

# **PX Operator Training Manual**

## I. Introduction

Hello. This manual is primarily meant as a training tool for our PX operators, but should also be useful in part to any guest at one of our beamlines. This is by no means all you need to know, rather you should think of it as a bare minimum in order to do your job effectively. The goal of this manual is to provide a quick reference to common procedures, problems and solutions that you may encounter.

### 1. Overview

#### a. Overview of crystallography (le raison d'etre)

What is protein crystallography? Well, put simply, it is the study of protein crystal structure. In our case, the protein samples are cooled down to a temperature where crystallization occurs, so that we may use diffraction as a method of 'looking' at our samples. The following sections detail the basics, sparing the reader the gory mathematical details as much as possible, reducing the situation to (I hope) plain english. I strongly encourage the reader to continue their crystallographic education beyond this brief synopsis, and include the mathematical theory.

#### -synchrotron radiation

Protein samples are small. Very small. They are so small, in fact, that the resolution needed to get a 'picture' of their structure is on the order of 1 angstrom. The way we achieve this resolution is through x-ray light, which has a wavelength comparable to the protein substructures we are interested in. How do we get x-rays? We use a device called a 'synchrotron'. What a synchrotron does, is get electrons moving at velocities very near the speed of light, approximately 2800MeV (1eV is the energy necessary to move an electron through 1 volt of potential). The electrons are kept moving in the circular 'ring' by magnets. As they move around in a circle, they experience a centripetal acceleration.

A funny thing happens to electric charges when they experience acceleration. Accelerations cause their electric fields to 'kink' (think about it in terms of Faraday field lines!), with the kink moving outward at the speed of light. This kink contains the information that the charged particle has moved, and is called a 'light wave'. When the light wave interacts with some object, it collapses and is detected as a particle called a photon. The energy of the photon depends on the wavelength of the light wave, with shorter waves corresponding to higher energies. In the case of our synchrotron, these waves are of very short wavelength, and are called "x-rays". They spray out tangentially to the 'ring', and get caught in our beamlines. Our neighbor, the UV ring, has a smaller curvature, and emits ultraviolet radiation.

A few important terms are associated with the synchrotron storage ring:

1. Beam Current,  $i$  is a definition of the total number of electrons circulating in the ring (usually in milliamperes).
2. The Energy,  $E$  of each circulating electron (in MeV) - Energy is often defined by the means of  $\gamma$  parameters. The ratio between  $E$  and the electron rest energy is :

$$\gamma = E/m_0c^2 = [1 - (v/c)^2]^{-1/2}$$

where  $v$  is the speed of the electrons.

#### -Bragg scattering

Ok, so now we know where the x-rays come from. How do they tell us what the protein looks like? Well, we make use of a phenomenon known as diffraction. Diffraction of light occurs when the inter-atomic distances are comparable to the wavelength of the light source. What happens is, the light comes into the crystal, and gets reflected off of the crystal planes. Simple enough, right? But wait, there's more! As the light waves are reflected off of neighboring planes, a funny thing happens to them... they (the waves) interfere with each other. Depending on where your detector is, you will see either constructive or destructive interference. Constructive interference will manifest itself on your detector as a bright spot, while destructive interference will look like... well... nothing at all!

#### -reciporical lattice/Fourier transform

As you may have guessed, we can use these dark and light spots to reconstruct the protein's molecular structure. We recognize that our 'diffraction pattern', which we see on the detector is indeed the 'Fourier transform' of the reciporical lattice corresponding to the atomic planes we scattered our x-rays off of. What the heck is a reciporical lattice? Well, simply put, it's a mathematical representation of the crystal structure, related to the actual lattice by a series of cross products. Ok, so now what the heck is a Fourier transform? It's a mathematical method of representing waveforms. Again, I encourage the reader to further explore this subject, not just because it's your job to, but because it's also really cool! Have you ever used a laser-pen with one of those

funny caps that make the laser-dot look like some kind of picture? I have one that looks like a mouse, and my cat loves to chase it around the house. Anyway, what the cap has, is the Fourier transform of the picture!

### -phase problem (MAD)

Unfortunately, it's not quite enough to measure intensity information. Included in the Fourier analysis are several quantities of interest. One is the electron density, and another is the structure factor. The problem arises because the structure factor is complex (meaning the imaginary number,  $i$ ), and we define intensity as its square. Thus, we lose phase information, which we need to complete our Fourier synthesis (make up the molecular structure).

To retrieve the phase information, we make use of a technique called Multi-wavelength Anomalous Diffraction. In a MAD experiment, one adds a heavy atom to the crystal. There will now be a resonance between the heavy atom and the x-rays, which will cause a phase (and amplitude!) shift. What this should do, is break the symmetry between the front and back of the Bragg planes. From this broken symmetry, we may retrieve the phase information if we collect the same data set at two different wavelengths.

In practice, we generally use three wavelengths. The heavy atom's reflection peak, its inflection point, and some 'remote' or longer wavelength than the other two. To do this, we make use of our fluorescence detectors.

### -More information

For more detailed information regarding crystallography, please visit [Introduction to Crystallography](#), a tutorial written by Bob Sweet.

### b. Who's who

At first, it may be difficult to keep everyone and their functions straight. The approximate breakdown is as follows:

#### ADMIN

Denise Kranz  
Renie Rosati

#### BEAMLINE SCIENTISTS (PX)

X8c- LiWei Heung  
X12b- Deiter Schneider  
X12c- Bob Sweet, Anand Saxena  
X25- Lonny Berman, Mike Becker  
X26c- Annie Heroux  
FedEx- Howard Robinson

#### BEAMLINE TECHNICIANS

X8c- Leon Flaks  
X12b- Tom Langdon  
X12c- Sal Scalfani  
X25- Steve LaMarra  
X26c- Joe Mansury

#### SUPPORT

Systems Administrator- Matt Cowan  
Programming Guru- John Skinner

#### PX OPERATORS

Bill Nolan  
Shai Vaday  
Joe Mansury  
Stu Myers

### c. PX OP jobs and responsibilities (simple list)

Your job, should you decide to accept it, includes (but is not limited to) the following:

- Aiding our users in the execution of their experiments.
- Performing Fed-Ex experiments under the direction of Howard Robinson.
- Troubleshooting any problems that may arise during your shift.
- Maintaining the px-operator logbook.
- Aiding any of the above personnel the execution of their jobs.
- Keeping the LN<sub>2</sub> dewars full.
- Being a bright and cheerful representative of BNL Biology!

## 2. Computers (hardware)

- Layout (general, with diagram)
- Passwords

During the course of your job, you will have to know many (about two dozen!) passwords. My suggestion if you lack a photographic memory, is to write all your passwords down on a piece of paper, and carry it around with you in your wallet. Of course they're not going in the manual!

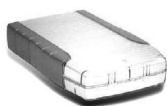
## c. Data Backup (general)

### 1. User Laptop

A number of experienced users of both the NSLS and other synchrotrons are finding that it's quite convenient to bring a laptop computer to back up data collected at the NSLS, and indeed, we are actively encouraging it. The storage of data to the user's own computer, which can be connected to the data-storage disks by 100 BaseT ethernet, eliminates operating system or filesystem compatibility issues, mount/umount/export complications, and problems regarding device availability or compatibility. The recommended archiving system is the use of ftp to store data to a laptop's external drive, such as a firewire system, or to a laptop's internal drive. We find that this scheme allows large amounts of data to be stored relatively quickly and easily on surprisingly inexpensive media.

[Instructions](#) for using DHCP to connect to BNL's network on a number of operating systems are available on the NSLS web-site.

A [web-based form](#) must be filled out on-line from the computer being connected within 30 minutes after the initial connection to prevent the network port from being deactivated. Please do NOT allow the users to plug their laptops into any old Ethernet cable!



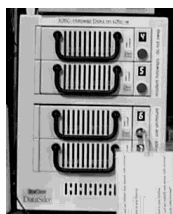
*Firewire drive.*

### 2. Tape Drives (types)

#### -Hotswap

These are removable, fragile, and expensive tape drives, which some users might bring to back up their data. Their operation is as follows:

1. Unmount the disk you wish to swap out. To do this, issue the UNIX command:  
`sudo umount/[diskname]`
2. Turn the key to unlock the disk. A light should begin blinking. Once the light stops blinking, you should see a "u" on the drive to denote "unmounted".
3. Pull out the old disk.
4. Put in the new disk.
5. Turn the key. Again the light should begin blinking. Once it stops, a number should appear denoting the hotswap drive number.
6. Mount the new disk. To do this, issue the UNIX command:  
`sudo mount/[diskname]`



*Hotswap drives at x26c.*

#### -8 or 4mm

These guys are somewhat easier to manage than the hotswaps. One thing to keep in mind is that the command lines differ a bit whether you are using the grey SGI machines (UNIX) or the black Dell (Linux) machines. Block size default is 20 for Linux, 1 for UNIX. The backup commands are as follows:

**UNIX:** `tar cvf/device location/drive name/your data directory`  
**Linux:** `tar cvb 20 -f/device location/drive name/your data directory`

If you want to confirm that a tape has been made, use the following commands:

**UNIX:** `tar tfv/device location`  
**Linux:** `tar tvb/device location`

Many of the drives are mirror drives, meaning that they can be used to make multiple copies of the data at once. To switch between mirror and independent modes, use the following procedure:

1. On the lower drive, press MENU twice
2. The screen reads "MODE=IND" press SELECT
3. Press MENU until the lower screen displays the mode you want.
4. Press SELECT. The screen should read "(CANCEL)"
5. Press MENU until the lower screen reads "(CHANGE)"
6. Press SELECT.

*Common Problems/Solutions:*

*Can't write tape.*

*Try: Cleaning with a cleaning tape.*

*Try: Check wire connections. If a connection is loose, tighten the cable and re-boot the computer. The computer MUST be re-booted to find the disk drive.*

*Try: Looking for the drive. On UNIX machines, use the command "hinv". On Linux machines, use the hardware browser (you will need the super-user password).*

*Try: Check to see that the drives are really called what you think they are called.*

*Try: Using one of the GUI programs on the desktop.*



8,4mm drives at x26c.

*The following sections describe beamline computers, which are in a state of re-working as of this writing. Check back for updates.*

- d. X8c
  - computer names, what they do, common problems/remedies
  - data backup hardware, what they do, common problems/remedies
  - restart procedures
- e. X12b
  - computer names, what they do, common problems/remedies
  - data backup hardware, what they do, common problems/remedies
  - restart procedures
- f. X12c
  - computer names, what they do, common problems/remedies
  - data backup hardware, what they do, common problems/remedies
  - restart procedures
- g. X25

-Starting a new user

Visitors to this beamline are all expected to use the same username: x12cuser. When starting a new user, create for them a directory (mkdir command) in an appropriate data disk (usually /w13/data1 or /w13/data2) and have them create their own subdirectories as needed, there.

-Computers at x25

### Common problems/solutions

Detector quadrants are blank/switched in position: This is most often a computer problem.

Try: Restarting cbass software.

Try: Rebooting the Q315 computers in the rack (see procedure under beamlines: general)

Diffractometer not responding:

Try: Cycling the power. Procedure:

1. Turn off optix, cbass if running
2. Turn off diffractometer (black switch on the back)
3. Turn on cbass, watch for it to have problems communicating to the diffractometer
4. Turn off cbass
5. Turn on the diffractometer
6. Restart cbass, optix

- data backup hardware, what they do, common problems/remedies

#### h. X26c

-computer names, what they do, common problems/remedies

-data backup hardware, what they do, common problems/remedies

### 3. Computers (software)

#### a. UNIX

-common commands (english to UNIX)

Most often in instruction books, you will find a list of UNIX commands followed by the english explanation of what the commands do. I find this about as transparent as a cashmere sweater. After all, If you knew the command already, you don't NEED its english explanation. Instead, I'll provide a list of the most common tasks you will perform, followed by their UNIX commands (as always in this manual, commands are printed in blue. Anything in [brackets] denotes a variable that you input).

-Print current directory: **pwd**

-list directory content: **ls**

-Change directory: **cd [newdirectory]**

-Print a list of all devices mounted to current machine: **df**

-Show a hardware inventory: **hinv**

-Remove something: **rm [something]**

-Print current user logged in: **whoami**

-Print all processes running: **ps -e**

-Print all ace (or whatever) processes running: **ps -e |grep ace**

-Print size of directory in kilobytes: **du -k**

-Connect to another computer: **telnet [computername]**

-Backup a directory to a tape drive: **tar cvf/device location/drive name/your data directory**

-Make a directory: **mkdir [directory]**

#### b. Linux

Linux is a shareware version of UNIX, created by a European fellow named Linus Torvalds. If you are one of those silly anarchist types who hates everything Bill Gates and Micro\$oft, you will be happy to learn that our Linux machines now out-number our Windows'.

In using Linux, the beginner will almost certainly stick to the "lindows" GUT's of Red Hat, which are far more user-friendly than the similar X-box GUT's on the SGI UNIX machines.

#### c. CBASS/optix

#### d. HKL2000/Denzo

#### e. X8c peculiarities

-firewall

-optix, shutter control

-Leon (just kidding)

### 4. Beamlines, general

#### a. Detectors (how they work)

1. Quantum 4: In this type of detector, we have four modules (quadrants) combined into an array. Each module contains a phosphor plate which, when hit by x-rays, emits a photon of lower (visible) wavelength. These emitted visible photons pass through a very cool, and VERY expensive optical taper, then end up by hitting 1" by 1" semiconductor CCD chips. The job of the CCD is to record an optical pulse as an electric one, digitized,

then fed into a computer system for processing. The CCD chips in the Quantum 4 have a resolution of 1150 by 1150 pixels. The total active area of the Q4 detector is 0.188m<sup>2</sup>.



2. Quantum 315: This detector type works very similarly to the Q4. Instead of a 2x2 array (four quadrants) however, the 315 sports a 3x3 array with a total active area of 0.315m<sup>2</sup>

Power-down procedure:

1. Quit optix and Cbass gracefully.
2. telnet w10
3. type: **ps auxww | grep det**
4. kill the process with "det" in title
5. type: **ps auxww | grep ccd**
6. kill the process with "ccd" in title
7. on Q315 PC rack, quit 'remote operate detector' on each of 5 PCs (need to press green or red buttons above to shift between PCs)
8. on Q315 PC rack, start 'operate detector' on each of 5 PCs (info will scroll; will read 'any\_fail: 0', or something like that, when each is ready)
9. on Q315 PC rack, PC #1, start 'quantum console' connect to detector enable temperature control ramp to 10 °C (It is critically important to RAMP the temperature. Simply setting it will break the very expensive CCD chips!!)
10. after they all reach 10 °C, quit 'quantum console' and quit 'operate detector' on all 5 PCs
11. go to power supply behind hutch. towards the bottom are 3 controllers; each has 2 buttons -- the upper one is the temperature control, and the lower one is the power. if we consider button #1 as being the lowest button on the lowest controller, and button # 6 as being the highest button on the highest controller, turn off buttons in the following order: 6, 4, 2, and then 5, 3, and 1. ie. turn off things top down, first the temperature control, and then the power. Unless there are explicit instructions, do NOT turn off the main power or mess with buttons on the top!!! (we want to avoid disasterous problems with the detector vacuum, etc.)
12. You are now able to switch fiber optic cables, etc., per instructions.

Power-up procedure:

1. go to power supply behind hutch. towards the bottom are 3 controllers; each has 2 buttons -- the upper one is the temperature control, and the lower one is the power. if we consider button #1 as being the lowest button on the lowest controller, and button # 6 as being the highest button on the highest controller, turn on buttons in the following order, waiting 5 seconds between pressing each button: 1, 3, 5, and then 2, 4, and 6. ie. turn on things bottom up, first the power, and then the temperature control. Unless there are explicit instructions, do NOT press the main power or mess with buttons on the top!!! (if the main power had been off, please see other documentation) (we want to avoid disasterous problems with the detector vacuum, etc.)
2. on Q315 PC rack, start 'operate detector' on each of 5 PCs (info will scroll; will read 'any\_fail: 0', or something like that, when each is ready) (need to press green or red buttons above to shift which PC)
3. on Q315 PC rack, PC #1, start 'quantum console' connect to detector enable temperature control set temp to 0.0° C when they all reach 0.0 (will read 'final ...'), ramp to cold operating temperature
4. when they all have reached their final temp. (about -45 °C), quit 'quantum console, and quit 'operate detector on all 5 PCs) (careful: do NOT set temp to cold operating; rather, ramp it; if it cools too fast, it could crack the chips)
5. on Q315 PC rack, start 'remote operate detector' on each of 5 PCs
6. start cbass; start optix; good luck ...

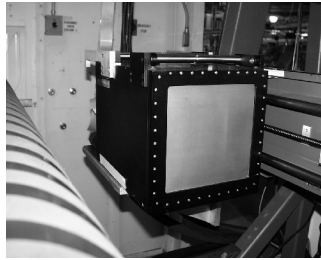
PC re-boot procedure (Q315-ADSC rack)

1. Quit 'remote operate detector' on each of the PC's you want to re-boot
2. Click the 'start' button on the taskbar, and follow instructions leading to a re-boot
3. After PC is rebooted, press ctrl-alt-del at the prompt to do so
4. Click on 'OK' when it asks you for a password (there isn't one)
5. Double-click 'remote operate computer' on each of the PC's re-booted
6. Restart cbass, optix, yadda yadda...



*Q315 PC rack at x25*

3. Brandeis 4: This is yet another CCD-type detector, and works similarly to the others. It is on X12c.

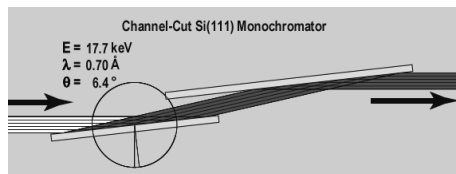


*Brandaís 4 detector at x12c*

-Flourescence detector

b. Monochromators

The beam coming out of the storage ring is called a white beam, it has many wavelengths. In crystallography we want to use a monochromatic light. So we use a monochromator to do just that. Our beamlines are equipped with crystal monochromators. A Platinum coated silicon crystal is used to diffract light from its 111 planes to produce a monochromatic x-ray of wavelength that is governed by the crystal's angle. This is based on Bragg's law.



*Graphic of a monochromator*

c. Mirrors



Mirrors are similar to the monochromators in that both are made of coated silicon crystals, and operate on the same principle. The mirrors are shaped like a section of a torus, and are movable on several axes to facilitate the focusing of the x-ray beam.

- d. Slits/attenuators
- e. Shutters

Shutters are exactly what they sound like. They are used to completely stop the x-ray beam. Typically a beamline will have three shutter groups. The first (counting from the ring down to the crystal) is the **Safety Shutter**. This shutter is operated by NSLS personnel, who will open it when all the safety approval red-tape is completed. The next is the **Photon Shutter**. This shutter is usually located downstream, somewhere close to the hutch. This is the shutter you open directly after the interlock procedure by pressing the green button on the hutch wall. When it is opened, you should hear a "whoosh" of air. The final shutter assembly is on the diffractometer, just upstream of the crystal. This is the shutter you open via the optix program, or through CBASS.

- f. Water cooling/proteus

The detectors make use of a water-cooling system, which may at times require servicing.

- g. Hutches  
-interlock procedure

Each beamline has an interlock procedure that must be followed before x-ray beam is allowed into the hutch. The process is long and cumbersome on purpose, as it is a safety feature. Each beamlines procedure is slightly different, but not enough so to go into. The general procedure is:

1. Press button for personal check station #1, located on a wall inside the hutch. A red light will go on.
2. Leave the hutch and close the door.
3. Remove the key from the door, and put it into the keyhole at check station #2.
4. Turn the key and press the button for check station #2. An alarm will sound for 20-30 seconds
5. When the alarm turns off, you may open the photon shutter, and proceed with the experiment.

-emergency stop

In the event that you find yourself trapped in the hutch, and you are concerned that someone is preparing to put x-rays in, you may press any of the red **EMERGENCY STOP** buttons that are strategically located throughout the hutch. This will result in one of two things:

1. If the Photon Shutter is closed, the Safety Shutter will close, and NSLS personnel will need to come and question you.
2. If the Photon Shutter is open, the electron beam in the ring will be dumped, NSLS personnel will need to come and question you, and you will be subjected to angry grumbles from the rest of the scientists on the experimental floor.

- h. Goniometers

Goniometers are XYZ adjustable platforms which are mounted on the diffractometers. Users will generally bring their own pins, which become mounted to the goniometer via a small magnet.



*XYZ adjustable goniometer*

-alignment, adjustment

Alignment of the goniometer is accomplished by adjusting the micrometer-scale screws with a special adjustment tool. Generally, we use a special alignment pin-head (instead of a needle) and center it within the field of a microscope camera.

-fluorescence paddle

A special paddle may be placed on a goniometer which, when struck by an x-ray beam, will fluoresce. Primarily, this technique is used to adjust the position of the horizontal lines displayed on the viewing screen, denoting x-ray position.

i. Diffractometers (see specifics for each beamline)

j. Beam Stops

The x-ray beam is quite intense, and will readily destroy the expensive CCD detectors at full strength. For this reason, it is important to employ a 'beamstop' which will block any direct x-rays from the collimator.

k. Nitrogen

-fill procedure

Follow the fill procedure as written on the wall at the NSLS fill station.

l. Coldstreams

Most of the beamlines use the Oxford Cryosystems cryostream cooler. The system consists of a cylindrical coldhead, an electronic temperature controller, a diaphragm air pump, a dry air unit, and a line dryer. Supplying the LN<sub>2</sub> which the system uses, is a small dewar (which holds about 1.5 day's supply of continuous use LN<sub>2</sub>) an electronic autofill controller, and a large supply dewar (which you will fill). It is very important to know that the coldhead and the temperature controller are a matched pair. The individual characteristics of the sensors within the coldhead are known by its specific controller. The serial number on the coldhead is on its top, next to the nitrogen line. The serial number of the controller is on the top right-hand corner of the rear panel. The number of the matched coldhead is shown near the coldhead connections on the rear panel.

-cool/warm procedure

Coldstream cooling procedure:

1. Check that the power is OFF (switch on the back of the controller)
2. Turn power ON, wait for system to do self checks. When it's done, screen reads "ready for use"
3. Press PROGRAM button, use up/down arrows to choose RAMP press ENTER
4. Cursor should be in column RATE. Set rate using arrow keys to 300k/hr. Press ENTER
5. Cursor should be in the TEMP column. Use arrow keys to adjust temp to 100K. Press ENTER
6. Cursor should be in FUNCTION column (below RAMP). Press START.
7. Press SCREEN

*Common problems/solutions*

*Ice block warning: An ice block warning, which reads "Ice Block warning will trip at 2 L/m" on the controller, indicates that the system will shut down eventually because of moisture build-up in the coldhead, but it may not occur for several hours. Since it takes a couple hours to warm up the system to remove the ice block and then to bring it back down to temperature, it is sometimes advantageous to finish collecting a set of data before the ice block is attended to if the nitrogen flow rate has not yet fallen significantly and the unit continues to run well.*

*Try: Drying out the coldhead. Blow dry air at no more than 5 L/min through the flow path of the cold head, and replace the line dryer (the line dryer is probably saturated). If the ice block continues, make sure there are no leaky connectors and replace the diaphragm in the pump if needed. A good indication of a leaky connector is the heat percent. The coldstream should run with a stable heat percent between 10 % - 20 %, a lower heat percent is probably due to a leaky connector.*

*Icing on the crystal: If ice crystals or "snow" begin to accumulate on your crystal, and you do not believe the conditions under which the crystal was frozen are to blame, a likely cause is the misalignment of the coldhead nozzle or the need to adjust the dry air sheath surrounding the nitrogen stream.*

*Try: Aligning the coldhead. A crucial decision is how close to put the snout of the Cryostream to the specimen. We suggest that you bring the cooler down until you can see the shadow of its business end in the crystal-viewing microscope, then back it off until that shadow is just at the edge of the field of view. Some beamlines have an alignment pin which you may place on the coldhead nozzle to determine the optimum minimum distance.*

*Try: Optimizing the laminar flow. The plume of nitrogen, which is produced by the laminar flow of an outer dry air stream and an inner nitrogen stream, can be optimized with the "flashlight test." The plume is comprised of an "invisible" section (that directly emerges from the coldhead nozzle), followed by a wispy, visible plume. It is desirable to lengthen the invisible section as much as possible. With the lights out in the hutch, shine a flashlight towards the coldstream nozzle to highlight the nitrogen plume. To maximize the*

*length of the invisible section of the plume (within which the crystal will reside), adjust the dry air flow. It is not recommended, however, to adjust the dry air flow to levels much higher than 10 L/min; the proper laminar flow will not be maintained and the crystal could be blown from its support.*

*Coldstream cannot reach programmed temperature but is stable: The coldhead may have lost its vacuum.*

*Try: Replace with spare cryosystem if available.*

*Condensation/ice on coldhead SUCT connector: An inward leak in the SUCT path may have thrown off the heat exchange in the coldhead.*

*Try: Replace cryosystem with spare.*

*Icing on nozzle:*

*Try: Changing the flow of dry air. It should be between 10 to 15 L/min. Failing that, dry air compressor may need to be serviced.*

*Shutdown due to large temperature error: Ice in the small dewar is formed on the plastic sensors tube. When the auto fill is attempting to fill the dewar warm gas cannot escape, causing the coldstream fault.*

*Try: Replace with spare. Service by venting pressure, then thawing out sensor tube.*

#### i. mounting/un-mounting crystals

There are several techniques for the mounting and un-mounting of crystals. Basically you practice, and determine which strategy works best for you. The bottom line is that crystals should never be both outside the cryostream, and outside of liquid N<sub>2</sub>. Crystals will typically arrive on a needle, attached to a cap, attached to a vial, attached to a cane, sitting in a supply of LN<sub>2</sub>. The basic strategy is as follows:

1. Put on gloves. Usually the white thermal inner glove with a plastic rubber outer glove
2. Raise the Kappa axis to a nice 45 degree angle
3. Move the detector back to at least 400mm
4. Fill your tall (cane) dewar
5. Fill your small (working) dewar

*Tip: always cover your dewars when not immediately in use. This helps prevent frost and ice from accumulating on the edges.*

6. Transfer the cane you will be using from the dry shipper to the tall dewar
7. Dip your forceps into the small dewar to cool it off. Wait for it to stop boiling.
8. Lift the cane from the tall dewar, and quickly grab the needed vial with the forceps.
9. Quickly transfer the vial to the small dewar.

*Tip: never let your vial be out of a LN<sub>2</sub> supply for more than 30 seconds.*

10. Use a second pair of cooled forceps to grab the vial from the side
11. If the vial is a screw-on type, unscrew it but leave the cap laying on the vial
12. Quickly move the vial up and attach the cap to the goniometer's magnet
13. Remove the tall dewar from the hutch, and replace the cane into the dry shipper.

#### 5. Beamlines, specific (specs, mechanical layout (with visuals), common problems/remedies, checklists, alignment procedure)

*Under maintainance at this time*

- a. X8c
- b. X12b
- c. X12c  
-B4 stuff  
-NONIUS
- d. X25

#### -Manual Control of Diffractometer:

The manual-control panel in the x-ray hutch allows one to drive the diffractometer by hand.

1. The **distance, 2-theta, phi, omega, and kappa** buttons will move their respective translations in the positive (+) and negative (-) directions. To move the motors, press and hold the switch in the direction desired, and release the button to cease motion. These controls do not enable one to move the motors to exact values, though movement to absolute positions may be accomplished through the DET level of CBASS.
2. The red **STOP** button should be pressed if a collision of moving parts is imminent. Once pressed, the STOP button will halt all diffractometer motor functions until the "home" button in CBASS is pressed (you may have to exit and re-enter CBASS first).

3. The **JOG** button will move phi 90 degrees in the direction (+ or -) specified. This function is particularly useful when aligning the crystal to the camera crosshairs.
4. The **Home** button in the Det level of CBASS will return phi, omega, kappa, and 2-theta to the zero (home) position and will drive the detector to its maximum distance. When executed, allow "home" a minute or two to finish before issuing further commands to CBASS.

#### -Crystal-Visualization Microscopes:

There are two video microscopes mounted over and under the crystal position, viewing at approximately 45 degree angles. There are video monitors in easy viewing overhead at the diffractometer, and slaves outside. There are electronic cross-hairs to show the position of the rotation axis (single vertical wire) and the x-ray beam (double horizontal wires, between which the x-rays can be found). The microscope positioned below the crystal is set to its maximum magnification, while the microscope above the crystal is adjusted to its lowest magnification. Illumination comes from a fibre-optic lamp sitting above the specimen. Note that it rarely helps to turn the lamp up above its lowest setting -- if it seems not bright enough, put it closer. Uninitiates are not encouraged to adjust the cross-wires, nor to try to focus the microscope. When in doubt, adjust the crystal to be stationary during rotation and ask your host for advice.

#### -Cross-hair generators:

Sometimes you will find that the cross-hair generator has locked you out. The password for each is generally on the back of the hand-controller.

#### -Q315 detector

The detector type at x25 is the Quantum 315. See specifics in previous section.

#### -photon wiggler

The brightness (flux) associated with x25 is due to her 27 hybrid-pole photon wiggler. This device is generally maintained by Lonny Berman, and Steve LaMarra.



27 pole hybrid photon wiggler at x25

#### -hutch laptop

Inside the x25 hutch is a PC laptop computer which is set up to run the diffractometer. It is mainly there to provide a more exact measure of goniometer and detector positions to the user than are possible with just the diffractometer alone.

##### *Common problems/solutions*

*Computer will not control goniometer.*

*Try: rebooting the laptop. Once re-booted, use Netscape and go to its set homepage.*

*Try: operating the diffractometer with the collection computer via CBASS. If this doesn't work, the power may need to be cycled (see below).*

*Try: Cycling the power. Procedure:*

1. Turn off optix, cbass if running
2. Turn off diffractometer (black switch on the back)

3. *Turn on cbass, watch for it to have problems communicating to the diffractometer*
4. *Turn off cbass*
5. *Turn on the diffractometer*
6. *Restart cbass, optix*

- e. X26c  
-Q4 stuff  
-NONIUS

## 6. FedX projects

- a. Dry shippers

These are the containers that the crystal samples arrive to us in. They are typically dry (hence the name) of LN<sub>2</sub> upon arrival, and need to be filled. Similarly, they need to be emptied (of LN<sub>2</sub>) before they are shipped back.

- b. Crystal screening

This is the process by which we determine what crystals are in the best shape, and are to be given priority in terms of data collection. Typically, we will mount in turn each crystal on a cane, take a five image collection, then attempt to index it using HKL2000. The results of this screening are to be recorded, and thus priority given to the best crystals when more time is available for collection.

- c. Data collection procedure

We have a standardized collection procedure, described here.

- d. Forms (or whatever the final procedure turns out to be)

## 7. Administrative stuff

- a. SAF
- b. BOR, EOR
- c. BLOSA
- d. Shipping procedure (Fed-Ex)