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Data Processing of X-Ray Diffraction Images



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Quick Operating Procedure for X-Ray Data Collection

Procedure for Starting X-Ray Generator and Cryo Devices

This is Quick Start and Stop Guide. For detailed procedure please consult the Standard Operating Procedure (SOP).

X-Ray Facility (XRF) users are **required** to take the Radiation Safety Course offered by FSU Safety Office and **should be trained** by the XRF Director prior to using the instrument.

1. All personnel should use a radiation badge during the operation of x-ray data collection.
2. **Turn** the valve in the building chilled-water inlet to 'OPEN' position.
3. **Turn** the switch on the chilled water circulator.
4. **Press** the 'ON' switch in Haskris water chiller.
5. **Enter** the value of hour meter reading on the log book.
6. **Turn** the power X-Ray Power 'ON'.
7. **Switch** the ON button on the R-Axis IV++ vacuum pump.
8. **Switch** the ON button on the R-Axis IV++ Controller (Blue Box).
9. **Turn** the switch to ON in AD 41 Dry Air Unit and ensure that the flow meter shows ~13 LPM of air flow
10. **Turn** the switch to ON in Oxford Cryo Stream Controller located on the back of the unit.
11. Confirm that there is enough liquid nitrogen in cryo dewar.
12. Press the Blue Start button.
13. **Ensure** t enough helium gas is available on the Primary or the Reserve gas cylinders.
14. **Ensure** that the flow selection knob on the Flow Control Box is set to 'SLOW PURGE' mode, the flow indicator is at 35 and helium is flowing to Osmic mirror system. **NOTE: Failure to flow helium will irreversibly damage the mirrors and is extremely costly to fix, so ensure the flow.**
15. **Press** the 'START' button in the 'VACUUM' Control Panel.
16. **Press** the 'ON' switch in the 'X-RAY' panel.
17. Wait until the digital multi meter's reading to fall below 180mV. Then **press** the White 'ON' button located just below TARGET.



X-Rays are ionizing radiation and are harmful to humans. This equipment produces X-Rays when energized. So the users should follow recommended protocols. If you are in doubt always consult the X-Ray Facility Director before using it.

18. The target will start rotating. Simultaneously a green light in the Lamp Post will light-up.
19. Press the White 'ON' button below X-RAY. Red light below X-RAY panel and on Lamp Post will light-up.
20. TUBE VOLTAGE LED will increase to 20 TUBE CURRENT LED will increase to 10.
21. Increase the voltage by six (6) kV/ minute. **Increase** the current by ten (10) mA/ minute. Repeat these steps until desired or the maximum allowable power is reached.
22. The maximum load for a 0.3 x 3.0 mm² filament is 5.4 kW and **should never be crossed**. However, **44kv and 94 mA** are recommended for trouble-free operation.
23. To carry out an experiment the right-side port of the generator should be opened. Opening and closing of the x-ray generator shutter controlled by data collection computer and therefore the shutter switch on the x-ray generator should to left at "EXT" position.
24. The amber light at the Lamp Post will also light-up when RIGHT x-ray port is open.

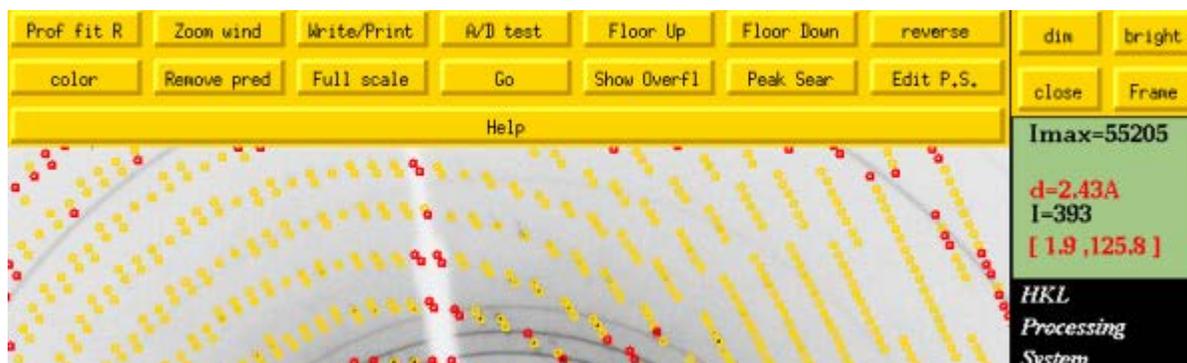
25. Proceed with your experiment.
 - **IN AN URGENCY:** During the experiment for emergency need to shut down the generator, push the Red Round Button labeled “**EMERGENCY**” located between the X-RAY and VACUUM Panels.
 - **IN AN EMERGENCY:** If the user is unable to reach the generator but needs to shut down the system, please push the Red Circular Button labeled “**EMERGENCY STOP**” located near the entrance to the Facility. Please call Thayumana Somasundaram following any emergency situation. The telephone number is located on x-ray facility interior doors.

Procedure for Stopping X-Ray Generator and Cryo Devices

26. Complete the experiment. Reduce the TUBE VOLTAGE and the CURRENT in steps until they reach 20 kV and 10 mA, Press the X-RAY ‘OFF’ red button located in X-RAY panel.
27. Enter the final value of hour meter in the log book.
28. Press the red STOP button in the VACUUM panel.
29. Turn the knob in Helium Control Box to ‘Stop’ position.
30. Flip the red handle to ‘OFF’ position in the circuit breaker at the wall.
31. Wait for 15 min for the target to cool down and switch OFF the Haskris water chiller.
32. Turn the black knob from ‘HAND’ to ‘OFF’ position on the water-chiller circuit-breaker.
33. Press the Red Stop button in Oxford Cryo Cooler and switch off AD 41 Dry Air Unit.
34. Switch OFF Vacuum Pump and R-Axis Controller.
35. Close the chilled water valve.
36. Return the Radiation Finger/Body badge back to the storage box at the Facility.
37. Report any problems or concerns to [Thayumanasamy Somasundaram](#).

Data Processing of X-Ray Diffraction Images

Indexing, integration, merging, and scaling of images using HKL Suite



Introduction to HKL Suite

In this module we will learn how to process the X-ray diffraction data (images) we collected for the lysozyme crystals. We will cover simple data processing using HKL Suite of programs: Denzo, XdisplayF, and Scalepack [Ref: Z. Otwinowski and W. Minor, " **Processing of X-ray Diffraction Data Collected in Oscillation Mode** ", *Methods in Enzymology*, Volume 276: Macromolecular Crystallography, part A, p.307-326, 1997, C.W. Carter, Jr. & R. M. Sweet, Eds., [Academic Press](#) (New York)]. We will, however, not show procedures for processing difficult crystals (twinned, high-mosaicity, and anisotropic crystals) in this tutorial.

First, it is important to gather data collection parameters like sample-detector distance, oscillation start angle, oscillation width angle, detector type, beam center, and wavelength while or immediately after collecting data. These values are necessary before we begin the data processing. At home, we use Rigaku RU-H2R/H3R Copper rotating anode generator equipped with Rigaku R-Axis IV⁺⁺ detector or marCCD165 detector, Rigaku Confocal mirror system, and Oxford CryoSystems Cryocooler. The wavelength of home-source data is fixed and is 1.541Å (0.1541 nm) and the sample is maintained at 100°K to reduce radiation damage. Typically, we use sample-detector distances in the range of 90-120mm. Beam center values for R-Axis IV⁺⁺ {x beam: 150.70, y beam: 149.80} and marCCD165 {x beam: 82.2, y beam: 81.7} are posted next to the computers that control these detectors.

R-Axis IV⁺⁺ detector is controlled by a Windows 7 64-bit computer while marCCD165 detector is controlled by a Red Hat Linux computer. However, the data *processing* is done on Ubuntu Linux machine (`gauss.sb.fsu.edu`). So after the end of the run, if you are using the R-Axis IV⁺⁺ detector transfer the acquired data using `sftp` (secure file transfer protocol) to `gauss`. However, if you are using marCCD 165 detector (and hence Linux o/s), the data will be mounted under `/disks/xrf/spruce/FSU-Data`. It is assumed that the user has some basic knowledge of Linux commands and operating system following the CRF Boot Camp given by Dr. Michael Zawrotny. XRF Boot Camp documentation on Basic Linux Computing is helpful too.