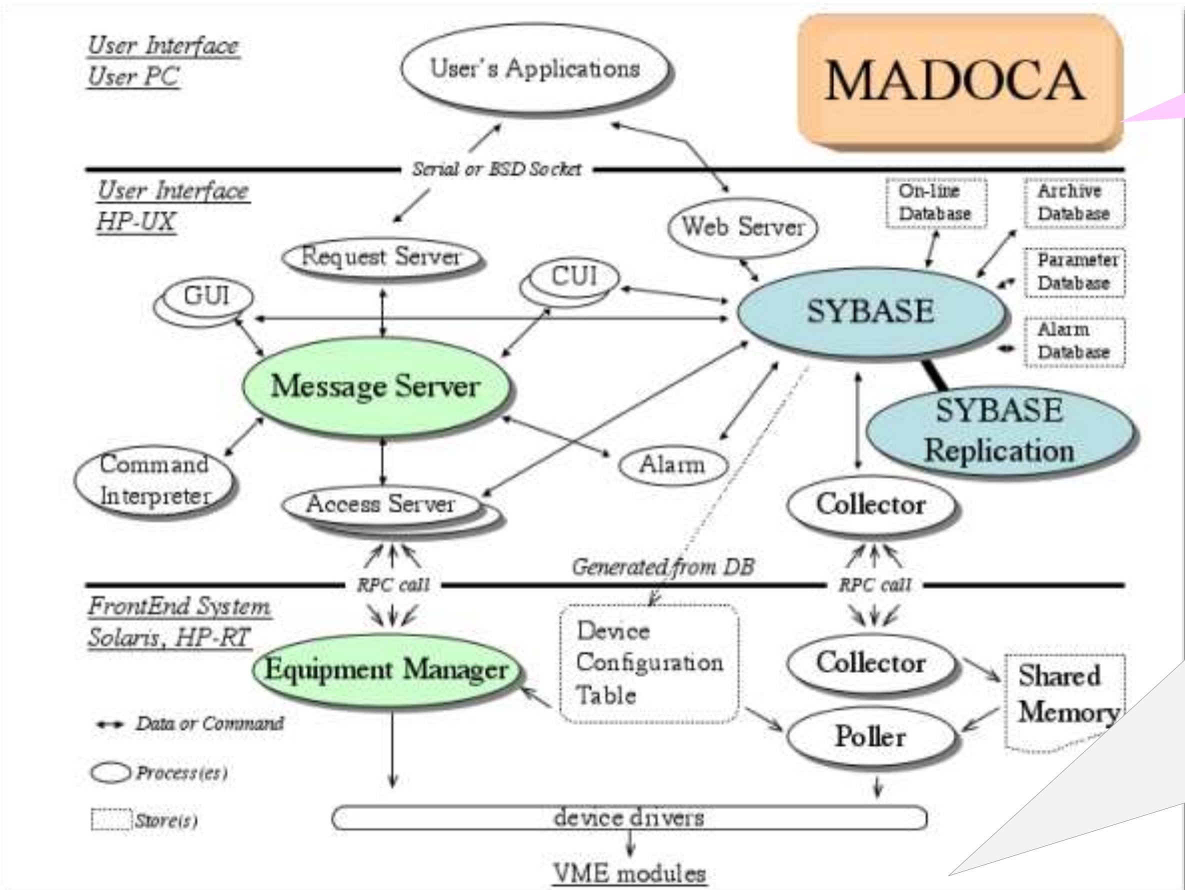


Common among BLs;

- GUI
- NIS user account
- Data storage (SAN)
- Compute server

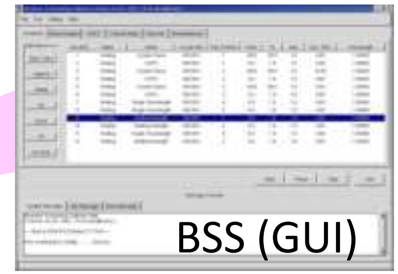
MADOCA (Message And Database Oriented Control Architecture)

A control framework for accelerator, storage ring and beamline control developed by JASRI Controls & Computing Division at SPring-8.

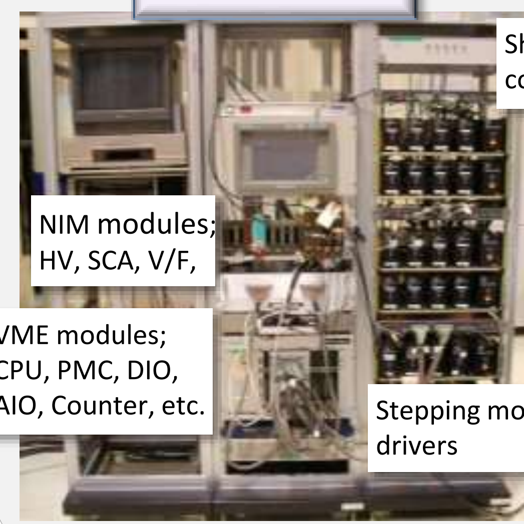


R. Tanaka, et al., Proc of ICALEPCS '95, (1995)

Socket connection



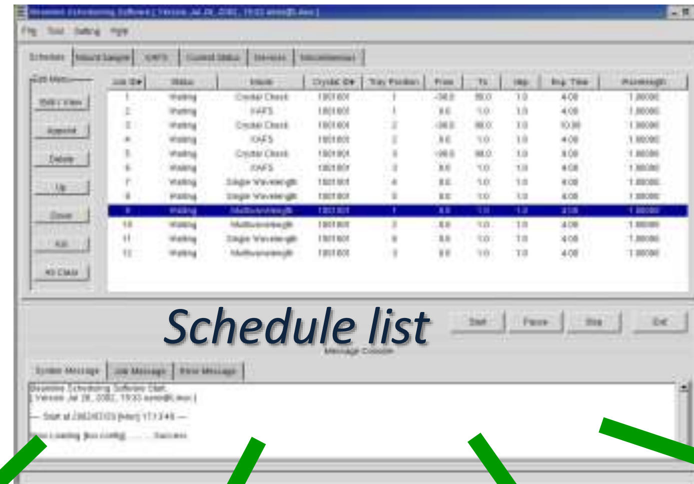
BL26B2 ST-VME



BSS (Beamline Scheduling Software)

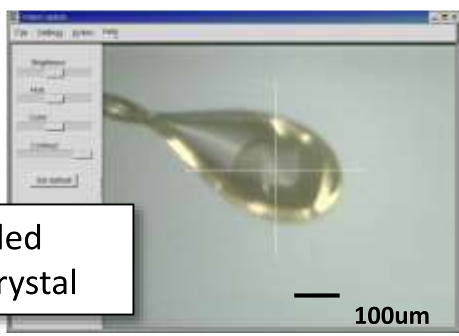
Standard GUI for all MX beamlines at SPring-8

- All-in-one control
- Job list for multiple conditions
- Load text BL configuration file
- Language & library:
  C, GTK+2, OpenGL, V4L2
- Platform: Linux



Schedule list

Diffraction measurement



Cryo-cooled protein crystal

Sample Exchange, Centering

Exp. Conditions

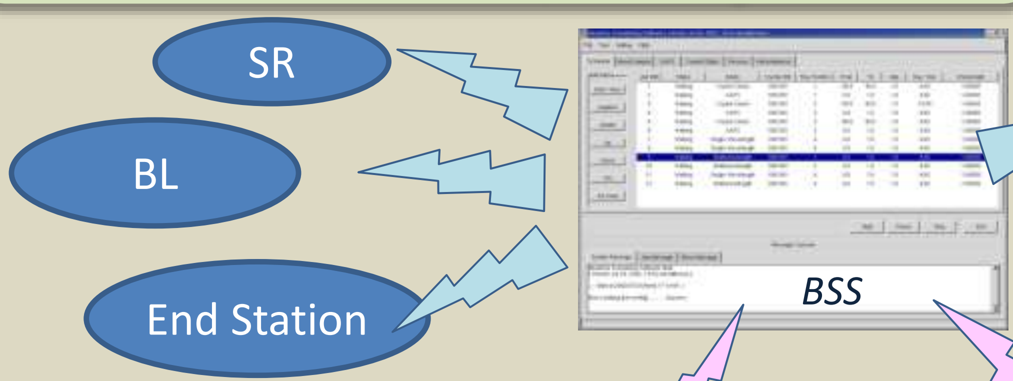
XAFS measurement

G. Ueno et al., J. Synchrotron Rad. (2005). 12, 380-384

Routine crystallography at SPring-8

Automatic data collection

- Sample Exchange, centering
- Set wavelength, beam optimization
- Detector distance, goniometer settings
- Diffraction data collection, monitoring SR status



Sample Changer SPACE at BL26B1

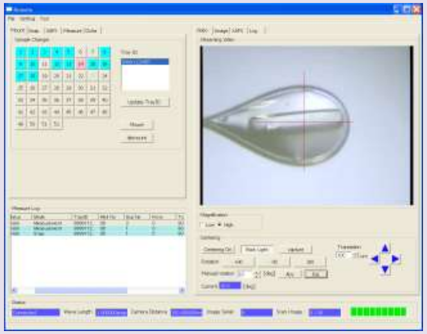
Murakami et al., J. Appl. Cryst. (2012)

Available pin types

Remote data collection

D-Cha

- Web-base interface
- Language: Perl
- Database: PostgreSQL
- **Mail-in data collection**



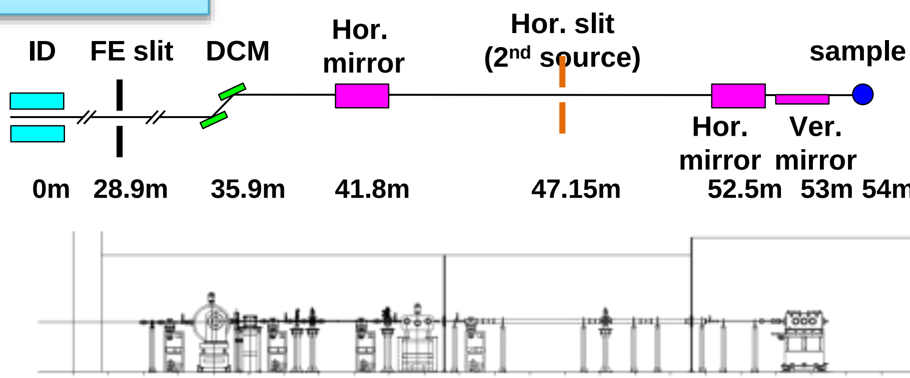
SP8Remote

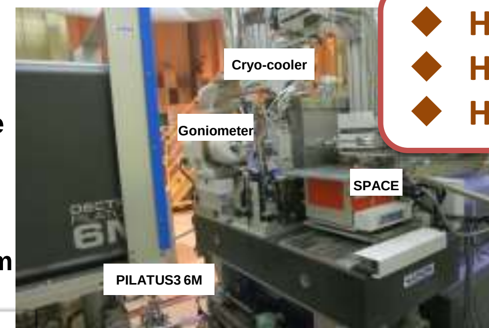
- Platform: Windows
- Language: Python
- GUI TK: wxPython
- **Remote control**

Okazaki et al., J. Synchrotron Rad. (2008)

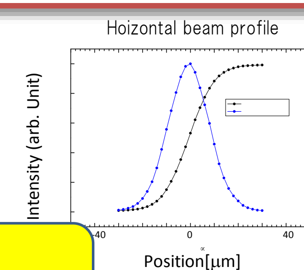
Micro-crystallography at SPring-8

BL41XU



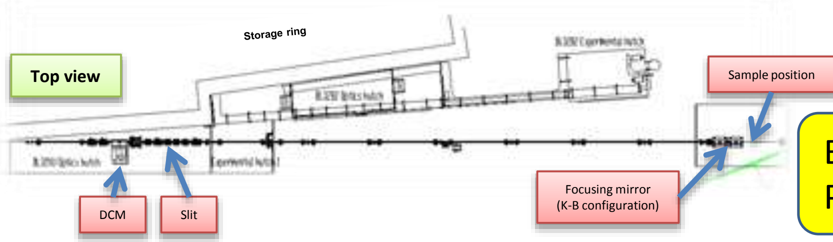


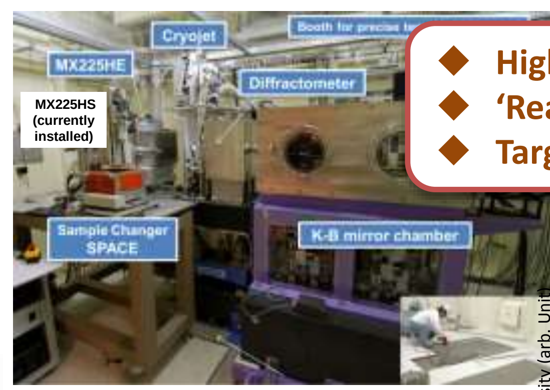
- Highest photon flux
- High speed data collection
- High precision data collection



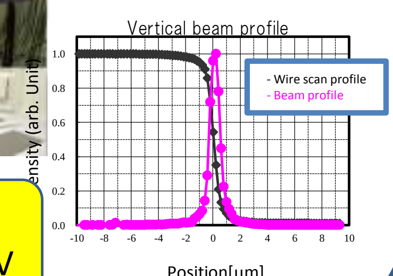
Beam size: 12x20 um
Photon flux: 1×10^{13} phs/sec@12keV

BL32XU





- Highly-brilliant beam
- 'Real' micro beam
- Target sample size < 10um



Beam size: 1x1 um
Photon flux: 2×10^{12} phs/sec@12keV

Data collections with ingenuity

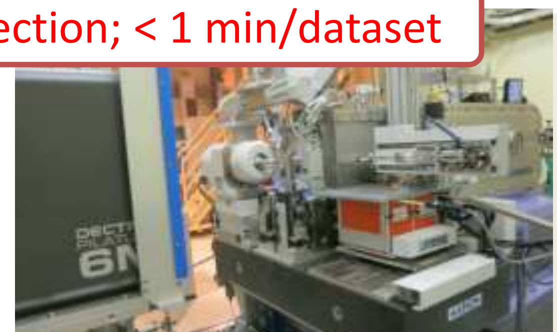
Shutterless data collection for helical scan and rapid raster scan

Helical data collection

Fast data collection; < 1 min/dataset

Cryo-cooled needle-like crystal

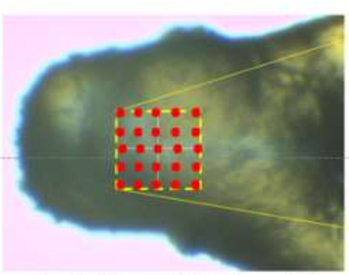
3D translation
& Rotation
+
High speed detector
with external trigger



Pilatus3 6M at BL41XU
100 Hz for 0.172mm 6M pixels.

Raster scan

Quick search of invisible samples



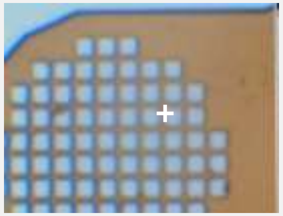
Crystal loop with LCP crystals

2D translation
+
High speed detector
with external trigger

MX225HS at BL32XU.
10Hz for 0.08mm 8M pixels.
Faster by binning.

Demo
W-needle (1um-point)

- Beam at white cross
- Cross size: 20um



Demo
Empty mesh (30um stp)

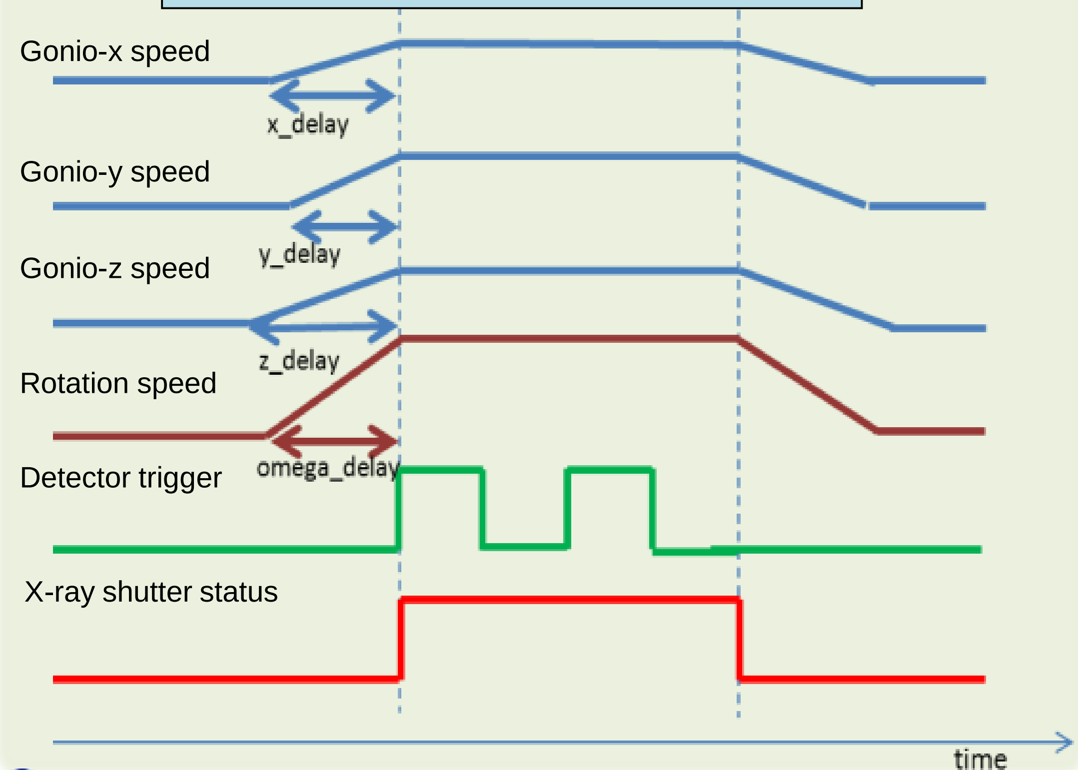
Synchronization by *Blanc8* control unit

- A multi-functional control unit developed at SPring-8
- Com express mother board with SATA connector
- Risercard for PCIe and PCI slots
- I/F for VGA, USB, GbE, RS-232C

Timing jitter ~ 2msec

Positional error < 1um
(where frame rate < 50Hz,
tr. Speed < 500um/s)

Timing chart of hardware control



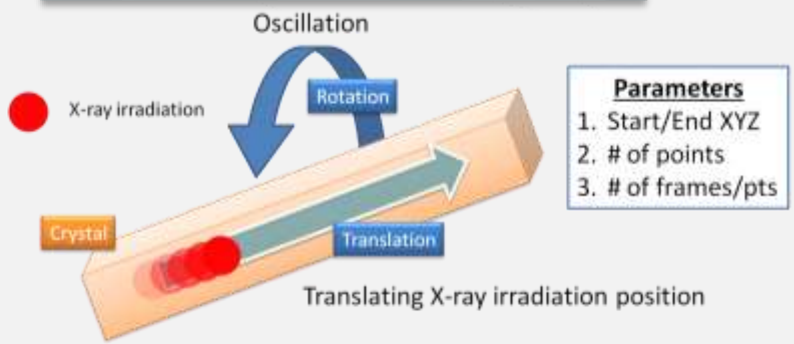
Blanc8 at BL26B1 end station

Ishii, M., *et al., Proc of ICALEPCS2009,* (2009)

Support for data collection strategy

Dedicated software for micro-crystallography to overcome Radiation Damage

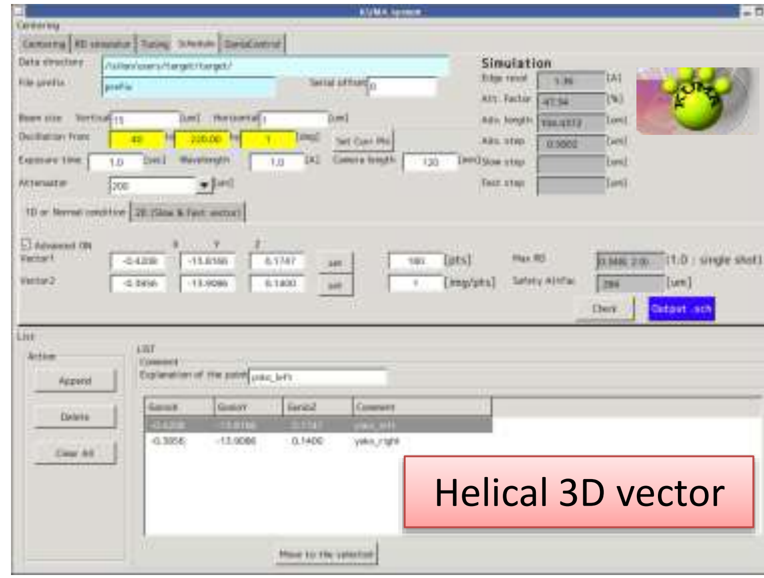
Helical data collection



Flot, D., *et al* (2010) *JSR*, 17, 107-118

KUMA

(Kesshou-wo Ugokashitari Mawashitari-suru Application)



Hirata, et al., J. Phys.: Conf. Ser. 425 (2013)

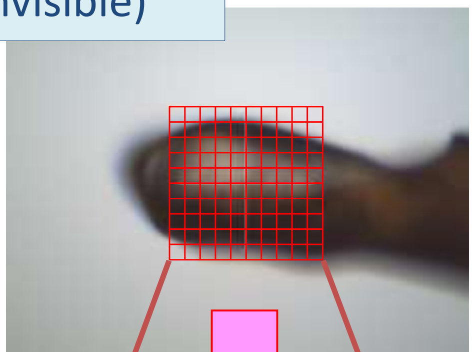
- Language: Python
- GUI TK: wxPython

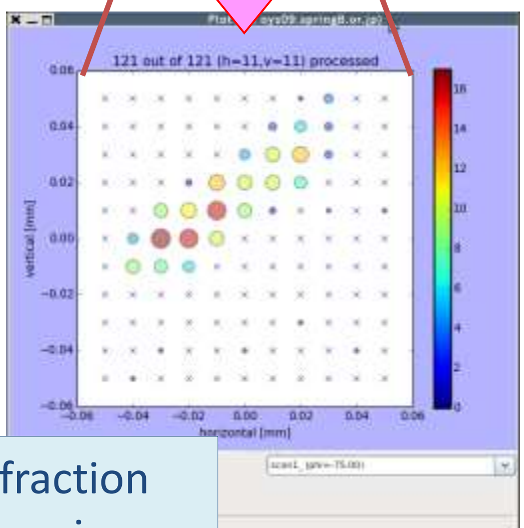
1. **Set up 'helical' vector**
2. **Determine oscillation conditions (oscillation width/frames)**
3. ***Exposure conditions for 'safety' data collection against RD.***

Important! Radiation damage by 1um beam propergates more than 2um!

‘Real-time’ visualization of raster scan results

LCP crystals (invisible)



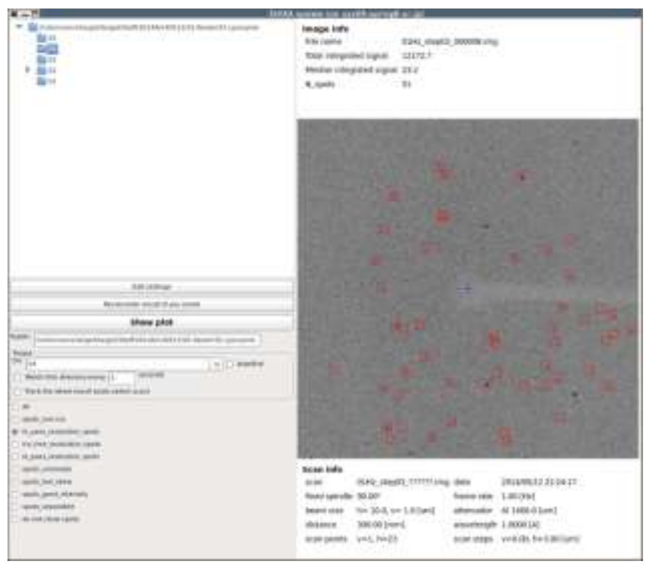


Diffraction mapping

SHIKA

(Spot-wo Hirotte Ichiwo Kimeru Application)

- Analyze low angle area of diffraction image (5-30Å)
- Back ground estimation for each 50x50pixels
- Find diffraction spots criteria; I/sigma > threshold
- Score an image by # of spots or total integrated intensity.



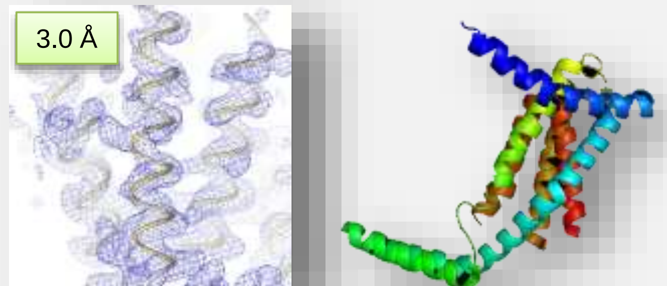
- Parallel processing with compute server (24 cores, Xeon)
  ~30 frames/sec
  (225HS 8x8 bin)
- Language: Python, C++
- GUI TK: wxPython
- Developed based on *DISTL (Sauter, 2013)*

Examples of structures determined at BL32XU

Channelrhodopsin (MAD)

H.E. Kato et.al., Nature, 482, 369-374 (2012)

YidC(membrane protein insertion) (SAD)

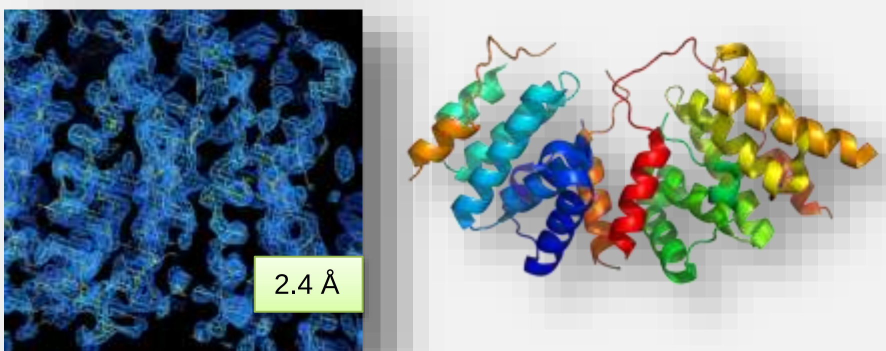


K. Kumazaki et al. Nature(2014)

MATE multidrug transporter

Y. Tanaka et al., Nature (2013)

H^+/Ca^{2+} exchanger (SAD)



T. Nishizawa et al. Science (2013)

Examples of de-novo structure determination

Protein	Crystal size	# of crystal (phasing)	HA
Channelrhodopsin	10 x 30 x 150 um	1	Hg
MATE	10 x 20 x 30 um	1	Se
CAX	10 x 10 x 80 um	1	Hg
YidC	10 x 10 x 10 um	1	Hg
Claudin	10 x 10 x 80 um	1	Se

Structural Biology at SACLA

- Japanese first XFEL facility at SPring-8 site.
- X-ray pulse laser with 10 fs duration.
- Repetition rate 60Hz.
- Accelerator and beamline device control by MADOCA.

Applications

CXDI

MX

SFX

http://sfxproject.riken.jp/about.html

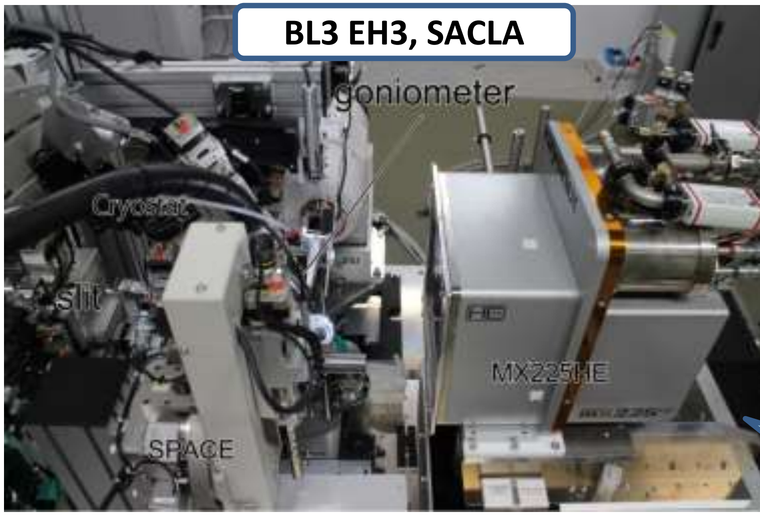
etc.

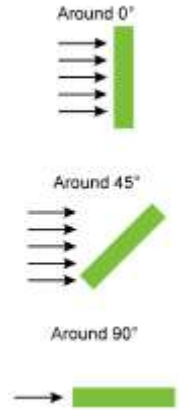
A femtosecond XFEL pulse outruns radiation damage!

Time scale of radiation damage

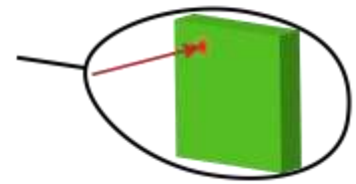
- Primary damage
  - Photoelectric effect
  - ~ sub-femtosecond
- Secondary damage
  - Generation of reactive particles
  - ~ picosecond

Goniometer based diffractometer for Step scan data collection at SACLA

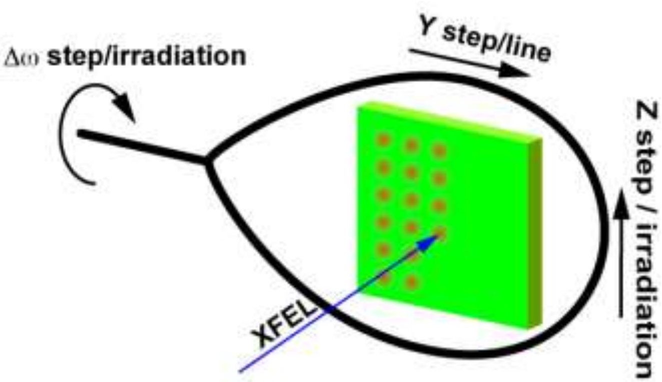


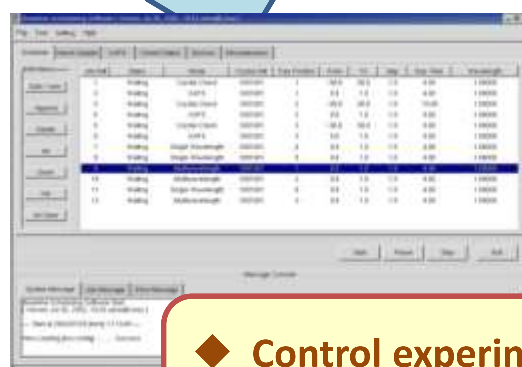


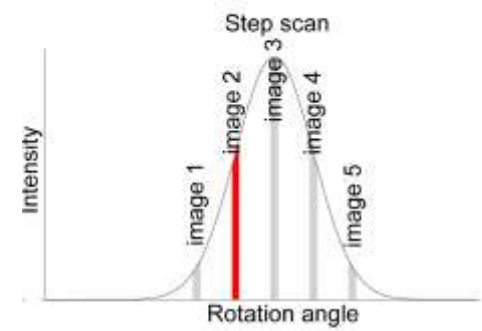
Beam paths are located 50 μm apart.



Δω ≤ one-third of crystal mosaicity





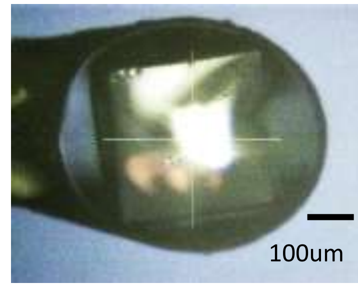


- Control experiment by BSS through MADOCA
- Scheduling list of irradiation points.

1.9Å damage-less structure of bovine Cytochrome c Oxidase

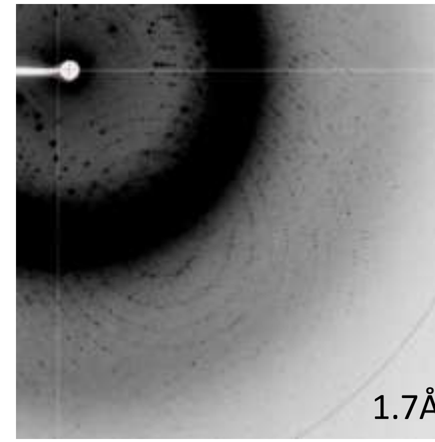
Hirata, K., Shinzawa, K. *et al.*, *Nature Methods* (2014).

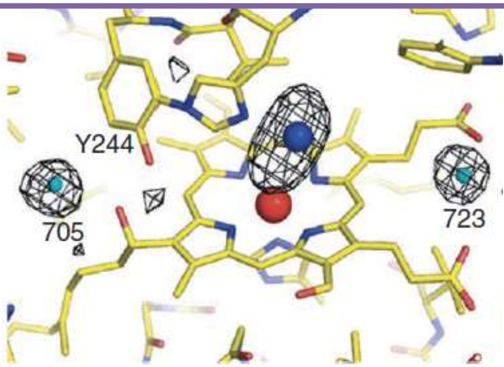
Data collection:	BL3-EH3, SACLA
Photon energy:	10 keV
Pulse duration:	< 10 fs
Pulse photon:	3.5×10^{10}
# crystals used:	76
# images collected:	1396 (1107 processed)
Rot. Step:	0.1˚

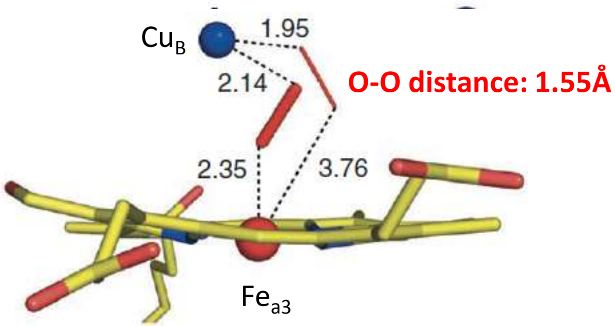


CcO crystal
M.W. 420kDa
Cell volume 6.7MÅ³

Diffraction image







Comparison of peak heights of water molecules

Intensity data	Fully oxidized form			CN-bound oxidized
	SACLA	SPring-8 BL32XU	SPring-8 BL44XU	SPring-8 BL44XU
Resolution (Å)	27.33 - 1.90	48.38-1.90	40.00 - 1.95	40.00 - 2.00
R_{work} / R_{free}	0.195 / 0.230	0.178/0.205	0.181 / 0.208	0.191 / 0.218
Peak heights				
A*	3.4	4.2	4.9	3.3
705	9.0	7.8	7.1	6.3
723	9.0	11.6	8.6	11.7
Ratio†	0.38	0.43	0.62	0.37
O-O distance	1.55Å		1.70Å (Aoyama, H. *et al.* (2009) *PNAS*)	

Summary

- At SPring-8 all MX beamlines are operated with a standardized GUI and a control system.
  - routine crystallography,
  - micro-crystallography,
  - femtosecond crystallography at SACLA

- Further integration of BL control software (KUMA, SHIKA, Remote GUI, etc.) is desired.

- Further automation e.g. real-time processing for high-speed detector by upgrading SHIKA system etc. is planned.

Beamline staffs and system development collaborators

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