24ID-C operation manual
and troubleshooting

(under permanent construction and improvements)

This manual is NOT finished and will be NEVER finished (version of June 2013)

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General Computer Layout
Our computer network can be roughly separated into five parts: (1) data visualization computer, (2) two computers for the beamline control, (3) MD2 operation computer, (4) three computers for data processing and structure solution and (5) data storage and backup. **You should use computers in category (1) and (2) ONLY for data collection and the beamline controls (no Web browsing, games, music etc)**. In addition, there are two more shared computers in the back user area.

The control room for 24ID-C station is pictured in Figure 1.

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**MD-2 Operation computer** is NOT used by users. The “pgpmac” program should be always running: Check Troubleshooting section on how to restart the whole MD-2 and how to restart ALL user beamline control software.

**Data visualization computer** is used only for current diffraction image visualization. To start the ADXV software: find on the desktop a diamond-shaped icon called “AutoADSC” and double –click on it, a new window with green font will open. Next on “auxiliary” computer in the CONSOLE script under detector tab select “UTILITIES” and choose “Restart ADSC system” – a new GUI will start after in a while. All other functions of the ADSC GUI are for exclusive use of the beamline personnel.

**Beamline Control Computers** The beamline control software runs on two different Windows computers, namely "master" and "auxiliary". The video from these computers is output to 3 rightmost LCD monitors (see picture above). You can switch between these computers by pressing corresponding button on KVM switch below the top shelf. All user operations are done on "auxiliary" computer only.

To start the main script, find on the desktop the icon EXEC_Intel (or it may be called EXECUTOR).
A new empty window will appear. Click on **File** and choose "Prior .CXE Selection", drag the mouse to the right to select the script "C:\CONHEAP\PHI_MD2_PILATUS_NM.CXE". As the beamline is under constant development, the current name of the script may be different; the support beamline personnel will notify you. Click on "Execution Option" and choose "Execute Script"

Now you should see the new window coming (Figure 2):

![Figure 2. MD2 support table script PHI_MD2_PILATUS_NM.cxe](https://example.com/fig2)

This is your MAIN working area and you can do all following functions:

- access the crystal centering interface (3-click alignment, focusing, lights, kappa-goniometer, snapshots collection, basic robot operations) **XTAL CENTERING tab**
- access data collections interface with different protocols **DATA COLLECTION tab**
- adjust the orientation of the MD2 table relative to the beam (**MD2 AUTOALIGN**) 
- change the size of the beam aperture (**MD2 APPERTURE**); check he current aperture value
- change energy and run **EXAFS** and **CHOOCH**
- access the robot control script by pressing **ROBOT LOADER** tab (see next chapter)
- maximize the flux and optimize the beam position (**TRIM_X** and **TRIM_Y POSITION**); move in/out the sample sensor **RETRACT/INSERT SS**;
- retract/insert the cold-steam nose **RETRACT/INSERT CRYO**;
- open/close sample shutter; check the status of the main shutter; get APS ring status
- define the desired beam flux (**TRANSMISSION** button)
- check the current values of the energy, flux (arbitrary units), transmission (in % of the full beam), detector distance, ADSC state and the status of the beamline software (busy/ready)
- access several functions of the script running on the MASTER computer (**MASTER OPS**)
- change cold stream parameters (be very careful!) (**CRYOJET** button)
- move the fluorescent detector IN/OUT to adjust the signal (**SCINT DET POS** button)
- recover from the shutter errors (**WAKE SHUTTER** button)
- read several parameters (to communicate with you support personnel, if requested)
- save or recover the tune state of the beamline, or the current state of the MD2 table

**ATTENTION:** Users are *forbidden* to reboot any beamline computers, unless explicitly authorized by the support personnel.
General notes for trouble-free operations:

Note 1: Please always watch the status of the control system: When the system is busy doing something the indicator in the middle on the left side will say so (red button and SYSTEM BUSY), don’t do anything until the system tells you SYSTEM READY. It is normal for the software to display SYSTEM BUSY for a couple seconds at anytime.

Note 2: Do NOT open the hutch door or close the main shutter, if the system is changing energy or collecting EXAFS scan or doing the autoalignment or collecting diffraction data. This may result in the mechanical damage to MD2. Besides, the beam may be lost because the system will try to optimize zero level flux and will move the mirrors to the wrong positions: Check the troubleshooting section how to recover the lost flux.

Note 3: Only ONE user operation is permitted at a time; i.e. don’t try to align the sample when changing energy, etc. Also crystal centering should be done only at one computer: either inside or outside the hutch.

Note 4: The system responds to the location and time where and when the mouse is released, so be careful NOT to drag the mouse during the "mouse click". Please also avoid double-clicking.

Note 5: The robot is SLOOOW – You can’t make it moving faster: every operation is a set of many instructions and sanity checks, which should be completed before the software can accept a new command. Please be patient, and wait every instruction to be properly finished. Aborting, canceling or just double-clicking may push the system in the delirious state with long recovery time.

Note 6: Please always watch the status of the robot system: When the robot is ready for the new operation/command it will display green ROBOT READY status.

Note 7. Please follow ALL instructions from your support personnel. Any deviations or shortcuts are dangerous and will lead to unpredictable results and/or physical damage of the equipment.

Note 8. During the non-business hours, if there is any problem, first check the troubleshooting section of this manual, before calling your support personnel.
Robot Operations

Inside the Hutch

Figure 3. The main working window for all robotic applications INSIDE the hutch

Unload/Load Pucks

1. Remove any mounted crystals on the goniometer, either manually or, preferably, by pressing DISMOUNT button.

2. Click on PARK/SAFE, select "park gripper" and script will ask you to confirm. Click YES. The robot script will then perform several sanity checks. The sample sensor will detect if there is a pin on the goniometer. If a sample is present, the robot script will ask you to remove the sample or allow the robot to place the sample back in the dewar. If you choose to manually remove, the sample sensor will retract to allow you to reach the pin. The sample sensor may be manually moved—click on “Sample Sensor”. Once the spindle is unoccupied, and MD2 is ready, the robot script will park the gripper.

You should wait and NOT touch anything before the gripper stops in Park Position (facing goniometer).
3. Click **UN/LOAD PUCK**. Next a new popup window will appear – select a puck position (A B C D) or CANCEL the operation.

After puck selection, the dewar will rotate that position directly under the opening and allow for placement of a puck. There are four positions in the dewar: A, B, C, and D. Keep track of which puck goes in which position. After the gripper has been parked, you can access the dewar.

4. Turn off the fan for better viewing. The switch for the fan is on the top of the light-yellow box (left figure). Before pucks can be loaded, the lid must be removed and placed in the lid resting location (i.e on the top of the fan control box): see figures below.

5. Remove the puck from the shipping cane. Turn over the puck so that the thick part of the puck is face up. Attach the puck pusher to the puck. Orient the puck so that the notch is facing the center of the dewar. Load your puck into the dewar by lowering into the liquid nitrogen and feeling for engagement of the notch with the pin. It is recommended to slightly tilt the pusher toward the center of the dewar. When the puck is engaged, then it will not move in the dewar and the puck pusher will be perpendicular to the opening of the dewar. The puck will also be seated inside all three pins. Once the puck is engaged, push down with the plunger (of the puck pusher) to separate the two pieces of the puck. Then lift straight up in order to avoid disturbing the pins. Visually check the alignment of ALL pins, use long (and cooled) forceps to place properly tipped over pins.

6. After loading your puck into the dewar, select the next position by first clicking **UN/LOAD PUCK**, confirm, and then choosing the next position. Due to the sanity checks, this step is slow. Repeat steps 3-5, until up to four pucks are in the dewar.
7. Once all your pucks are loaded, replace the lid on the dewar (make sure the pin on the dewar is engaged in the lid) and turn the fan back on.
8. You must cool the gripper before samples can be mounted. Click COOL GRIPPER. The gripper should enter the dewar and begin cooling. Once the proper temperature has been achieved, you will be able to select a puck (click A, B, C or D) and a sample (click a number in the puck picture). Now, sample mounting can proceed. It is recommended to mount the first crystal inside the hutch, to check proper alignment of the robot. Once PuckID and Sample# changed color to yellow/cyan – press “MOUNT” button and watch the robot mounting the first crystal.

Now all robot operation should be done OUTSIDE the hutch, using the robot script running on auxiliary computer.

### Outside the Hutch

All robot operation can done OUTSIDE the hutch, using either the XTAL CENTERING tab (most used robot functions only) or the full robot script running on auxiliary computer – by press tab “ROBOT LOADER” You should see the following screen (Figure 5):

![Figure 5: “ROBOT LOADER” window for all robot operations](image)

To mount/unmount samples:
1. You must cool the gripper prior to mounting your first sample. Press “COOL GRIPPER”
2. To mount your first sample click the Puck ID (A, B, C or D), then click the Sample ID (#1-16). The Puck ID and Sample ID colors will change to red as the robot moves to the sample location. When the dewar move is complete, the Puck ID color will change to yellow and the Sample ID will change to
cyan.
Make sure that the BUSY light is green (not bright red), ROBOT READY and the status is DISMOUNTED

3. Then click MOUNT under Mount Ops. The robot will start mount the selected crystal;
   Be Patient – it takes ~50 seconds to finish!
When the sample has been mounted, the voice will state, "Ready to Align Sample." At this time, you may align/center your crystal. The robot is still busy doing first de-icing of the gripper and second cooling it. Please do not click anything in the robot window at this time

4. Mount Next Sample. With a sample mounted on the goniometer, you have two choices.
   * If you wish to go to the very next sample in the puck, then click MOUNT NEXT. The robot will dismount the sample and mount the next sequential sample. When it is complete, the voice will state, "Ready to Align Sample." Be patient – it takes ~115 seconds for this step.
   * If you wish to go to new sample in the same puck, then click the Puck ID and then select Sample ID of the desired sample. The puck IDs will change to red, then back to yellow and Sample ID will change to cyan when your selection has been accepted. The robot will dismount the current sample and automatically mount the chosen sample. When the mount is complete, the voice will state, "Ready to Align Sample." Be patient – it takes ~125 seconds for this step.
   * If you wish to mount a crystal from a different puck, it is recommended to first click DISMOUNT before selecting your new sample. The robot will return the currently mounted sample to the puck. Once the crystal is dismounted, the BUSY light is green (not bright red), ROBOT READY, click the Puck ID and then select Sample ID of the desired sample. The puck IDs will change to red, then back to yellow and Sample ID will change to cyan, then your selection has been accepted. The robot will move to the new location and automatically mount the chosen sample. Be Patient – it takes ~70 seconds to finish! The voice will then state, "Ready to Align Sample."
   * The above behavior may be changed by checking “Automount Disabled”: in the Maintenance submenu – the crystal will be NOT automatically mounted after new selection. You need to click MOUNT to mount the selected crystal.

5. Dismount -This command returns the currently mounted sample to the puck. (duration ~80 sec)

6. OPTIONS button will give you the menu with different washing scenarios. The most commonly used are:
   POST_WASH to wash the crystal after mounting : it instruct the robot to move the crystal back to the dewar, move it back and forth to remove any ice on the loop surface and mount it back.
   WASHING MACHINE – The whole dewar is strongly agitated several times to remove ice from many crystals at once; it is recommended to perform this operation after loading your pucks
   ENABLE PRE_WASH – Before mounting every new crystal, the gripper will be agitating up and down with every crystal before mounting.

7. If a long time has passed between sample mounts (such as the duration of a long data collection), De-Ice Gripper and then Cool Gripper prior to dis-mounting. Ice builds up on the gripper during long intervals and will enter the storage dewar if not removed by de-icing. Ice in the storage dewar is drawn to any crystals present.

8. To move the gripper into a safe state for maintenance (or after finishing the experiment) - click SAFE GRIPPER.
Crystal Centering and Data Collection

Two tabs: XTAL CENTERING and DATA COLLECTION tabs are regularly used during most of user operations.

**XTAL CENTERING** is the extension of the centering mode of MD2 software for use with minikappa goniometer and several robot manipulation tasks.

![General view of "XTAL CENTERING" tab](image)

Figure 6. General view of "XTAL CENTERING" tab

There are several embedded operations in this window (from left top corner clockwise):

1. Robot puck ID number (current) and the button to mount next crystal - "SELECT SAMPLE". You can also dismount a currently mounted crystal, mount new crystal, wash it and select pre-wash cycle. When "BUSY" light is red - the robot is busy doing something, wait before clicking.

2. Section to control camera zoom, lighting of the crystal, and ability to acquire or view your crystal picture. StartServer button should be used only if the separate visual window is not started automatically (to be used by beamline personnel only).

3. The function to acquire STAC strategy parameters from RAPD for better crystal alignment (i.e
long axis close to omega rotation axis) and align the center of the crystal at new kappa/phi. Make sure you are using the correct values and the most recent ones (check timestamp). “Align” will automatically re-center the crystal to the new kappa/phi orientation in small steps.

4. Controls to manually adjust Kappa/Phi/Omega for mini-kappa; if using single-axis goniometer-only omega should be used; predefined crystal rotations to check the alignment. Users are advised not to change kappa angle more than 40 degrees in one step, as the crystal may go out of the cold stream.

5. Three button to change MD2 mode: Center(default), Data collection and Sample Transfer

6. Buttons for crystals rotation and input field for a specific phi/omega angle.

7. 3-CLICK alignment tools, including alignment for crystal rastering: First, CLEAR the database (if necessary), second do 3-CLICK at the beginning of the raster vector, press RECORD; third, repeat 3-CLICK centering at the end of the rastering vector, and press again RECORD, it should be number 2 in the left box. DISPLAY will show consequently the beginning and the end of the raster vector and REPLAY will repeat two alignment processes. More than two points may be created for complex cases. 3-click alignment should be completely finished, before proceeding to the next operation. ABORT may be used to leave the "3-click" function.

8. Snapshot button to collect snapshots – a snap constructor dialog will appear

9. Buttons for manual movements of the crystal: vertical, horizontal and along the beam (out-focus plane). 1, 10 or 50 microns increments.

**DATA COLLECTION** tab is used to control ADSC snapshot/data collection parameters.

Figure 7. General view of "DATA COLLECTION" tab (discrete vector run in progress)
1. **SNAP CONSTRUCTOR** button opens the following dialog window:

Here you choose all parameters for snapshots. First, you need to define the root directory for your data collection (unless that was not done earlier) - by clicking on "Select Browse Root". It should be the *images* sub-directory below the directory assigned to your group. Next click on "Browse" button and select, or create a new directory, for current project. Confirm by clicking “Update”. Next, type the file prefix in the corresponding field. Click "Reset Index", if necessary. Select "Pair Snap Acquisition". When "Paired Snap Acquisition" is ON, two images separated by chosen offset value are collected, and LABELIT/RAPD will autoindex them together with much higher success. Type in Initial phi, oscillation range, exposure time, detector distance and transmission. For high-resolution data collection or to resolve overlapping reflections, it is possible to move detector up or down – use “Segments Vertical Lift” field. Click "Update". Resolution Limits are updated once detector distance is defined and UPDATE is pressed. EXECUTE leads to collection of defined snapshot(s). EXIT closes the dialog window and return operations to DATA COLLECTION tab window.

*Note 1:* It's VERY important to click "Update" on this window and similar ones for data collection; otherwise previously collected image(s) may be overwritten.

*Note 2:* Detector distance should be in the range of 1000 to 150 mm.
2. **RUN CONSTRUCTOR** - the following dialogs opens, depending on what is selected when DATA MODE was pressed:

2A. (default) STATIC ALIGNMENT is for usual data collection:

![Run Constructor Dialog](image)

Figure 8. Run constructor dialog with five separated runs.

Here you choose all parameters for standard data collection: Every field can be manually edited, as necessary. First, you need to define the root directory for your data collection (unless that was not done earlier) - by clicking on "Select Browse Root". It should be the *images* sub-directory below the directory assigned to your group. Next click on "Browse" button and select, or create a new directory, for current project. Next type the file prefix in the corresponding field. Select binning mode (1x1 or 2x2), and "Friedel Flipping" and corresponding wedge size (if required). Shutterless Ops should be ON. Type in Run ID, Number of frames to collect, Detector distance, Transmission, Initial omega/phi angle, Oscillation range, and Exposure time. Repeat for another run ID, if necessary (figure above shows an example of five runs of 40 degrees each). Once all values are checked, press "Update" and "Execute". EXECUTE leads to collection of defined run. EXIT closes the dialog window and return operations to DATA COLLECTION tab window.

Data collection can be aborted by pressing "ABORT RUN" button on dialog menu or “**ABORT RUN**” button in the middle right area of “DATA COLLECTION” tab – see Figure 7. This operation is slow and several images may be collected, before the run is completely aborted; please wait and do not press any other buttons.
More complicated scenarios are defined when pressing "**DATA MODE**" button - see menu at right -->

2A. STATIC ALIGNMENT – described above
2B. STAC PREDICTION - under development
2C. BEST STRATEGY - under development.

2D. **DISCRETE VECTOR SCAN** - discrete data collection all over the predefined vector along your crystal (vector is defined by recording two (or more) points along the crystal on Centering tab window (see above). A new setup window will appear where you choose the aperture size, zoom view and omega angle for visualization. Next select rastering parameters: Inset - the shift to the right from the leftmost raster vector point for start of data collection. Offset (usually more than aperture size) is the shift along the raster vector for next data set collection. Cycles - number of data sets to be collected. Phi Offset - is the shift of phi/omega angle between different data collection points. Note: for continuous data collection PhiOffset should be equal number of "Frames" number times "Del Omega".

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**Figure 9. Discrete vector scan dialog and graphical representation of data collection spots.**

Pressing "**GENERATE RUN DATA**" will open a graphics window in the top left corner, where you may monitor relative positions of every run along the raster vector. Change Omega values to check the alignment at different orientations. Press RUN and a “Run Constructor” dialog will appear (Figure 8).

In the "Run Constructor" dialog window you should setup subdirectory for data collection and file prefix (see above), check the correctness of all runs. If there is any mistake, Click "Exit" and go back to
"DATA MODE" and make all changes in the "DISCRETE VECTOR SCAN" dialog. If all runs are correct and all values are checked, press "Update" and "Execute". EXECUTE leads to collection of several defined runs along your crystal. EXIT closes the dialog window and return operations to DATA COLLECTION tab window.

The CONTINUOUS VECTOR SCAN (described next) provides better protocol to decrease radiation damage, as the crystal is rotated and shifted after every collected image.

2E. CONTINUOUS VECTOR SCAN – (sometimes called helical scan) – more advanced mode as compared with "DISCRETE VECTOR SCAN" - continuous data collection all over the predefined raster vector. In this mode the crystal is translated/rotated after EVERY image collected to better spread the radiation damage. In the “ContinuousVectorScan” dialog window you should choose the aperture size, zoom view and starting omega angle for crystal visualization.

Figure 10. Continuous vector (helical) scan dialog and a graphical representation of data collection along the crystal.

LeftInset and RightInset are offsets from predefined raster vector ends (not zeros - if you want to collect data on a shorted portion of the crystal). They may be negative if you want to extend data collection beyond predefined vector. Omega defines the initial view of the crystal, there are +/-5 and +/-45 degrees rotations buttons.

Next, setup all data collection parameters based on the calculated strategy by RAPD (initial omega/phi angle, oscillation range, number of frames, exposure time, transmission and detector distance.

Pressing "GENERATE RUN DATA" will open a graphics window on the top left corner (Figure 10), where you may monitor relative positions of every image point along the raster vector, with all data collection point simulated in red.

Pressing “RUN” will open ADSC data collection dialog which show only one run and it should not be edited (except run number and directory name). Just press "Update" and "Execute" for data collection.
**2F ALIGNMENT BY DIFFRACTION** - is used to find your crystal in an opaque/non-clear loop: the loop is rastered by a very weak beam and the software analyzes all collected images to find one with the better and stronger diffraction. In the following dialog window you select the necessary parameters:

![Alignment by diffraction](image)

Figure 11. “Alignment by diffraction” dialog and a graphical representation of data collection along the crystal (top left graphics window).

It is recommended to use 70 micron aperture for big loop and use Omega value corresponding to plane of the loop. The "Metric" parameters are used by DISTIL to recognize best diffraction: "Good B Spots" is the best choice for now. "Overlap" defines the overlap between different search points: use a negative number for a sparse search.

Press "DEFINE SCAN" and a new window will appear in the top left side, use the left mouse to define a rectangular search box. The final search area will be automatically defined as a convolution of your search box and image centering server oval covering the loop.

Change transmission value to 1% or less and define the desired detector distance. Exposure and DelOmega should be 1 second and 1 degree (or less). Press "RUN SCAN"; the system will move the loop through all red circles/ellipses and store the corresponding coordinates. After that on ADSC computer the diffraction images will appear. All images are stored in "/gpfs5/users/necat/phi_dfa_1/in" directory and will be deleted after exiting. The message will appear when the run is complete.
A table with processed results will appear (see figure below), on which it is possible to select different points and check again the corresponding diffraction (can be also done by making selection in lower left graphics window by clicking on different spots).

![Table of Results](image)

**Figure 12. A summary of the diffraction metrics after running “alignment by diffraction”, “occluded view” or “raster snap” mode**

Once the region of the best/strongest diffraction is found, pressing “MOVE TO” will center that portion of the loop for subsequent data collection. Or, if it is necessary, you may save all diffraction images in the specified directory by clicking on “SAVE RAME DATA”.

You may proceed to scan the orthogonal orientation using the “OCCLUDED VIEW” mode.

Click EXIT to close the Diffraction Alignment dialog.

### 2G. RASTER SNAP

In the dialog window choose the aperture size, zoom and omega angle for specific crystal orientation. Next select rastering parameters: LeftInset and RightInset are offsets from predefined raster vector ends (not zeros - if you want to collect data on a shortened portion of the crystal). Frames - number of snapshots to be collected along the crystal. Select starting angle, oscillation range, exposure, transmission and detector distance.

Pressing "GENERATE RUN DATA" will open a data collection dialog and in a graphics window in the top left corner you may monitor relative positions of every run along the raster vector at the orientation defined by OMEGA value. You may change it for different view, and press again "GENERATE RUN DATA"; once satisfied, click EXECUTE and on ADSC computer you may monitor the incoming images. After the completion of the "raster snap" run another window with simple results will pop-up:

You may analyze (and plot) the results table to find the best portion of the crystal. Or you may select any position of the crystal and press "move to" to center that portion of the crystal.

### 2H. OCCLUDED VIEW Alignment

- With very thin crystals, it is often difficult to visually discern the location of the crystal when in the plane of the loop. First, you need to center at any point in the loop, when the crystal is visible (perpendicular view of the plane of the crystal). Second, choose the view corresponding to the plane of the loop (Occluded View), and define a box for scanning, by pressing “DEFINE SCAN”. The software calculates a series of points for scanning perpendicular to the plane of view based on user-entered criteria, and then takes snapshots at each point, when RUN
SCAN button is selected. RESET button permit you to reset and change scan parameters.

A summary of the diffraction metrics (similar to figure 12) is displayed and you can select a point corresponding to diffraction maxima which causes the MD2 x,y,z centering stages step to the selected scan point coordinates. This point can then be entered into the vector database for use in other vector-based scanning methods.
How to change the energy

Click MONO ENERGY in the left top portion of the menu; you have two choices of changing energy – using “edge selector” (you choose the desired element from the periodic table and its K-, L- or M-edge) or clicking on “specify discrete energy” (you type any value in the range 6000-20000eV); click “cancel” if you change your mind. See the beamline personnel, if you need to use energy above 13600eV.

![Energy change dialogs](image)

Unless you change energy below 8200eV, it takes less than one minute to change energy, wait until the green light indicates “SYSTEM READY/STABLE”, in the top left portion of the menu click both TRIM X and TRIM Y POSITION (the X/Y beam position values should have green color). The energy change below 8200eV involves changing the mirror strip – it takes about 10 minutes to finish. So, be patient.

The system will perform an autoalignment after EVERY energy change more than 500eV. Please be patient –let the system to find and center the beam!!! (~1-2 minutes)

Note: Do NOT open the hutch door or close the main shutter, if the system is changing energy or doing autoalignment. This may results in the mechanical damage to MD2 and the beam may be lost.
How to collect EXAFS scan

To collect EXAFS scan from your protein crystal - use the biggest crystal for EXAFS and save the best crystal for diffraction data collection. The crystal for the EXAFS scan needs not be single, but the size matters. Currently, the beam flux used to collect EXAFS may damage your crystal. Metal foils can be also used for EXAFS to calibrate the energy or your support personnel may suggest the correct energy for your element, based on the previously collected EXAFS data.

Check the energy and/or move it manually (see above how to change energy) close to the desired edge. For energies below 8200eV it is recommended first to change energy to desired value and only after that perform EXAFS.

The full procedure of EXAFS is fully automated now: click on XAFS scan – the new window with periodic table will appear (bottom part of figure 13); choose your element and the desired edge (within the range of 6000-14000 eV). The system will change energy automatically, and it will perform an autoalignment after EVERY energy change more than 500eV. Be patient, wait until it’s finished. Once it’s done a new popup input window will appear – see Figure 14.

First, check that the fluorescent detector did move toward the sample, if not, click on “TOGGLE YAP POSITION” (The lowest button in the EXAFS popup menu – Figure 14).

Second, check that INITIAL ENERGY is close to the desired energy (~20 eV below the edge); SCAN RANGE should be 40-50eV with energy step 0.5-1eV (don’t change INTEG TIME 5.0 and DAMP DWELL 200). In the DESC field put some info about your protein, put a file name in the OUTPUT field (you will need it later for processing by CHOOCH); the file extension should be “.dat”

Third, the flux of the beam should be optimized at the peak energy (or slightly above it) so that the fluorescent scattering from the crystal is not overloading the “SDD DETECTOR” (on rightmost monitor- left side of figure 15). Open the shutter (click on toggle shutter in the lower portion of menu) and watch the counts appear in the “SDD_DETECTOR”. You want make sure that the counts are in the...
range of 1-5 units, by adjusting the beam flux (less than 1% in most cases). Optimal counts are around 3-5 as shown in the Figure 15 below. Adjust the transmission, if necessary. Close the shutter.

![Figure 15. The view of the right most monitor of the beamline computer, illustrating proper flux for EXAFS](image)

**Forth**, press **ACCEPT RUN SCAN** and next **EXIT XAFS CONFIG**. Hit **ABORT XAFS SCAN** if you change your mind (this is valid only after first point is measured). Another graphics window opens on the rightmost monitor, in a minute it will be showing the acquisition of real-time fluorescent signal (blue), very rough derivative of it (red) and the baseline flux (green). It takes ~3-5 minutes to collect 50eV scan and the crystal is irradiated for less than one minute. Once it's done (the system will announce “EXAFS energy scan complete”, close the graphics window by clicking on “**MASTER OPS**” and select “CLEAR MASTER XAFS NTGRAPH”. This menu also has a button to abort running XAFS scan.

Now click on “**chooch**”, select your previous file name for exafs scan, select the anomalous element and write down the calculated inflection point, peak energy and the values for $f'$ and $f''$. *(note that for the very noisy exafs scan the results could be incorrect, always compare calculated value with the real fluorescent scan).* You may annotate both experimental and calculated plots (click on any point and press “annotate” and print the graph in a landscape orientation.

Change the beam flux back to the appropriate value for data collection. Use your preferred strategy to collect MAD data (like - peak energy first, inflection point next and finally – at remote energy); for SAD data collection move energy to the “peak” value.

*Note 1: Do NOT open the hutch door or close the main shutter, if the system is changing energy or doing EXAFS. This may results in the mechanical damage to MD2 and the beam may be lost*

*Note 2: Do NOT try to collect EXAFS scan from mercury containing proteins, as the Hg white line is extremely broad. Use any energy above 12310 eV to collect “peak” Hg data.*
Troubleshooting (in progress)

1. **Lost beam (no flux)** - make sure that all lights are green on PSS panel and there is beam in the ring (check TV screen). If the beam was lost in the ring, after the repair is done APS will announce “shutter permit is enabled” – after that, manually open station A shutter on a PSS panel and wait for 15 minutes for a monochromator to warm up.

   If there is a red light “Faults Trips” – call the floor coordinator (2-0101) to inquire about this problem. Still no flux – change energy to 12658eV (still no flux, change energy to 11000eV and again to 12658eV). Click “MAXIMIZE FLUX” in the top-left corner of the menu. Call your support beamline personnel if none of the above brings the beam.

2. **The experimental station cannot be searched** – Check that the red button on the yellow box is not pressed in (occasionally someone may lean and push it). Check PSS panel: If there is a red light “Faults Trips” – call the floor coordinator (2-0101) to inquire about this problem.

3. **Detector software does not collect any data** after pressing “collect” – may be the problem of MD2 software not running or communicating with EXECUTOR. On MD2 computer press F1 button, thus stopping the software. In the same window type “/pgpmac” and press return, wait for all field to populate; re-start EXECUTOR. Next on “auxiliary” computer in the CONSOLE script under “DATA COLLECTION” tab select “UTILITIES” and choose “Restart ADSC system” – a new GUI will start after in a while.

   It may be necessary to check for PILATUS software to be running. Exit EXECUTOR. On KVM switch press 4 and press button on the middle monitor. In the black window you may see a message “Access denied” – type “quit” and “exit” (hit returns). The black window should disappear. In the white window type “/camonly” and wait until all text output in the new black window stops. In the black window type “setenergy 12658” (or current energy with space), hit return, and wait for text output to finish. On KVM switch press 2 and press again button on the middle monitor to go back to EXECUTOR window. Re-start EXECUTOR.

4. Alarm is sounding and the detector is moving back because some foreign object went too close to the front of the detector. Report it to your support personnel and disable the alarm on the panel.

5. If any computer is slow or frozen, the corresponding software should be restarted. To close the frozen EXECUTER window, press Execution Options – Abort SCRIPT in the top menu (use Ctrl-Alt-Del only as a last resort to kill CONSOLE, (do not send any report to MS). To start a new CONSOLE script, double-click on the desktop icon called “EXEC_Intel”, click on “Execution Option” and choose “Execute Script”.

   The following software should be always running:
   - Master beamline computer (Far right computer; to access this screen use KVM switch): EXECUTOR - C:\CONHEAP\KONZUU_FULL_REDIS_ANPTEK.CXE script
   - Auxiliary beamline computer: EXECUTOR- C:\CONHEAP\PHI_PILATUS_NM.CXE script;
   - Robot computer (inside hutch) : C:\CONHEAP\ROBOT-C_CENTERING.CXE, in a separate windows: “Console Serial Port Server” and “Robot Remote Server”
   - MD2 operation computer: pgpmac (in the background).
6. Restarting the whole MD2 controller – this operation should be done ONLY after receiving authorization from your support personnel.

Switching off sequence:

1. Exit MD2 Control Program on the PC.
2. Go inside C hutch and locate the MD2 control box (back wall, upstream and behind the MD2).
3. Press the RED Motion Supply button to stop all motors.
4. Wait for 30 secs.
5. Press the Main Supply to OFF position.

Switching on sequence:

1. Press the Main Supply to ON position.
2. Press the GREEN Motion Supply button to enable all motors.
3. Wait for 30 secs.
4. Start MD2 Control Software on the PC.

7. Restarting GALIL server
Login to 164.54.212.68 or go inside the hutch and look for the crate behind the detector;
Open a new shell and make sure galil is not running by typing “ps –e |grep galil”
Type “cd /usr/local/galil_rpc_server”, type “./galil_rpc_server &”
Check again if galil is running by typing “ps –e |grep galil”, close the terminal
Master and auxiliary EXECUTOR scripts and ADSC software should be restarted after restarting galil server.

8. List of running servers (in progress):
Terminal is behind the adsc detector ; the KVM switch is behind the monitor
   164.54.212.67 – pc68_rpc_server and hei_rpc_server
   164.54.212.68 – pc68_rpc_server and galil_rpc_server
   164.54.212.66 (in the rack close to LN2 controller by Station A) – serial…, utility…, diamond…,
and pc68_rpc_server