IMSERC User Manual for Rigaku Synergy-ED

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INTRODUCTION

Use of this instrument is allowed only by qualified users after receiving training by a staff member. Do not run this instrument without approval from IMSERC staff. Failure to do so may cause damage to the instrument, produce invalid data, and result in additional fees and/or removal of all IMSERC privileges. This set of instructions is meant to serve as a guide for 'routine' data collection on the instrument. For custom experiments that are not covered in this user manual, contact a staff member. For the full list of modes, capabilities, and potential custom experiments that could be run on this instrument, please either contact a staff member or check the corresponding capabilities section at http://imserc.northwestern.edu/crystallography-instruments.html. Please read this user manual and acquaint yourself with the instrument.

A hard copy of this user manual can be found near the instrument. An electronic version of this user manual is linked to the desktop of the instrument computer and also available under the corresponding instrument section at http://imserc.northwestern.edu/crystallography-instruments.html by pressing on the 'User manual' button. If while using the system, something happens that you do not understand, please ***stop***, and ***get help***. In any event, be completely prepared to justify your actions. The cost of even minor repairs could be considerable.

SAFETY

All users of IMSERC must review the general safety policies at http://imserc.northwestern.edu/about-policies.html and the Crystallography specific policies at http://imserc.northwestern.edu/crystallography-policies.html. To become an independent user of this instrument, you must have the following safety training and certificates under your LUMEN profile:

- Laboratory Safety
- Personal Protective Equipment
- Filling and Maintenance of Liquid Nitrogen Tanks

You need the above certificates to be able to reserve time for this instrument on NUcore. Online classes and certification are offered at https://learn.northwestern.edu. Upon completion of the certificate, it will take an overnight to filter through the different systems and get into the files that NUcore uses. Additionally, familiarize yourself with the location of standard safety stations like eye wash and shower stations found outside of room BG70. Protective eyewear is required in this room, and gloves should be removed when using the computer.

DATA MANAGEMENT

Your personal data folder is created during training. Please save data under your personal folder, which must be located under your supervisor's group folder, otherwise you might not be able to access your data remotely. See a staff member if you do not have a personal folder on this instrument yet. For users that prefer to name their data folders using dates, use the order of YYYY-MM-DD or YYYYMMDD in the name, so that folders can be sorted chronologically by the operating system if needed.

Data from this instrument are copied in your supervisor's group folder on 'imsercdata.northwestern.edu' under 'xrd/Synergy-ED' every 20 minutes. Please follow instructions at http://imserc.northwestern.edu/about-general-faq.html#data for details about data access.

SOFTWARE

Data reduction and analysis can be performed with 'CrysAlisPro'. Software is installed on the instrument computer. For offline analysis, please use any the following resources:

- For registered IMSERC users, software can be downloaded from 'imsercdata.northwestern.edu' under the folder 'public/Crystallography/Rigaku'. Software is available for Windows only. Please follow instructions under 'Data Access' at http://imserc.northwestern.edu/about-general-faq.html#data on how to connect to the 'public' folder
- You have the option to use the instrument computer for analyses, but you must reserve instrument time through NUcore

DEFAULT INSTRUMENT STATUS

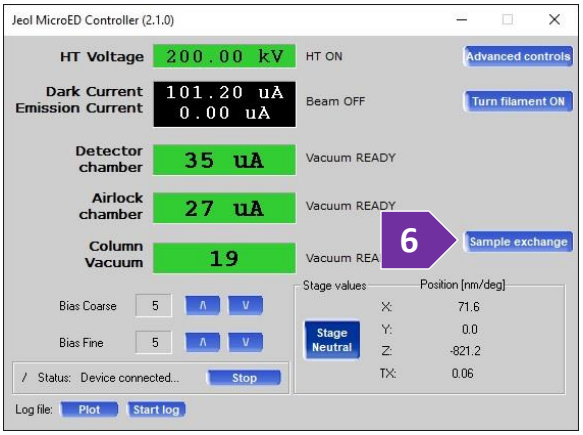
The default measurement mode of the Synergy-ED is ***room temperature or low temperature collections in transmission geometry using parallel electron beam at 200 kV***. Please notify the right staff member well in advance if you would like to run an experiment in a different mode than the one listed above, and you are not trained to perform the required mode-switch. Additionally, put a note on your NUcore reservation indicating the preferred mode of your measurement. For the full list of modes, capabilities, and potential custom experiments that could be run on this instrument, please either contact a Crystallography staff member or check the corresponding capabilities section at http://imserc.northwestern.edu/crystallography-instruments.html.

The default working condition of the Synergy-ED is as follows:

1. Computer screen is by default deactivated. You must start your reservation through NUcore to be able to turn on the computer screen. If screen is already on, start your reservation through NUcore
2. The default 'Synergy-ED' user account should be logged in. In case the computer was restarted, the password for the 'Synergy-ED' account is
3. State of the vacuum and high voltage indicators on the console at the northwest corner of the room should be green as shown in figure 3
4. Emission current should be off, dark current at around 101 uA, detector chamber and column vacuum levels green, and stage neutralized

When you are done with your measurement, please remember to:

5. Leave the acquisition software running
6. Press on the [Sample exchange] button on the 'JEOL MicroED Controller' window (figure 6). This will reset the goniometer and turn the filament off
7. Remove the holder from the column and remove the sample grid from the holder
8. In case you're using the cryo-holder, put the cryo-holder in the pumping station and start the 'Warm Up' procedure, so that the holder is ready for the next user. Check the 'Using the cryo-holder' section for more details



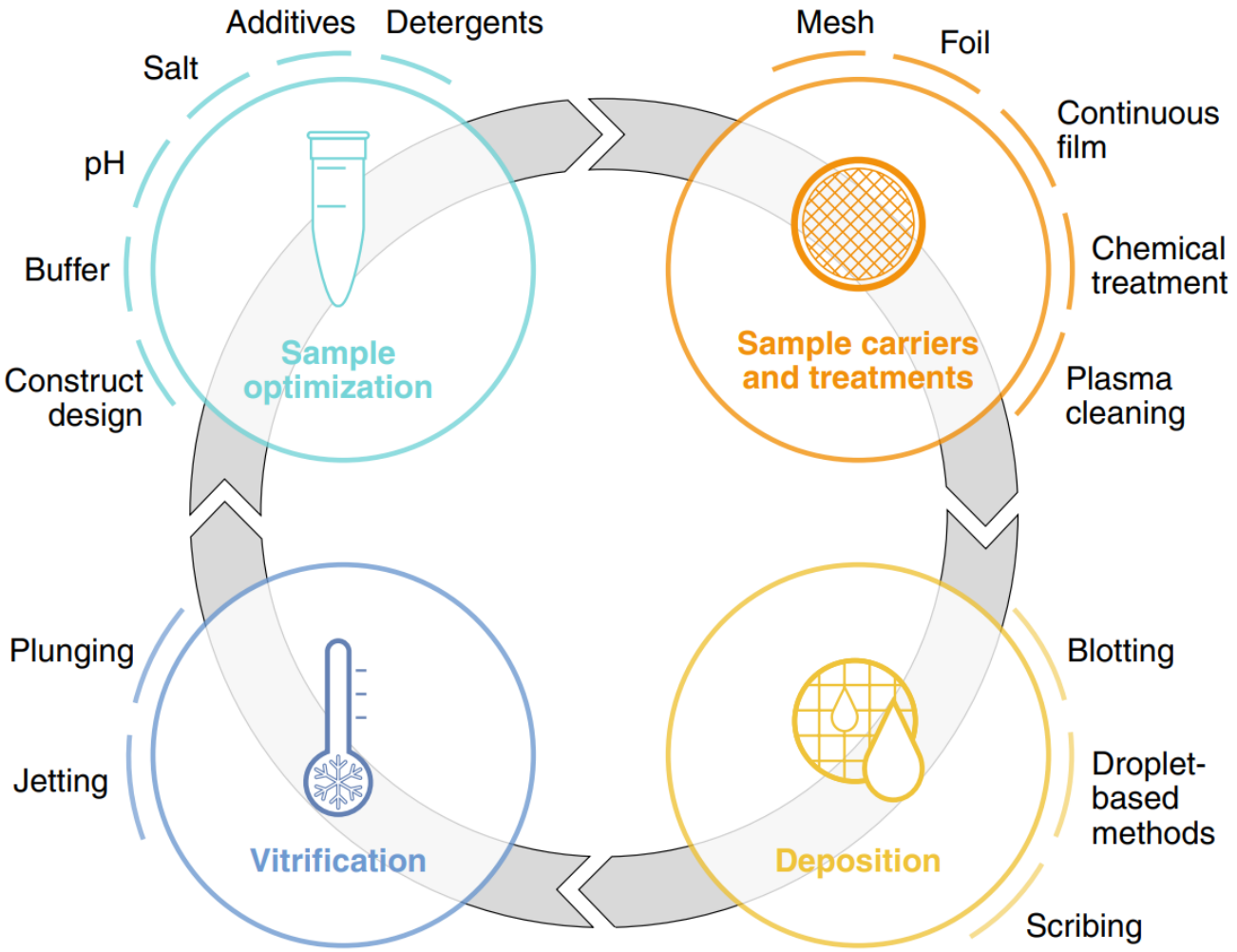
9. End your reservation in NUcore when all procedures are completed. i.e., account for any additional time that might be needed such as warming up the cryo-holder for the next user
10. Leave lab tables clean and tools/accessories organized. Thoroughly clean all tools/accessories with alcohol and kimwipes
11. If you are the last use of the day, please follow the detailed procedure regarding:
    a. The cryo-holder 'Warm Up' or 'Bake Out' process. Check the 'Using the cryo-holder' section for more details
    b. The ACD heater of the cryo-trap on the column. Check the 'Manual collection' section for more information

If there is an error or problem with the instrument which is not addressed under the 'Troubleshooting' section, please report the issue by following at least one of the steps below:

1. If you have already started your reservation using NUcore, please end your reservation and select the error reporting option with a brief description about the issue. Place the 'Stop' sign near the instrument computer to notify users immediately after you. 'Stop' signs are located on the shelf above the computers in BG51
2. If you have not started your reservation using NUcore, please report problems with the instrument at http://imserc.northwestern.edu/contact-issue.html and place the 'Stop' sign near the instrument computer
3. Contact a staff member for instructions

GENERAL STEPS FOR SAMPLE PREPARATION

Sample preparation is very sample dependent. For example, if sample is vacuum stable, a room temperature deposition of the solid or solid in solution on the sample grid might be more than enough. In any case, the sample must be crystalline. You could use powder diffraction to evaluate crystallinity if sample volume is sufficient. Promising crystalline samples should show reflections up to at least 30 degrees two theta when using Cu-radiation powder diffraction. An overview of potential sample preparation methods is shown in the figure below.



Sample preparation workflow is very sample dependent. Start with the simplest method by depositing the sample directly on an untreated sample grid and if this method does not work follow the illustrated workflow clockwise starting from the top right section. Adopted from "Nature Methods (2021), 18, 463 – 471 [10.1038/s41592-021-01130-6]"

If sample is vacuum unstable, cryo-conditions might be needed. Below are some general guidelines regarding unstable or biological samples:

- Samples with high solvent content: Proteins, porous materials, and hydrates are examples of samples that may require cryogenic conditions due to loss of crystallinity upon de-solvation. This can be prevented by freezing the sample before inserting into the diffractometer using the cryo-holder.
- Low molecular weight samples: Low molecular weight samples may sublimate under the vacuum of the electron column. If this is a concern, the specimen should be cooled to cryogenic temperatures before inserting into the diffractometer. In cases where room temperature sublimation is suspected but unknown, the ACD trap can be cooled while operating at room temperature.
- Crystalline suspensions: Suspensions of microcrystalline material can be analyzed by pipetting a small (typically around 1-3 µL) amount of the suspension onto a TEM grid. Excess solvent can either be removed by evaporation or by blotting with filter paper. If the sample is not stable to thorough drying, it can be frozen with small amounts of solvent present before analysis.
- Primary fixation with aldehydes (proteins): During this step proteins and, to a lesser extent, other cell molecules, become crosslinked by formaldehyde and/or glutaraldehyde molecules. Small mammals can be fixed by perfusion, whereby the fixative is introduced via the vascular system. Other samples need to be fixed by immersion and the specimen needs to be dissected no thicker than 1 mm in at least one direction.
- Secondary fixation with osmium tetroxide (lipids): This step ensures that lipids, for example the phospholipids forming membranes, are preserved and are not extracted during dehydration. During the fixation a black insoluble precipitate is formed on the membranes, creating membrane contrast.
- Tertiary fixation and contrasting with uranyl acetate: Uranyl acetate is a heavy metal salt which binds to proteins, lipids and nucleic acids, providing additional contrast. Some authors believe it also has fixative properties. Samples can be incubated en bloc in a solution of uranyl acetate before dehydration, but the stain can also be applied to the sectioned specimen before lead staining.
- Dehydration series with solvent (ethanol or acetone): A fixed specimen is dehydrated by incubation in a series of ethanol or acetone solutions. Solvent concentration is increased gradually so that water is removed gently, without causing artefacts, mainly shrinkage.
- Resin infiltration and embedding: Following dehydration, the solvent is replaced with a gradually increasing concentration of liquid resin (typically epoxy resin for ultrastructure studies). The specimen

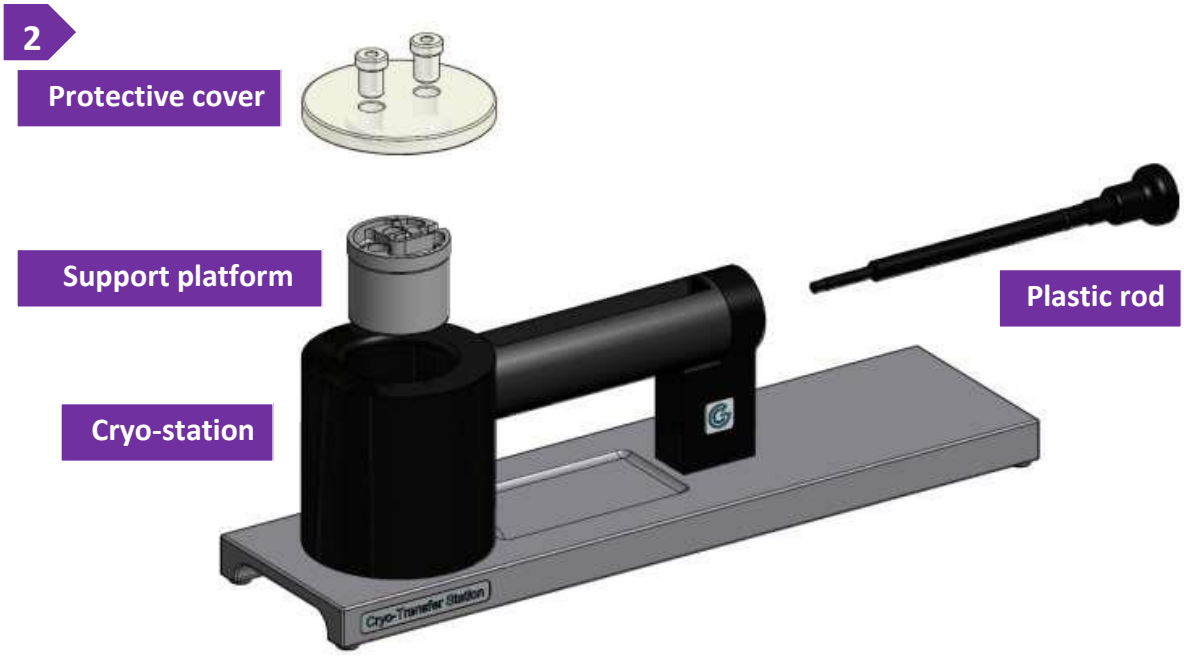
is placed in a mold filled with liquid resin and cured into a hard block using heat or UV light. After this, a sample can be stored indefinitely.

- Sectioning and mounting sections on specimen grids: A specimen embedded in hardened resin can be sectioned extremely thinly, at less than 100 µm. This allows for the electron beam to pass from the electron gun through the specimen to the detector. The sections are mounted on specimen grids which fit into microscope sample holder.
- Contrasting (post-staining): Biological specimens are naturally not very electron opaque as they are composed of atoms with low atomic numbers and the beam passes through them easily. To increase ample contrast, the sections can be post-stained with lead citrate. This heavy metal salt, similarly, to osmium tetroxide and uranyl acetate, binds to cell components and scatters the incident beam electrons. The areas of specimen section which scatter electrons more are recorded as darker pixels, which stand out against the brighter background. Use the flat plate disc holder in transmission mode for qualitative analyses on powder sandwiched between acetate, polyimide, or mylar foils (high throughput option up to 30 flat plates). Use the appropriate foil based on the details discussed during training. Polyimide and acetate foils are available to all users and supplies are located on the bench near the instrument

MOUNTING TOOLS

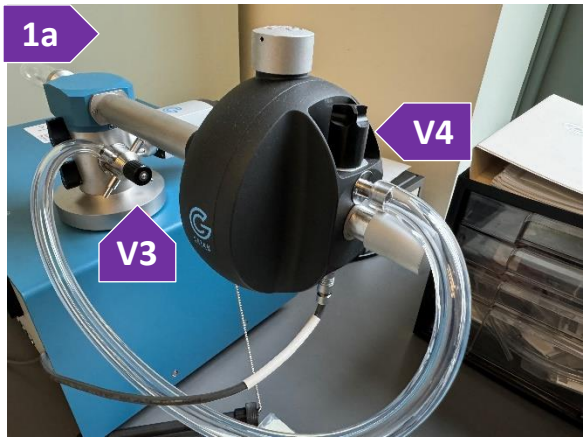
A wide variety of mounting tools and grids are available to use for handling samples. For cryo-related work, commonly used tools are shown in Figure 1 below.

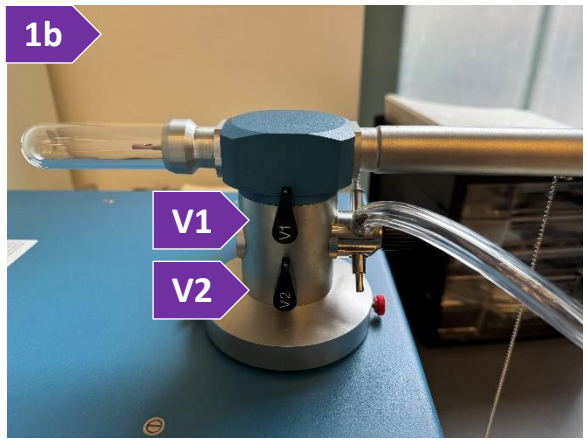
For cryo-related work, when using the Gatan Elsa 698 cryo-holder, the cryo-transfer station and its accessories are used. Cryo-station and its accessories are shown in Figure 2.



USING THE CRYO-HOLDER

The ELSA cryo-holder is a specialized holder with a large cryogenic dewar that can maintain temperature for around 8 hours. Please follow the steps below for the proper use and required maintenance by users of the cryo-holder:





1. Cool down the Elsa holder using the 'Cryo Transfer' mode (should be done on the desk with the pumping station)
   a. Ensure that the cryo-holder is on the pumping station as shown in figure 1a
   b. Ensure that the V4 and V3 valves (see labels of figure 1a) are closed. If not, close these valves first (close V4 and then V3) by rotating them clockwise. Do not overtighten the valves, finger tight is good enough. Remove hose from below valve V4 if necessary
   c. Ensure that valves V1 and V2 are open. See labels in figure 1b for the position of the valves which are shown open
   d. Attach the Elsa holder to the Elsa temperature controller using the cable (thick grey cord). Make sure to connect the red dots to one another (figure 1d)
   e. If controller is not on, touch on the Gatan 'G' logo to turn on the controller (figure 1e)
   f. Using the touch screen on the controller of the cryo-holder, switch to 'Cryo Transfer' mode and start by:
      i. Pressing on the [Mode] button
      ii. Pressing on the [Cryo Transfer] button
      iii. Pressing on the [Start] button
   g. Slowly add liquid nitrogen to the dewar of the cryo-holder until dewar is full. Wait for the temperature to reach around 140K. This might take 15-20 minutes. At the meantime, start the next step of preparing the cryo-transfer station
2. Prepare the cryo-transfer station by mounting the required accessories (see figure 2 under 'Mounting Tools' section)

a. Insert the plastic rod (see figure 2 under 'Mounting Tools' section)

b. Insert the support platform. Ensure that the platform is in the pins of the cryo-transfer holder. Once properly inserted, platform should not rotate or rock

c. Place the protective cover on the top of the platform

d. Wait for the cryo-holder to cool down below 140K (step 1g). Once the cryo-holder is ready, start adding nitrogen to the cryo-transfer platform by using the small diameter funnel (see figure 1 under 'Mounting Tools' section). Overall assemble should look like picture 2d. Wait for the support platform to reach cryo-genic tempeature

3. Remove Elsa holder from the dry pumping station (see figure 1a of the holder in the pumping station) and install the cryo-holder in the cryo-transfer station by following the step below:

a. Ensure that the V4 and V3 valves (see labels of figure 1a) are closed. If not, close these valves first (close V4 and then V3) by rotating them clockwise. Do not overtighten the valves, finger tight is good enough. Remove hose from below valve V4 if necessary

b. Close V1 and then V2 (see labels in figure 1b for the position of the valves which are shown open). Rotate each valve clockwise at 90 degrees. Closed is in the horizontal position

c. Close the shield of the holder by rotating fully clockwise the knob located at the center of the dewar of the cryo-holder (label 3c in figure 3f)

d. Turn the dry pumping station off. The on/off switch is at the back of the unit next to the power cord

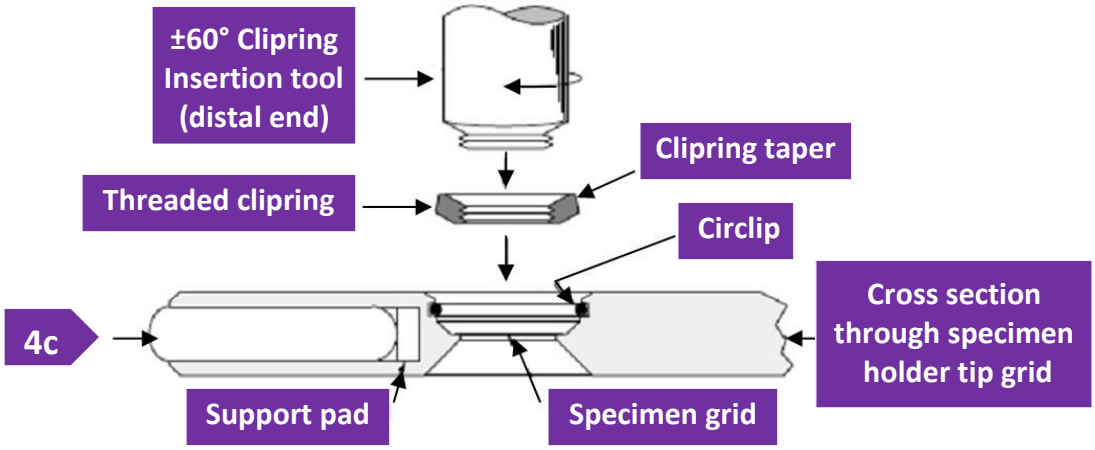
e. Once the 'MDP status' and 'System Status' light indicator in the front of the station are turned off, retract holder from dry pumping station

f. Transfer carefully the Elsa holder to the Elsa cryo-transfer station (figure 3f). The tip of the cryo-holder should secure into the support platform. A properly inserted cryo-holder in the support platform of the

cryo-transfer station is shown in figure 3g. You must ensure that the tip of the cryo-holder is secured into the support platform, as shown on figure 3g. Otherwise, the tip will get damaged/bent

g. Accessories of the cryo-station (see figure 2 under 'Mounting Tools' section) are located in the dehydrator which is on the prep table. The support platform contains a round reservoir for one grid box (see figure 3g) and a wide reservoir for work under liquid nitrogen. Additionally, there is a fill port for liquid nitrogen

4. Transfer sample to the Elsa holder
   a. Open the shield of the holder by rotating fully counterclockwise the knob located at the center of the dewar of the cryo-holder (label 3c in figure 3f)
   b. [Optional] Once the holder is at around 140K, you can unplug it from the temperature controller and move the Elsa with its cryo-transfer station over to the general bench
   c. Remove the clip ring from the holder tip (figure 4c) using the clip tool (see figure 1 under 'Mounting Tools' section). Place the silver end of the tool onto the clip ring and while applying a small amount of vertical pressure on the end of the clip ring tool, slowly turn the tool clockwise 1 - 2 turns until you feel resistance. Clip tool must be exactly vertical for the clip ring tool's thread to engage the thread of the clip ring



   d. Be careful not to turn too far as it will over-tighten the clip ring to the clip ring tool. Pull up to remove the clip ring. Transfer the cold sample grid to the tip of the holder using a pair of tweezers
   e. Secure the grid with the clip ring. With the clip ring still at the tip of the clip tool, insert the clip ring back into the grid holder by gently pushing down on it until you feel/hear a click. That click is the clip ring passing the circlip and getting secured underneath. While applying a small amount of vertical pressure on the end of the clip ring tool, slowly turn the tool counterclockwise 1 - 2 turns until the tool moves freely. Pull up to remove the clip ring tool.

f. Close the shield of the cryo-holder by rotating the knob fully clockwise (label 3c in figure 3f)

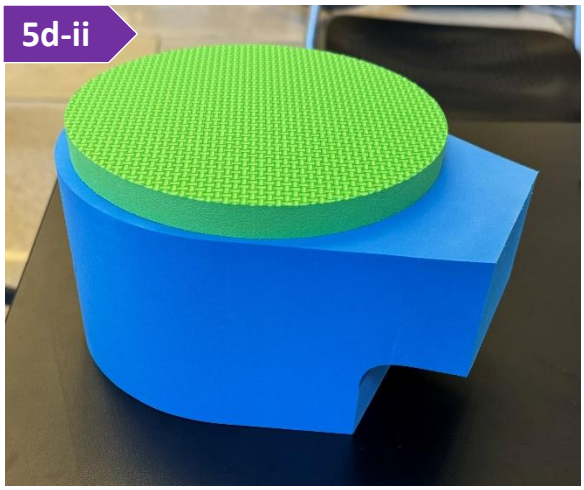
5. Insert Elsa holder into the microscope

   a. Tilt the goniometer to 45 degrees and insert the cryo-stage in the same way as you do for the room temperature holder, i.e., press on the [Go a] button (figure 5a). See details under the 'Manual Collection' sections

   b. Using the touch screen on the controller of the cryo-holder, set the target temperature by:
      i. Pressing on the [Mode] button
      ii. Pressing on the [Automatic] button
      iii. Pressing on the button that shows the temperature and typing the desired temperature value
      iv. Pressing on the [Start] button

   c. Open the shield of the cryo-holder by rotating the knob fully counterclockwise (label 3c in figure 3f)

   d. Before data collection start the drying process for the cryo-tools by:
      i. Putting tools/accessories in dehydrator. Ensure that the dehydrator is running by pressing on the 'Temp/Time' button multiple times until the temperature value is visible, and then on the 'Start/Stop' button. Temperature should not exceed 100 F
      ii. Dumping out all excess liquid nitrogen from the cryo-transfer station. Use the foam cryo-cup (figure 5d-ii) as a reservoir for the liquid nitrogen
      iii. Turning cryo-transfer station upside down, so that any condensed water drips out



   e. Start data collection (see details under the 'Manual Collection' sections). It might take 15-20 mins or more for the cryo-stage to thermally equilibrate at the target temperature. During the equilibration process, sample will drift, and you must wait until the drift is not significant before starting a measurement

   f. If icing of the sample grid is a constant issue, set the temperature at 170K while the sample and cryo-holder are in the column in the normal data collection position. Pumping time may range from 15-60 minutes depending on the amount of ice

   g. Once all data collections have been completed on the crystals of a grid, retract the Elsa holder from the microscope using a 45 degrees tilt on the goniometer

   h. Retraction of the cryo-holder is the same as the room temperature holder

6. If you are doing back-to-back measurements with the cryo-holder:

a. Prepare the cryo-transfer station as normal (step #2) while the cryo-holder is in the column. If certain tools are still wet, use the heat gun (figure 6a) to remove any moisture

b. Once the cryo-transfer station is cold, remove the cryo-holder from the column and install the cryo-holder in the cryo-transfer station as normal (step #3)

c. Transfer sample to the Elsa holder as normal (step #4)

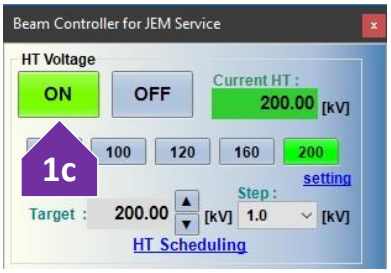
d. Insert Elsa holder into the microscope as normal (step #5)

7. Warm up and prepare the cryo-holder for the next user
   a. Insert holder into the dry pumping station and turn the pumping station on
   b. Dump out all excess liquid nitrogen from the dewar of the holder by spinning the holder upside down in the pumping station. Use the foam cryo-cup (figure 5d-ii) as a reservoir for the liquid nitrogen
   c. Once 'MDP status' and 'System Status' turn green, slowly open V2 and then V1. Leave V3 and V4 closed
   d. Attach the holder to the Elsa temperature controller
   e. Using the touch screen on the controller of the cryo-holder, run the 'Warm Up' procedure which takes 30 mins. Check the timer on the controller regarding the progress of the 'Warm Up' cycle
8. On Fridays or weekends, bake out the dewar and cryo-holder as follows:
   a. Insert holder into the dry pumping station and turn the pumping station on
   b. Dump out all excess liquid nitrogen from the dewar of the holder by spinning the holder upside down in the pumping station. Use the foam cryo-cup (figure 5d-ii) as a reservoir for the liquid nitrogen
   c. Once 'MDP status' and 'System Status' turn green, slowly open V2 and then V1
   d. Wait until most ice on the tip of cryo-holder has disappeared
   e. Connect the plastic vacuum hose that is already connected to V3 to the dewar
   f. Once most ice on the tip of cryo-holder has disappeared, slowly open V3 and wait for the vacuum reading to reach 10^{-5} Torr or lower. Make sure V3 is fully open
   g. Once 10^{-5} Torr is reached, slowly open V4
   h. Connect the cable from the temperature controller to the Elsa holder
   i. Using the touch screen on the controller of the cryo-holder, start the 'Bake Out' process
   j. The bake out process takes approximately 14 hours. Check the timer on the controller regarding the progress of the 'Bake Out' cycle

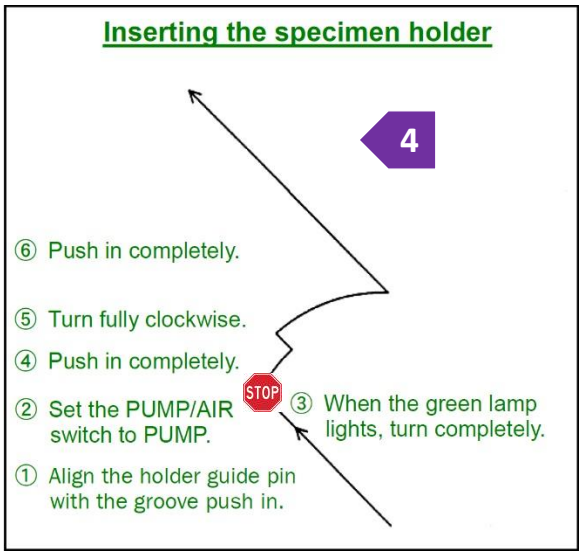
MANUAL COLLECTION

Please follow the steps below for a manual collection on a grain-by-grain workflow. For an automatic collection of multiple grains, check the 'Automatic collection/Queueing' section.

1. If you are the first user of the day:
   a. Take out the ACD heater from the anticontamination device (figure 1a)
   b. Fill the ACD dewar with LN_2. Use the white funnel (see figure 1 under 'Mounting Tools' section) and the 4 L dewar by the preparation desk. If the 4 L dewar is empty, refill it in the dry ice room MG91 (across receiving)
   c. Start your reservation on NUcore and turn the high voltage on using the TEM center software (no CAP) shown on the right monitor. Press on the [ON] button on the 'Beam Controller for JEM Service' window (figure 1c). If the 'Beam Controller for JEM Service' window is not visible, press on the 'Control' menu and select 'Beam'



2. If you are not the first user of the day, top off the ACD with LN_2
3. Launch CrysAlisPro (CAP) if it is closed and wait for a few seconds until the software initializes all connections to the hardware. A few electronic beeping sounds will occur during the connection process
4. Insert the holder with the sample grid into the column by (figure 4):
   a. Aligning holder guide pin with the guide notch on the goniometer and push the holder till it stops
   b. Keep pushing gently on the holder and do not rotate
   c. Pull on the PUMP/AIR switch below the goniometer, turn the switch up to the 'PUMP' position (the orange LED turns on, figure 4c)
   d. Wait till the 'EVAC Ready' LED turns green (figure 4d). Usually takes 1-2 minutes



e. Turn the holder 15 degrees clockwise until it moves forward a little by itself

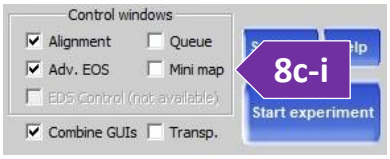
f. Once the little step forward has taken place, slowly turn the holder clockwise and let it slide all the way in gently by holding it back a little. Do not let it slam in. Controlled rotation is the only motion needed, do not move the holder sideways.

5. Press the [ED] button at the top right of the CAP window (figure 5). The 'JEOL microED Controller' window will appear

6. On the 'JEOL microED Controller' window (figure 6), ensure that all vacuum related textboxes are green otherwise the next option will not be enabled. ***Ensure that the 'Column Vacuum' reading is below 30 before you go to the next step.*** It might take 2+ minutes for the column vacuum to reach proper vacuum level

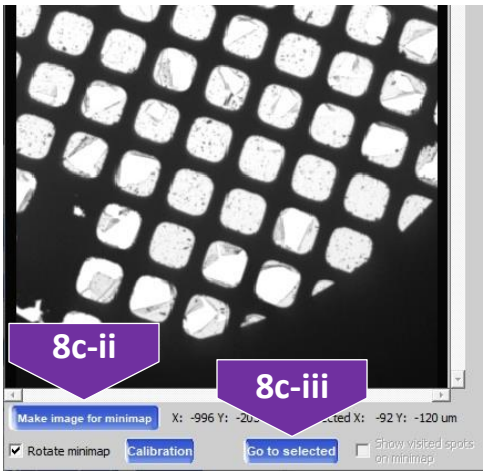
7. Turn current emission on by pressing on the [Turn filament ON] button (figure 7)

8. Locate sample grains by launching the 'Visual Mode':



a. Press the [START/STOP] button in the upper right corner of the main CAP window (figure 5a)

b. Select [Run Visual Mode] to launch the Visual (real space) mode. A real space image of the currently centered region of the grid will be displayed in the main window of CAP. If the image is black, most likely the beam is on the grid and/or in case you use a cryo-holder the shield is still closed (check the 'Using the cryo-holder' section for more details). Left clicking on a point on the image will automatically move that point under or near the central crosshair. An additional click may be needed to complete the motion of the point exactly under the crosshair. The region selected for diffraction data collection is shown as a circle around the crosshair.



c. Search for crystals using the trackball or mouse click. To see a larger area of the sample grid:

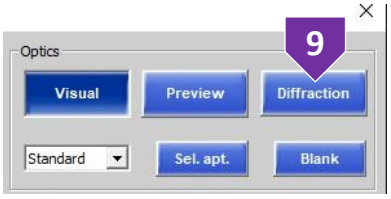
i. Tick the 'Mini map' option (figure 8c-i) at the bottom right side of the 'MicroED/3DED Control' window to show the mini map window

ii. Press on the [Make image for minimap] button to take a low magnification picture of a large fraction of the grid. It will take a few seconds for the system to create the minimap (figure 8c-ii)

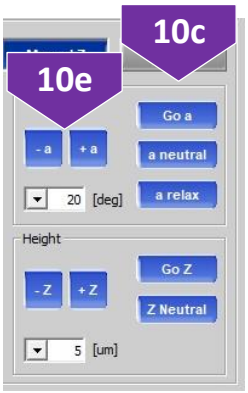
iii. To go to a specific spot shown on the minimap, left-click on the spot (no visual indication will appear yet) and press on the [Got to selected] button (figure 8c-iii). Stage will move to that spot

d. Look for crystallites that are isolated, nothing directly on the left or right of them, and not too close to the edge of the grid if a full goniometer rotation is needed. In any other case, the angles of rotation will be limited by the obstacles around the crystal of interest. Once an appropriate crystal is found, click the center of the crystal to bring the crystal near the crosshair, within the red circle (figure 8d)

9. Before spending time on aligning the crystal, you could switch to diffraction mode and verify that the crystal diffracts well. If this is the very first grain tested on the sample grid, follow the 'Aligning Apertures' session. Check apertures is typically done only once per sample grid. To switch to diffraction mode, under the 'MicroED/3DED control' window (figure 9), press on the [Diffraction] button. If the diffraction quality is not good, switch to 'Visual Mode' and search for another crystal



10. If diffraction quality is good, switch to 'Visual Mode' and perform the full eucentric centering:
    a. Before collecting diffraction data under constant rotation, the height of the sample stage must coincide with the rotation axis height. As in an X-ray diffractometer, this is achieved by minimizing the shift/movement of the crystal when the tilt angle is changed.
    b. Move the selected crystal to the center of the crosshair by left clicking on the crystal
    c. Click on the [a neutral] button (figure 10c) to relax the stage. If the crystal is not in the center of the crosshair after relaxation, repeat the alignment step above. Once the alignment looks okay, proceed to the next step and do not adjust the XY anymore (no left click)
    d. Specify the desired tilt angle step. For the very first crystal on a new sample grid, a very small step is recommended, e.g., 5 degrees. For later crystals, once the eucentric is aligned, a higher step can be selected
    e. Tilt the stage by pressing the [-a] or [+a] buttons (figure 10e). Stage will tilt by the number of degrees set in the dropdown menu below the [-a] or [+a] buttons. The tilt angle of the stage will be reflected by the indicator at the bottom left corner of the main display window
    f. If the crystal height is not at the center of rotation, the apparent crystal position will shift to the left or right after the stage is tilted. Adjust the stage Z position:
        i. By clicking the mouse wheel button (middle click) at the center of the crystal



ii. Multiple repeated clicks may be required due to stage relaxation effects

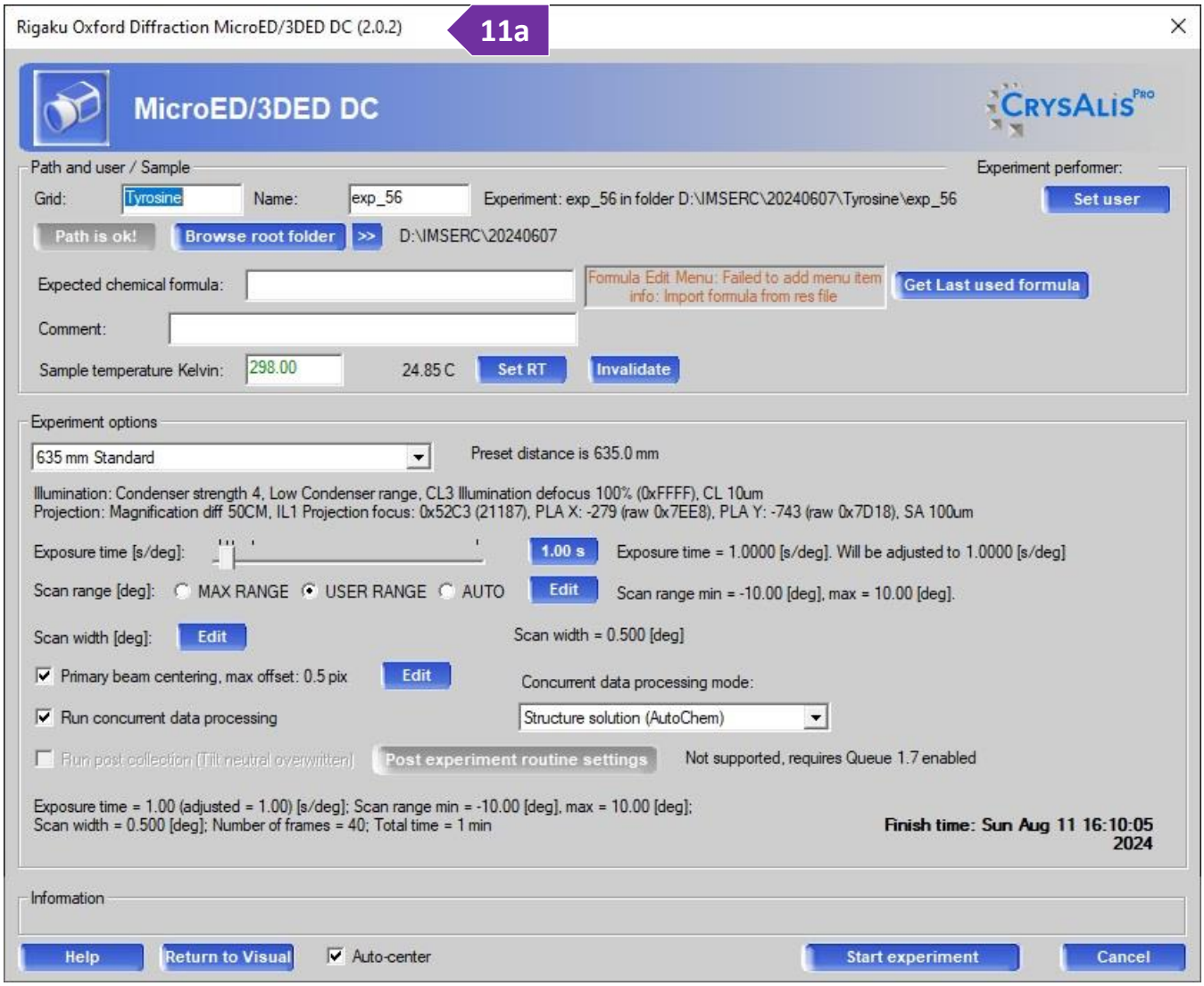
iii. Click the [a relax] button to change the tilt angle of the stage

iv. If the crystal position shifts during rotation by an amount exceeding approximately half the diameter of the central circle (diffraction area), repeat the procedure of the Z-alignment

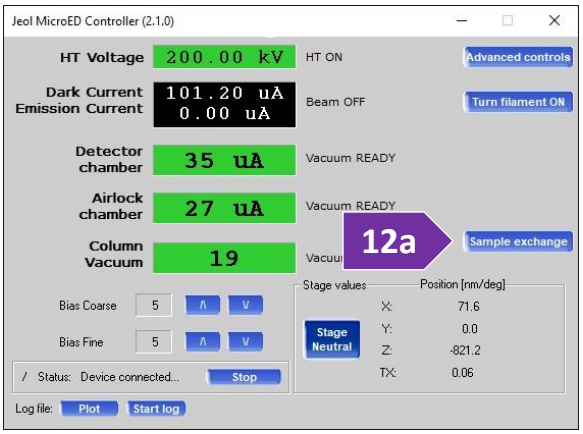
v. Check Z-alignment for the final tilt angle by pressing the [Go a] button and typing the desired angle. You need to repeat this step for both positive and negative angle limits to ensure that either part of the grid or other grains are not in the beam. Remember these values for the lower and upper limit of rotation as you will need them for the data collection

11. Once you are done with the eucentric alignment and have defined the minimum and maximum rotation limits for the rotation, you can perform the data collection:

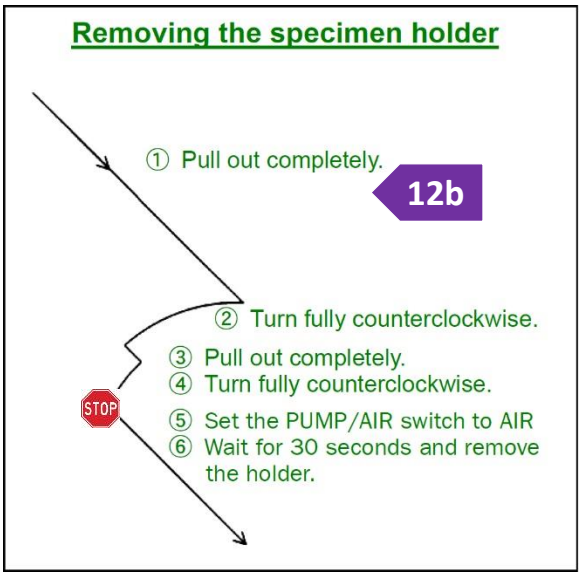
a. Press on the [Start experiment] button on the 'MicroED/3DED' Control window to show the data collection window (figure 11a)



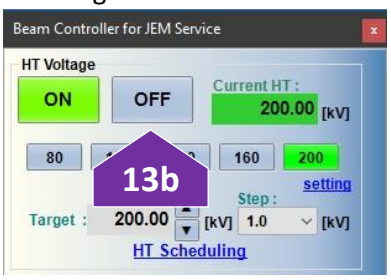
b. Enter the experiment conditions:
   i. Enter the 'Grid' name and 'Name' of the experiment/collection. CAP will automatically create a directory with the grid name and another with the experiment name. Ensure that you are saving the data under your personal folder shown next to the 'Name' field. To change the destination folder, press on the 'Browse root folder' button
   ii. Provide the chemical formula or expected chemical content that will be used by the auto solution program
   iii. Provide the sample temperature
   iv. Next to the 'Exposure time' bar, press on the exposure button and set the exposure time, e.g., 1 s
   v. Next to the 'Scan range' label, press on the [Edit] button for the tilt angle range
   vi. Next to the 'Scan width' label, press on the [Edit] button for the scan width, e.g., 0.5
   vii. Check the 'Primary beam centering' option
   viii. Check the 'Run concurrent data processing'
   ix. Press on the 'Start experiment' button to start the data collection



12. To remove/exchange sample grid after your measurement:
   a. Press on the [Sample exchange] button on the 'JEOL MicroED/3DED Controller' window in CAP (figure 12a) and wait for the goniometer to go to the home position and for the filament emission to turn off
   b. Remove the sample holder by (figure 12b):
      i. Pulling the holder until is stops. You will feel some resistance, as the holder is in vacuum at this point
      ii. Turn it fully counterclockwise until it stops
      iii. Pull it a bit until in stops
      iv. Turn it fully counterclockwise and stop
      v. Set the PUMP/AIR switch to the 'AIR' position and wait until the green LED turns off
      vi. Remove the holder from the goniometer
      vii. If you are measuring more samples, load a new sample grid and restart the process



viii. If you are done for the day and there is another user after you, remove the grid from the holder and leave the holder on the preparation table



13. If you are the last user of the day
    a. Ensure that there is no holder in the column
    b. Turn the high voltage off using the TEM center software (no CAP) shown on the right monitor. Press on the [OFF] button on the 'Beam Controller for JEM Service' window (figure 13b). If the 'Beam Controller for JEM Service' window is not visible, press on the 'Control' menu and select 'Beam'

    c. Retract the aperture on the condenser lens (CL) using the TEM center software (no CAP) shown on the right monitor. Press on the [OPEN] button on the 'Aperture Panel Controller' window (figure 13c). If the 'Aperture Panel Controller' window is not visible, press on the 'Control' menu and select 'Aperture'

    d. Retract the selected area (SA) aperture using the TEM center software (no CAP) shown on the right monitor. Press on the [OPEN] button on the 'Aperture Panel Controller' window (figure 13d)

    e. Install the ACD heater in the liquid nitrogen trap (figure 13e). The orange light above the switch on the mounting base of the heater should be always on as show in figure 13e
    f. Turn on the 'ACD' heater using the TEM center software (not CAP) shown on the right monitor. If the 'ACD Controller' window is not visible (figure 13f), press on the 'Control' menu and select 'Maintenance' and then 'ACD'

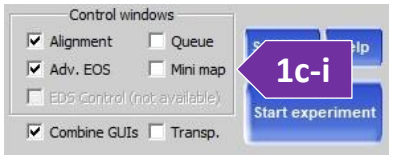
    g. Acknowledge that the apertures have been retracted (steps 13c and 13d)
    h. Update the second calendar in NUcore named 'XRD ED-Synergy ACD', so that all users are aware that the instrument is under heating and not available for at least 4 hours
    i. End your reservation in NUcore using the standard calendar named 'XRD ED-Synergy'

AUTOMATIC COLLECTION/QUEUING

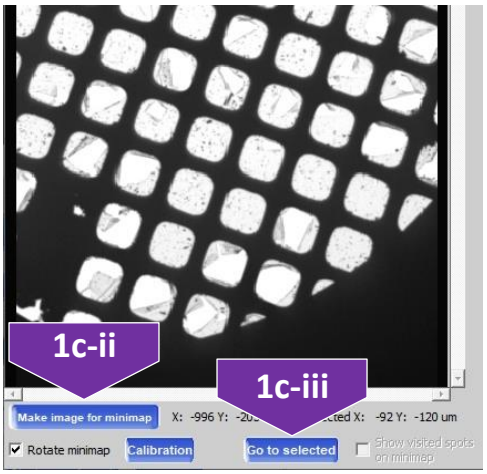
Instead of measuring diffraction data from a given crystal immediately after screening and centering, data collection (screening or full experiment) of multiple crystals can be automated by creating a queue. Once the queue is populated with several experiments, all data collections can be run sequentially and unattended. To use the queue function:

1. Perform the basic centering steps as described in the 'Manual Collection' section. Locate sample grains by launching the 'Visual Mode':

   a. Press the [START/STOP] button in the upper right corner of the main CAP window (figure 1a)
   b. Select [Run Visual Mode] to launch the Visual (real space) mode. A real space image of the currently centered region of the grid will be displayed in the main window of CAP. If the image is black, most likely the beam is on the grid and/or in case you use a cryo-holder the shield is still closed (check the 'Using the cryo-holder' section for more details). Left clicking on a point on the image will automatically move that point under or near the central crosshair. An additional click may be needed to complete the motion of the point exactly under the crosshair. The region selected for diffraction data collection is shown as a circle around the crosshair.

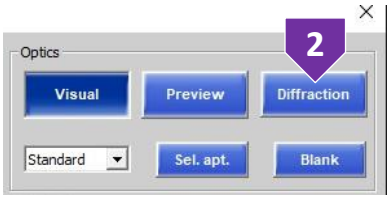


   c. Search for crystals using the trackball or mouse click. To see a larger area of the sample grid:
      i. Tick the 'Mini map' option (figure 1c-i) at the bottom right side of the 'MicroED/3DED Control' window to show the mini map window
      ii. Press on the [Make image for minimap] button to take a low magnification picture of a large fraction of the grid. It will take a few seconds for the system to create the minimap (figure 1c-ii)
      iii. To go to a specific spot shown on the minimap, left-click on the spot (no visual indication will appear yet) and press on the [Got to selected] button (figure 1c-iii). Stage will move to that spot

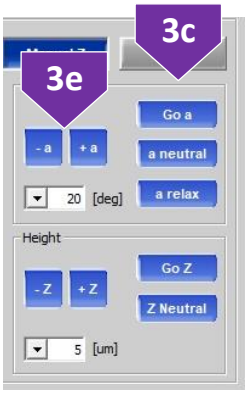


   d. Look for crystallites that are isolated, nothing directly on the left or right of them, and not too close to the edge of the grid if a full

goniometer rotation is needed. In any other case, the angles of rotation will be limited by the obstacles around the crystal of interest. Once an appropriate crystal is found, click the center of the crystal to bring the crystal near the crosshair, within the red circle (figure 1d)

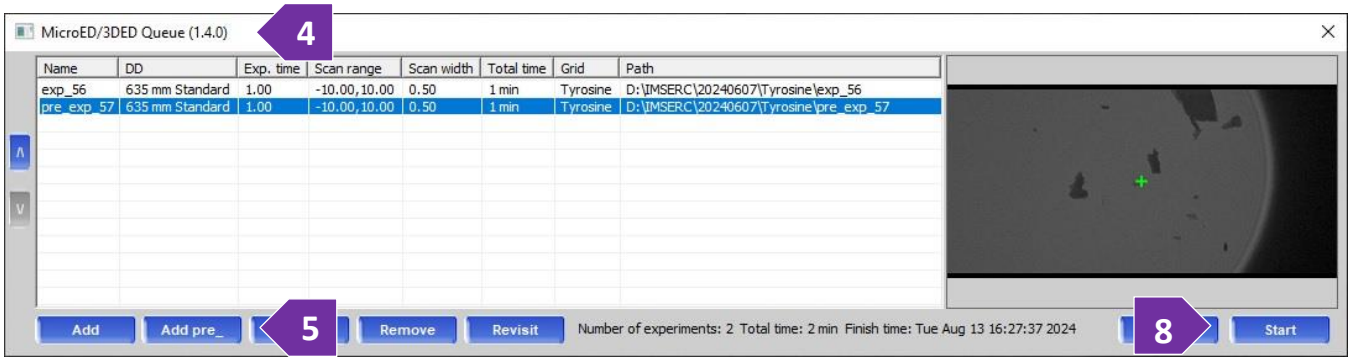


2. Before spending time on aligning the crystal, you could switch to diffraction mode and verify that the crystal diffracts well. If this is the very first grain tested on the sample grid, follow the 'Aligning Apertures' session. Check apertures is typically done only once per sample grid. To switch to diffraction mode, under the 'MicroED/3DED control' window (figure 2), press on the [Diffraction] button. If the diffraction quality is not good, switch to 'Visual Mode' and search for another crystal
3. If diffraction quality is good, switch to 'Visual Mode' and perform the full eucentric centering:
   a. Before collecting diffraction data under constant rotation, the height of the sample stage must coincide with the rotation axis height. As in an X-ray diffractometer, this is achieved by minimizing the shift/movement of the crystal when the tilt angle is changed.
   b. Move the selected crystal to the center of the crosshair by left clicking on the crystal
   c. Click on the [a neutral] button (figure 3c) to relax the stage. If the crystal is not in the center of the crosshair after relaxation, repeat the alignment step above. Once the alignment looks okay, proceed to the next step and do not adjust the XY anymore (no left click)

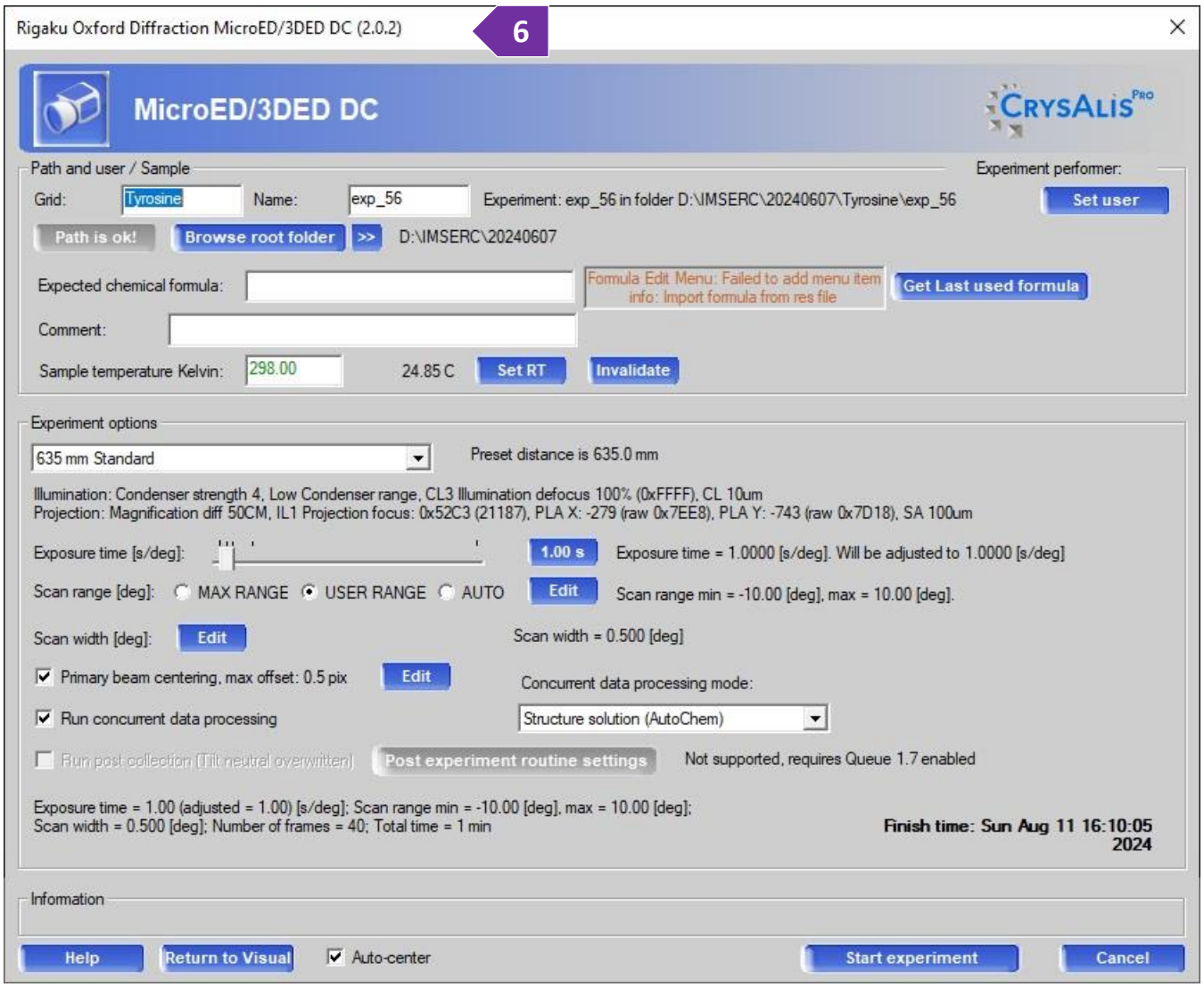


   d. Specify the desired tilt angle step. For the very first crystal on a new sample grid, a very small step is recommended, e.g., 5 degrees. For later crystals, once the eucentric is aligned, a higher step can be selected
   e. Tilt the stage by pressing the [-a] or [+a] buttons (figure 3e). Stage will tilt by the number of degrees set in the dropdown menu below the [-a] or [+a] buttons. The tilt angle of the stage will be reflected by the indicator at the bottom left corner of the main display window
   f. If the crystal height is not at the center of rotation, the apparent crystal position will shift to the left or right after the stage is tilted. Adjust the stage Z position:
      i. By clicking the mouse wheel button (middle click) at the center of the crystal
      ii. Multiple repeated clicks may be required due to stage relaxation effects
      iii. Click the [a relax] button to change the tilt angle of the stage
      iv. If the crystal position shifts during rotation by an amount exceeding approximately half the diameter of the central circle (diffraction area), repeat the procedure of the Z-alignment

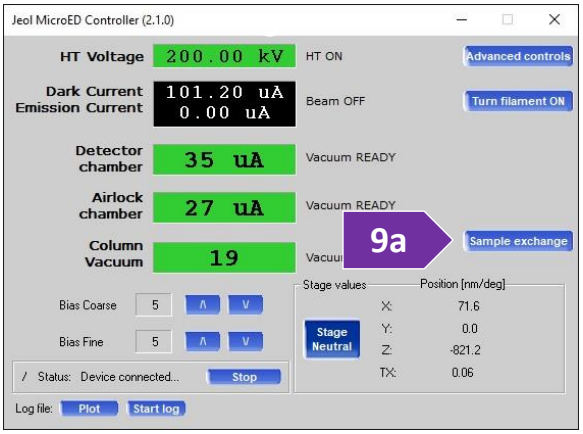
g. Check Z-alignment for the final tilt angle by pressing the [Go a] button and typing the desired angle. You need to repeat this step for both positive and negative angle limits to ensure that either part of the grid

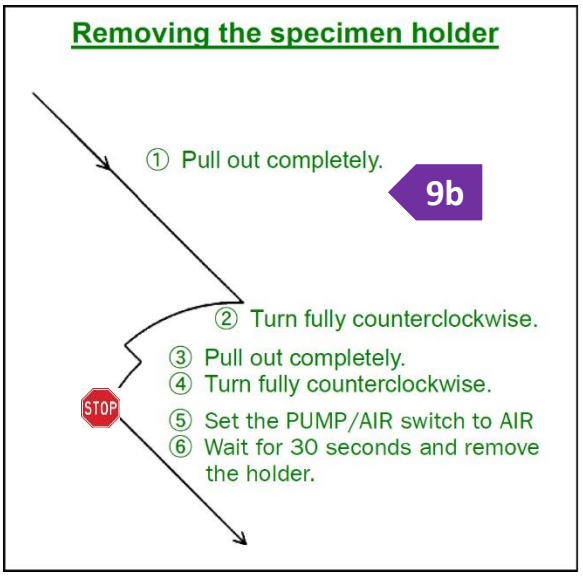


4. Enable the queue window by pressing on 'Queue' check option on the 'MicroED/3DED control' window. Queue window will appear (figure 4)
5. Instead of starting an experiment, press on the [Add] button (figure 5) on the 'MicroED/3DED Queue' window



6. The data collection setup window similar to that of a standard collection will appear (figure 6). Similar process can be followed for adding a pre-experiment by pressing on the [Add pre_] button. Enter the experiment conditions:
   a. Enter the 'Grid' name and 'Name' of the experiment/collection. CrysAlisPro will automatically create a directory with the grid name and another with the experiment name. Ensure that you are saving the data under your personal folder shown next to the 'Name' field. To change the destination folder, press on the 'Browse root folder' button
   b. Provide the chemical formula or expected chemical content that will be used by the auto solution program
   c. Provide the sample temperature
   d. Next to the 'Exposure time' bar, press on the exposure button and set the exposure time, e.g., 1 s
   e. Next to the 'Scan range' label, press on the [Edit] button for the tilt angle range
   f. Next to the 'Scan width' label, press on the [Edit] button for the scan width, e.g., 0.5
   g. Check the 'Primary beam centering' option
   h. Check the 'Run concurrent data processing'
7. After enqueueing, experiments can be reorders using the [Up] and [Down] arrow buttons located on the left side of the window
8. To start the queue, press on the [Start] button (figure 8) on the queue window
9. To remove/exchange sample grid after your measurement:
   a. Press on the [Sample exchange] button on the 'JEOL MicroED/3DED Controller' window in CAP (figure 9a) and wait for the goniometer to go to the home position and for the filament emission to turn off
   b. Remove the sample holder by (figure 9b):
      i. Pulling the holder until is stops. You will feel some resistance, as the holder is in vacuum at this point
      ii. Turn it fully counterclockwise until it stops
      iii. Pull it a bit until in stops
      iv. Turn it fully counterclockwise and stop
      v. Set the PUMP/AIR switch to the 'AIR' position and wait until the green LED turns off

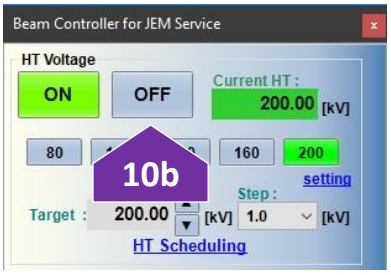




vi. Remove the holder from the goniometer

vii. If you are measuring more samples, load a new sample grid and restart the process

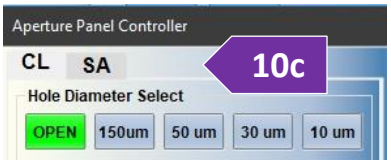
viii. If you are done for the day and there is another user after you, remove the grid from the holder and leave the holder on the preparation table



10. If you are the last user of the day

a. Ensure that there is no holder in the column

b. Turn the high voltage off using the TEM center software (no CAP) shown on the right monitor. Press on the [OFF] button on the 'Beam Controller for JEM Service' window (figure 10b). If the 'Beam Controller for JEM Service' window is not visible, press on the 'Control' menu and select 'Beam'

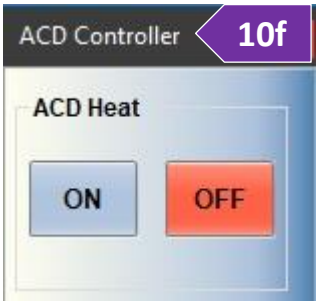


c. Retract the aperture on the condenser lens (CL) using the TEM center software (no CAP) shown on the right monitor. Press on the [OPEN] button on the 'Aperture Panel Controller' window (figure 10c). If the 'Aperture Panel Controller' window is not visible, press on the 'Control' menu and select 'Aperture'

d. Retract the selected area (SA) aperture using the TEM center software (no CAP) shown on the right monitor. Press on the [OPEN] button on the 'Aperture Panel Controller' window (figure 10d)

e. Install the ACD heater in the liquid nitrogen trap (figure 10e). The orange light above the switch on the mounting base of the heater should be always on as show in figure 13e

f. Turn on the 'ACD' heater using the TEM center software (not CAP) shown on the right monitor. If the 'ACD Controller' window is not visible (figure 10f), press on the 'Control' menu and select 'Maintenance' and then 'ACD'



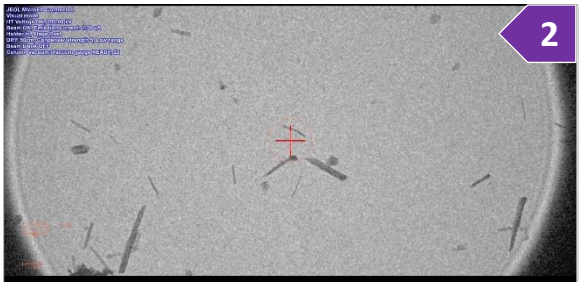
g. Acknowledge that the apertures have been retracted (steps 13c and 13d)

h. Update the second calendar in NUcore named 'XRD ED-Synergy ACD', so that all users are aware that the instrument is under heating and not available for at least 4 hours

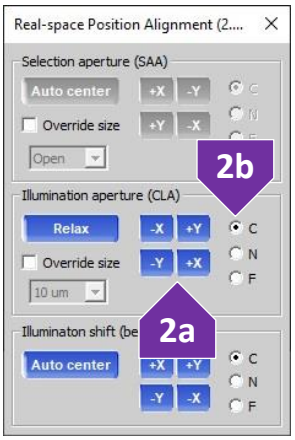
i. End your reservation in NUcore using the standard calendar named 'XRD ED-Synergy'

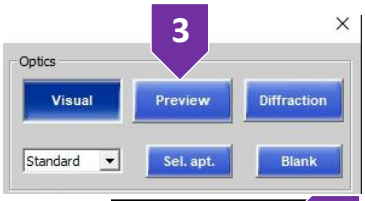
ALIGNING APERTURES

For every new grid, it is a good practice to check the alignment of the condenser lens aperture (CLA) used for real space, and the Selected Area aperture (SAA) used for diffraction. To perform/check the alignment, perform the following steps:

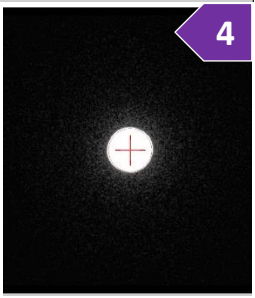


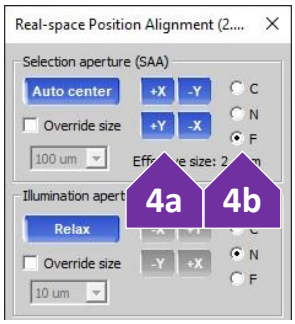
1. Start as normal by inserting your sample grid into the column, turning on the emission, and going into 'Visual mode'
2. The edges of the 10 µm CL aperture must be visible on the left and right sides of the screen (figure 2). If edges are not as shown in Figure 2, perform a CL aperture alignment by:
   a. Pressing on the [-X], [+Y], [-Y], and [+X] buttons under the 'Illumination aperture (CLA)' section of the 'Real-space Position Alignment' window (figure 2a)
   b. Adjusting the speed of aperture translation, if needed, by selecting the corresponding radio button for Course (C), Medium (N), and Fine (F) speeds (figure 2b). if the CLA aperture is jumping too much during the alignment process, press on the [Relax] button which is next to the [-X], [+Y], [-Y], and [+X] buttons
   c. Edges of the aperture do not have to be perfectly symmetric on the left and right sides





3. Check the alignment of the 100 µm SA aperture by switching to the 'Preview' mode by pressing the [Preview] button on MicroED/3DED control' window (figure 3)
4. The position of the SA aperture must be in the center of the screen as shown in Figure 4. If SA aperture is not in the center of the screen as shown in Figure 4, perform a SA aperture alignment by:
   a. Pressing on the [-X], [+Y], [-Y], and [+X] buttons under the 'Selection aperture (SAA)' section of the 'Real-space Position Alignment' window (figure 4a)
   b. Adjusting the speed of aperture translation, if needed, by selecting the corresponding radio button for Course (C), Medium (N), and Fine (F) speeds (figure 4b)
   c. Switch back to either 'Visual' or 'Diffraction' mode (figure 3) and resume your work



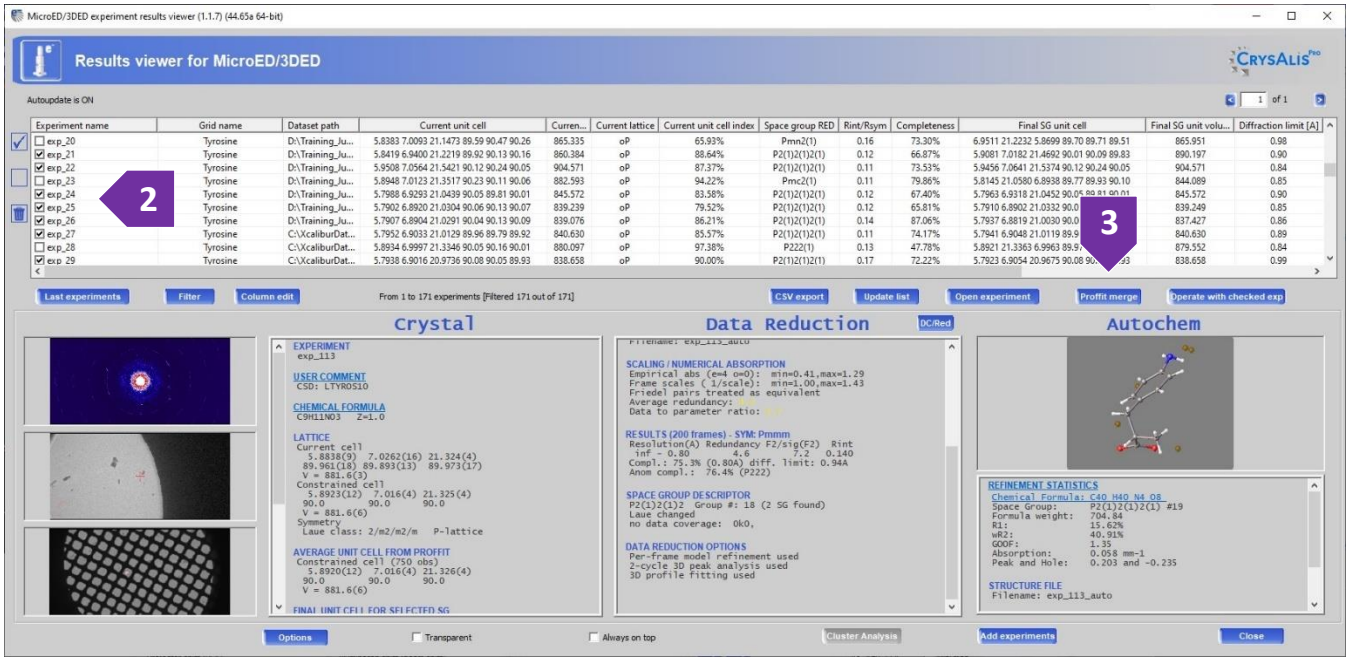


DATA REDUCTION

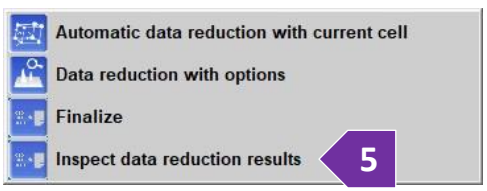
MERGING AND SCALING DATASETS

In electron diffraction for microED/3D ED collections, it is very likely that you'll have to merge and scale intensities of reflection collected from several crystals to optimize data completenss. The process of merging and scaling different data sets from the same sample includes the following steps:

1. Lauch the 'Results Viewer for MicroED/3DED' module by pressing on the  icon at the tool bar (bottom left side of CAP)
2. Select the datasets you'd like to merge by ticking the corresponding checkboxes in the list view (figure 2). You could use listing criteria for refining the list of visible datasets by pressing on the [Filter] button. Unit cells constants should agree as closely as possible. Reasonable criteria for merging selection are low Rint value, high unit cell indexing, and high diffraction resolution



3. Press on the [Proffit merge] button (figure 3) to open the 'Proffitmerge dialog' window which should contain the list of selected datasets. Additional experiments can be added or removed by pressing on the [Add] or [Delete] buttons on the 'Proffitmerge dialog' window, respectively
4. Set the name of the merged dataset using the 'Output file name' textbox at the bottom of the window, and press on the [Merge all files] button. This step will create the merged input file for the data reduction
5. On the main CAP window, press on the 'Data Reduction' tab, and select the 'Inspect data reduction results' option (figure 5)

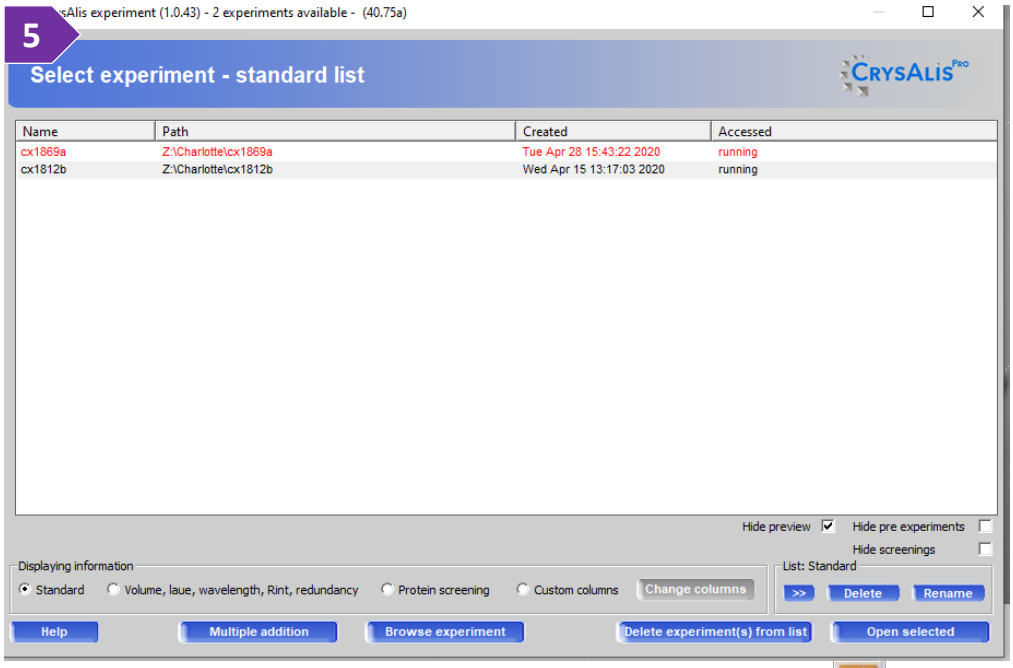


6. In the dropdown menu on the 'Data reduction results' window, select the file name of the merged collection, and press on the [Refinalize] button to initiate the re-finalization of the merged collection by launching the 'Finalization dialog' window
7. On the 'Finalization dialog' window, verify all parameters based on your conditions, and press on the [OK] button to start the finalization
8. Once the re-finalization is complete, the data reduction results for the merged dataset will be shown in the 'Data reduction results' window. If there are no further adjustments to be made, press on the [OK] button. The final merged data reduction output files are now available for the structure solution process
9. Press on the Olex2 button on the left hand side and select the INS file for the merged data. Then, proceed as usual for structure solution
10. Several combinations of different datasets may have to be attempted to get the best merged data and best structure. Also, it may be better to remove a bad dataset from merging if it worsens scaling results and structure refinements, even if completeness decreases a little

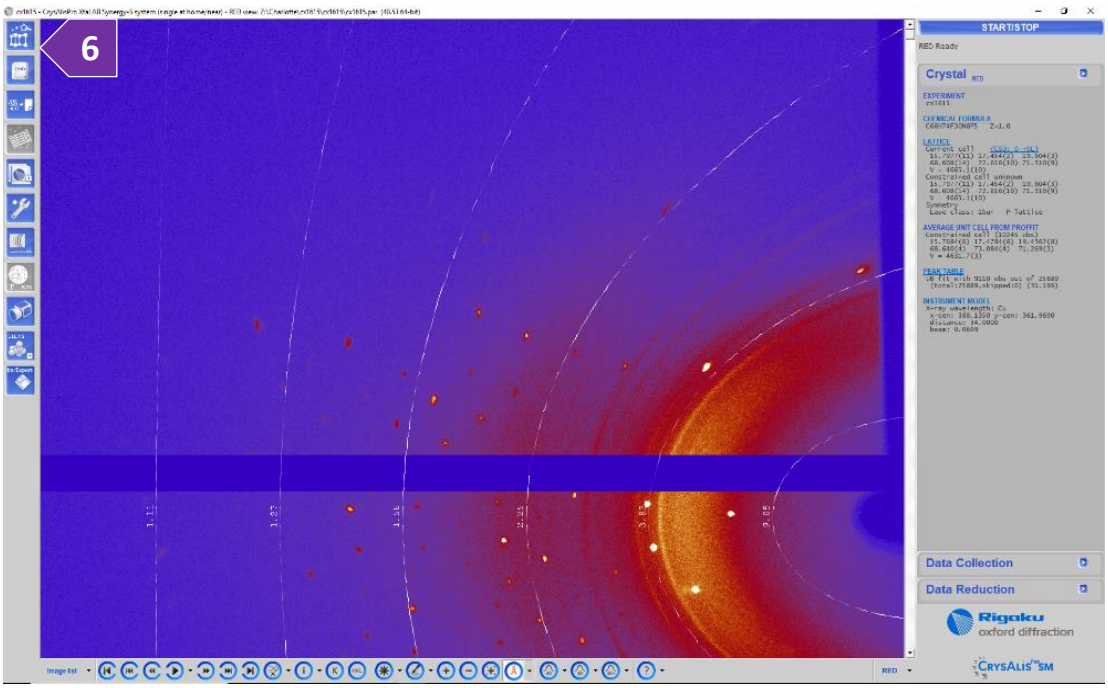
DATA INDEXATION

CAP automatically integrates data online, this section shows how to do it manually.

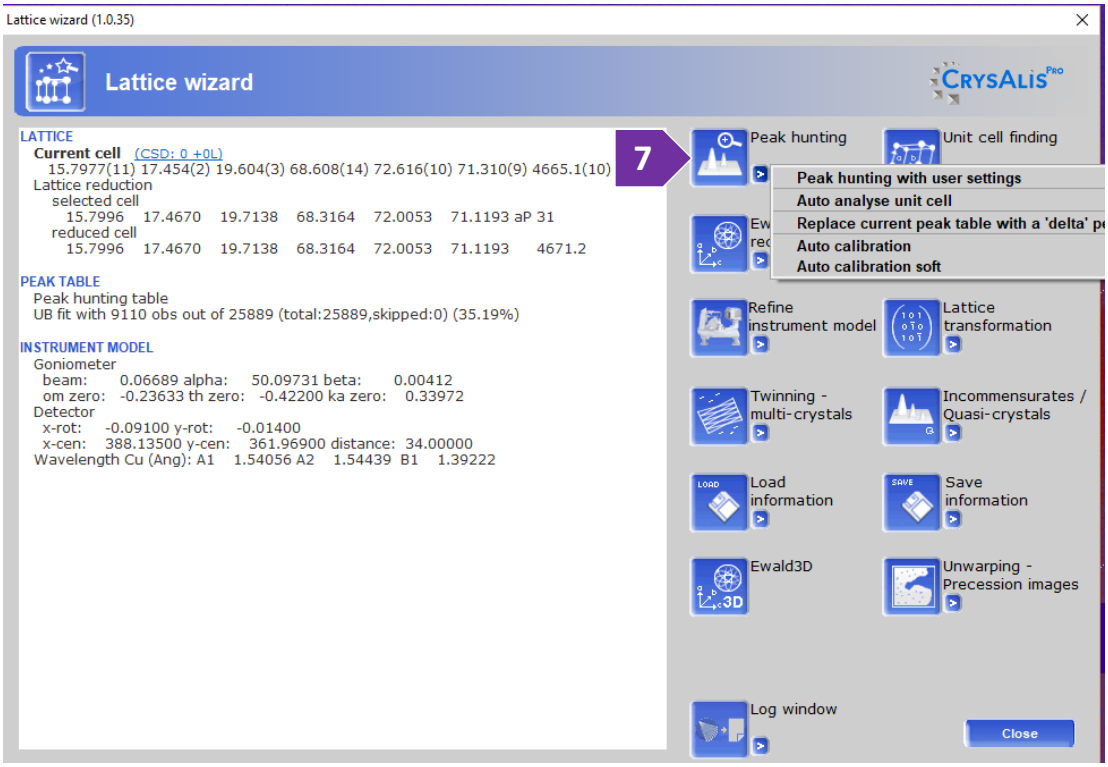
1. Copy the parent directory (i.e. the whole folder) for your data from the X-ray-live drive
2. Double click the CrysAlisPro (CAP) RED  shortcut icon on the desktop.
3. Double click the XXXXX.run file
4. Or in the Select Experiment menu, browse to your experiment. Select the XXXXX.par file.
5. Select the new "XXXXX" experiment file that has been created, and click "Open selected".



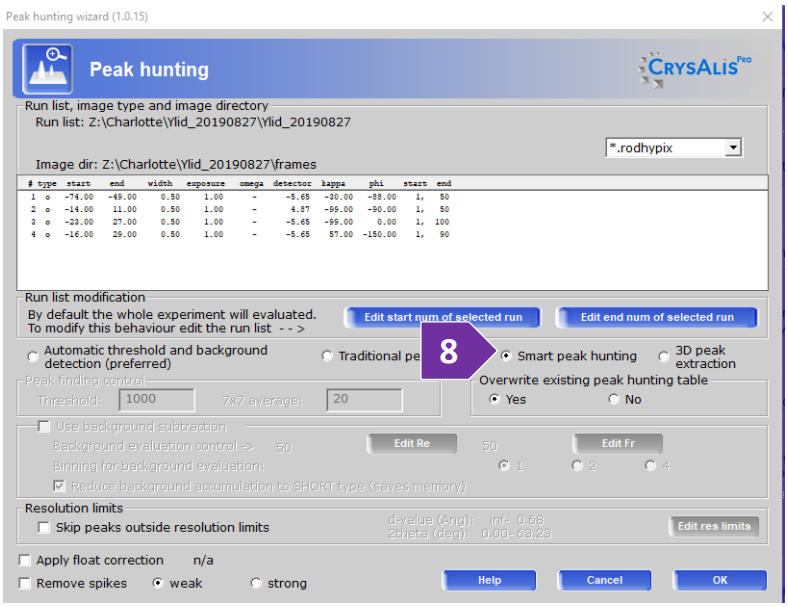
6. Your diffraction image should appear as below. Now select the "Lattice wizard"  button.



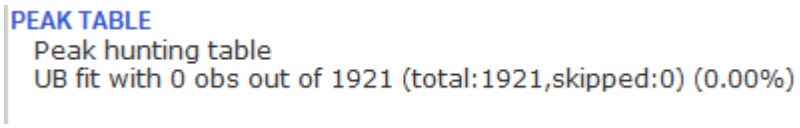
7. Click the “>” button under “Peak hunting”, and select “Peak hunting with user settings”.



8. In the next window, select “Smart peak hunting” and press ok.

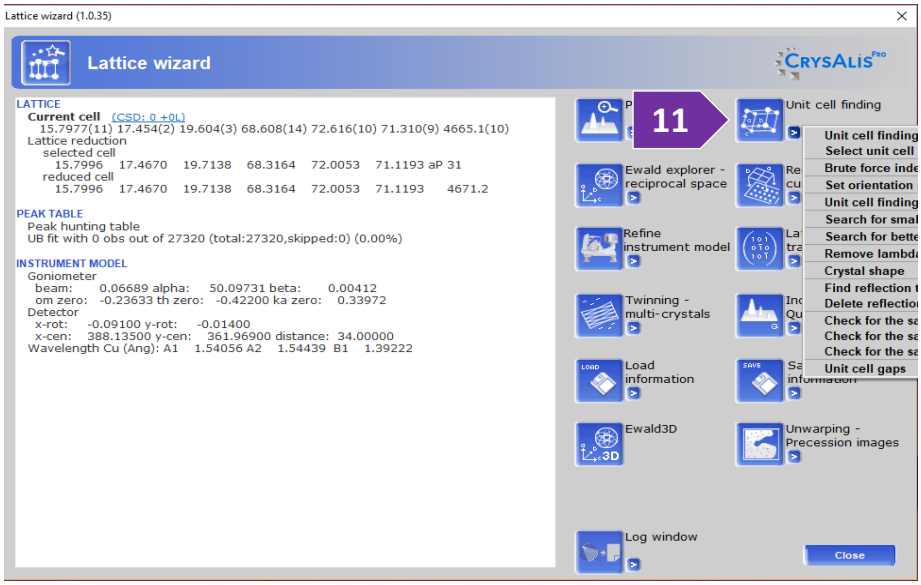


9. Say yes to overwriting the peak table. A window will pop up, and images will rapidly start to sequence as the peak hunting algorithm searches for diffraction peaks. You should see the strongest peaks being marked with "+"
10. At the end of the process, a unit cell is displayed, but 0% of the reflections have been fit to the unit cell. This

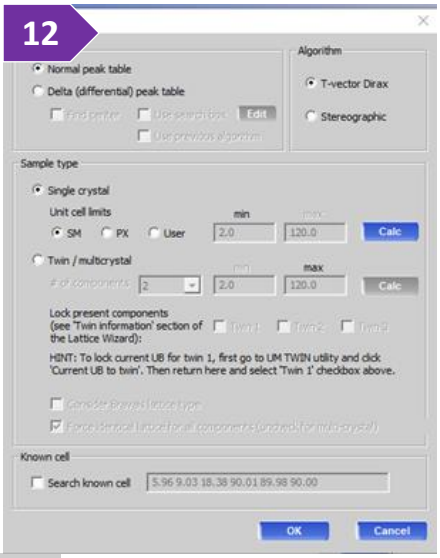


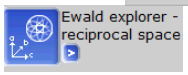
is because the displayed unit cell is from a previous peak hunt

11. Click the ">" button under "Unit cell finding", and select "Unit cell finding with options"

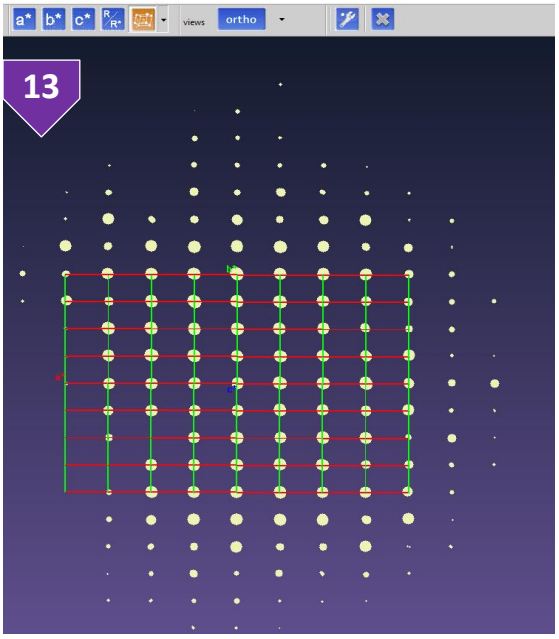


12. Ensure that “Normal peak table” and “T-vector Dirax” are selected. More importantly, “Single crystal” and “SM” should be selected in the Sample type filed. Here, you could also select “User” and set upper and lower bounds for unit cell parameters. For now, make sure your window looks like this image, and click “OK”



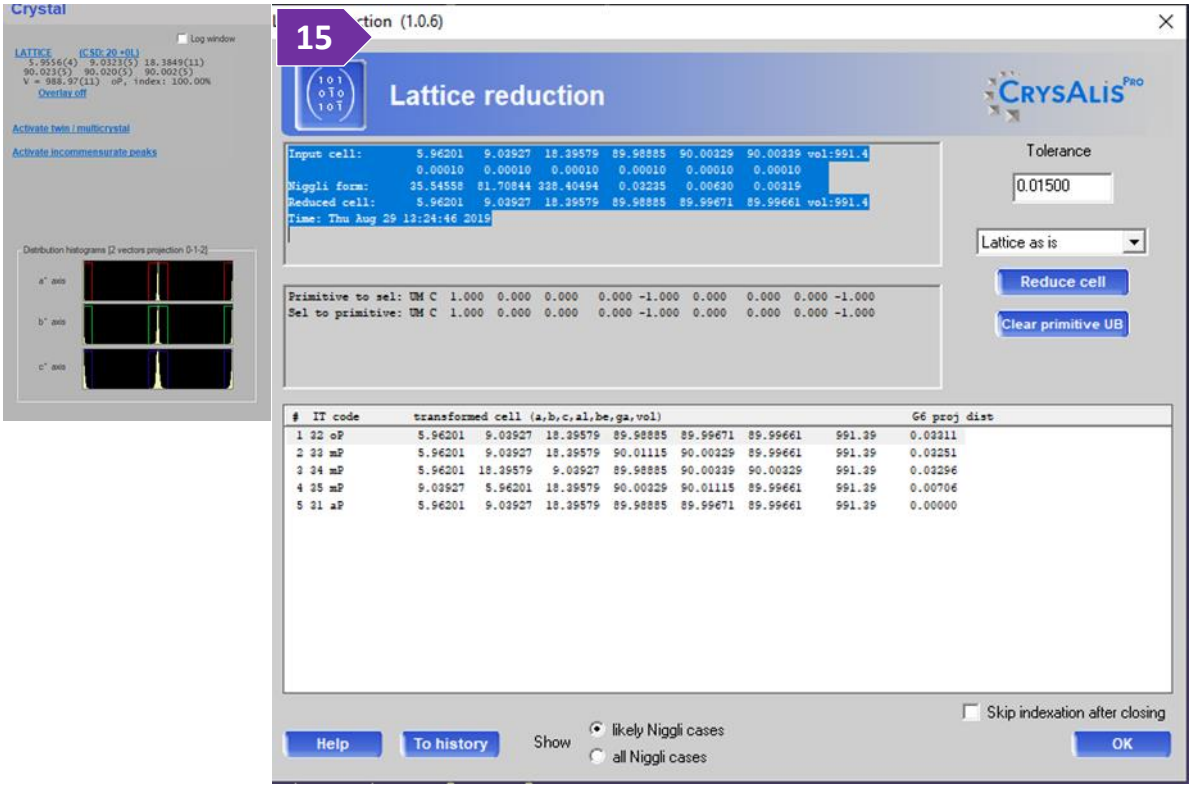
13. Click Ewald explorer 

and look at your reciprocal space. You may look down the different axis. You may turn the overlay on and off. If the cell is indexed properly, then the grid lines should be passing through reflections. If there are a lot of reflections NOT passing through the grid, or if there are grid lines NOT passing through reflections, then something is wrong.



14. You can look for twins here too and look at the histograms

15. From within the Ewald Explorer, check the predicting crystal system and lattice centering: Left click lattice and select “Modify lattice type”. A window appears listing possible unit cells from highest to lowest.



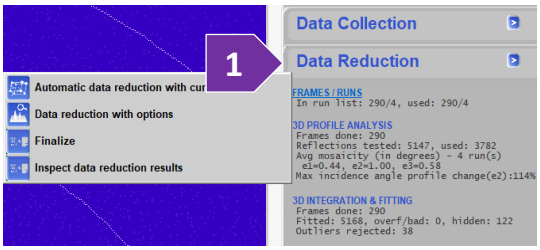
symmetry. The relative small figure of merit of 0.03 suggests that oP is correct. Click OK.

16. Close the Ewald viewer and Lattice Wizard.

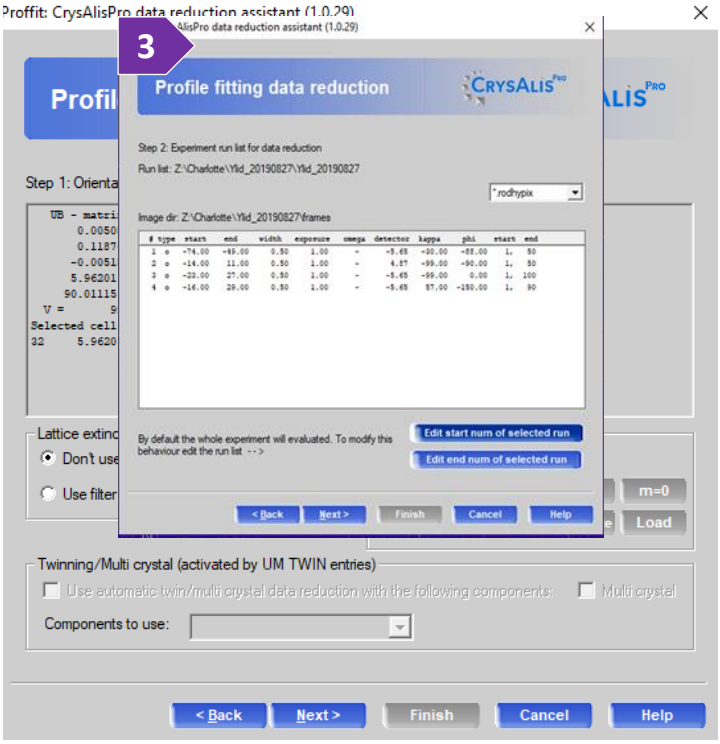
17. Click overlay spot prediction and play through your frames with the 10-foward button. Make sure the + marks are appearing on the peaks. Note: peaks marked ◊ and □ correspond to approaching and receding peaks, respectively. Click on to toggle the frame information on and off

DATA REDUCTION

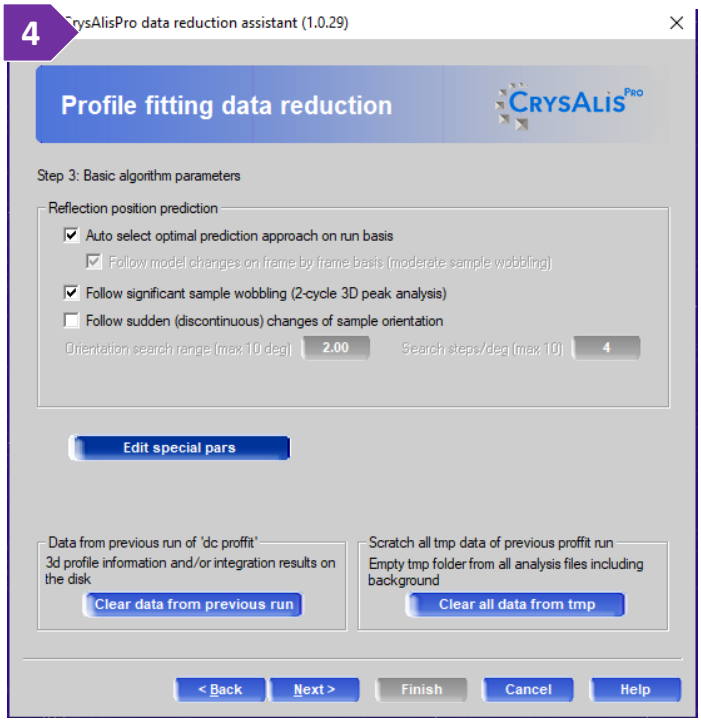
18. Click on “Data Reduction”, (this time not the carrot next to it). Click on it again and select “Data reduction with options”



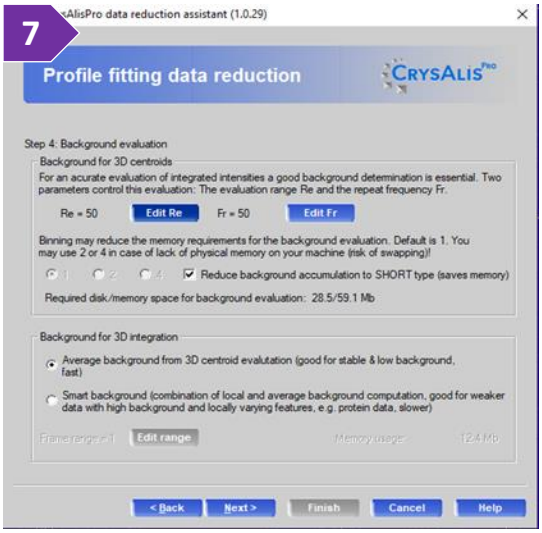
19. Ensure that don’t use filter is checked if lattice is not known. If the lattice is centered (A, B, C, I, F, R), click *use filter for*: and select the correct centering. Make sure Normal data reduction is checked and then click “Next”.



20. Check that CrysAlis[Pro] has found all runs. This list will dictate which images are integrated. If there were bad frames, omit them here. To delete bad frames, click on the run '#'. Then select edit start [end] number of selected run, depending on whether the bad frames are at the beginning or the end of the run. Change the start [end] number to exclude the bad frames. To omit an entire run, enter 0 (zero) as the start number. Press next.
21. You can change some parameters here. You can correct for sample wobble or sudden movement. Clear data from previous run and Clear all data from tmp. Make sure you click yes in warning windows.



22. Edit special pars “Use resolution limits”. Click “Edit limits” will enable you to reset your resolution.
23. Click “Edit high limit”, and enter “0.77” in the Editing high-resolution limit window. You should already have an idea on how far the crystal diffracts based on your unit cell analysis. Click “OK”, and then click “OK” on the “Resolution limits” window.
24. Click “Next” to Background evaluation window.



25. You can change to “Smart background” option for weaker data. Your window should appear as below.
    d. Re = # of frames used in background calculation
    e. Fr = How often calculation restarts.
    f. Make #s smaller if sudden changes between frames

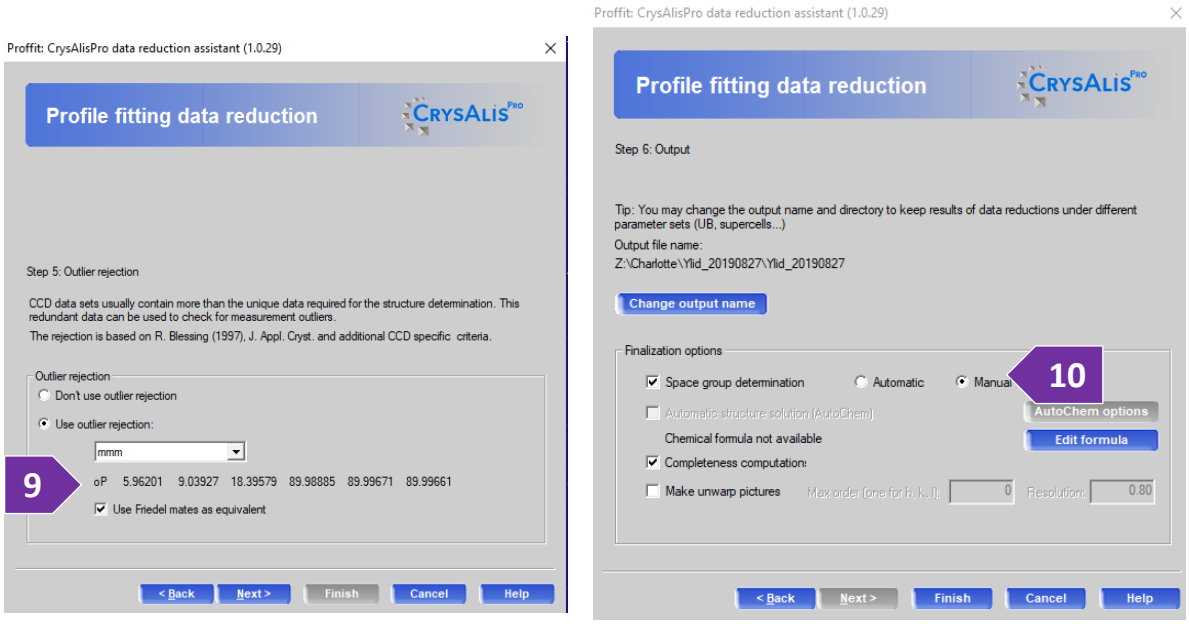
g. Background for 3D integration for average

h. Smart: weak data or Large variation w/in frame

i. Click “Next” to proceed.

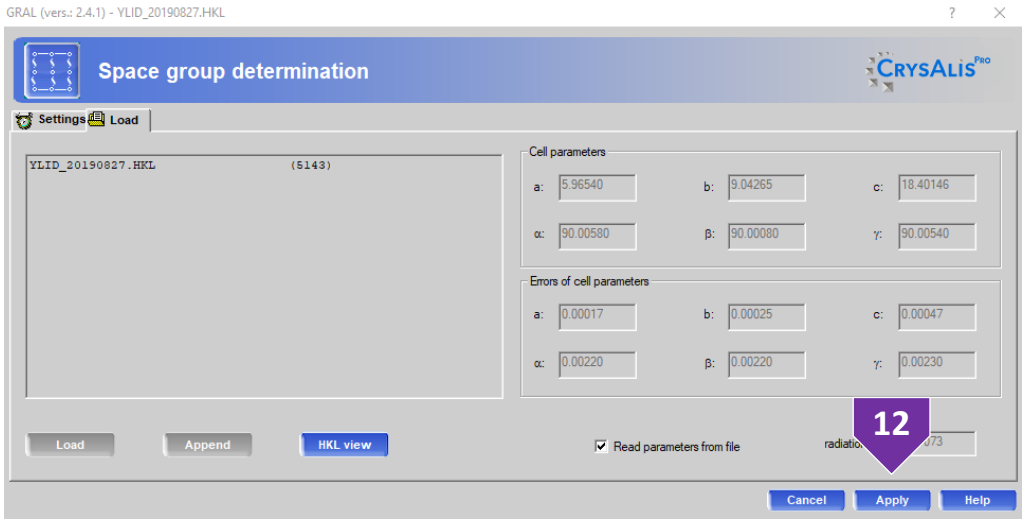
26. In the outlier rejection window, use the pull-down menu to choose the correct Laue group. Unless you know you have non-centrosymmetric space group make sure Use Friedel mates as equivalent is checked.

27. Change output name to something new, make sure that the Space group determination is set to “Manual”. Check the formula and Z.



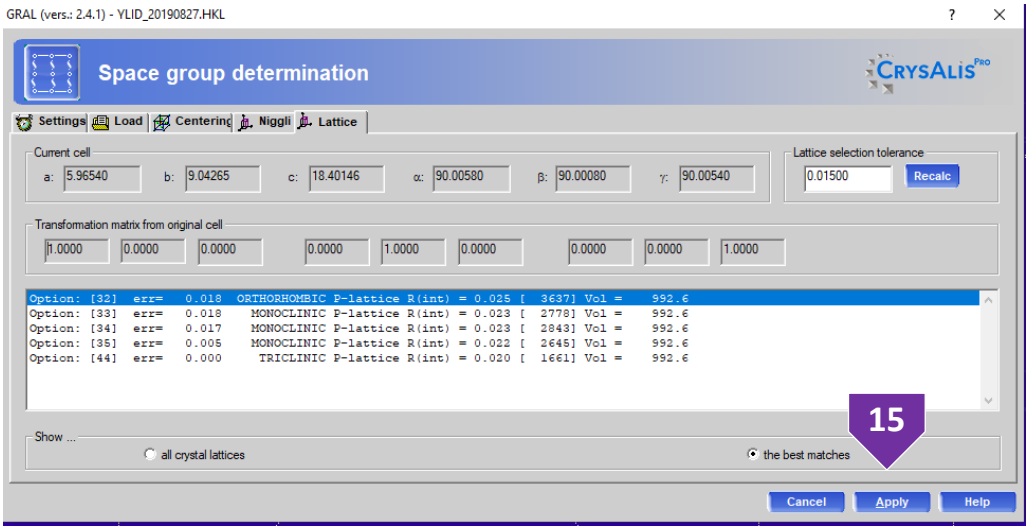
28. Click “Finish”, Watch the integration and monitor the output in the tab. The software runs through the dataset twice. First, the software locates the peaks (marked “+”) and develops a 3D peak profile. Second, the UB matrix and 3D profile are used to calculate the position and intensity of each reflection (3D integration & fitting). The peaks are marked by “integration masks” that give a visual idea of the size and possible overlap of the peaks.

29. Once the integration is finished, the GRAL window will pop-up (Like XPREP) and ask you to assign the space group. Click “Apply”.

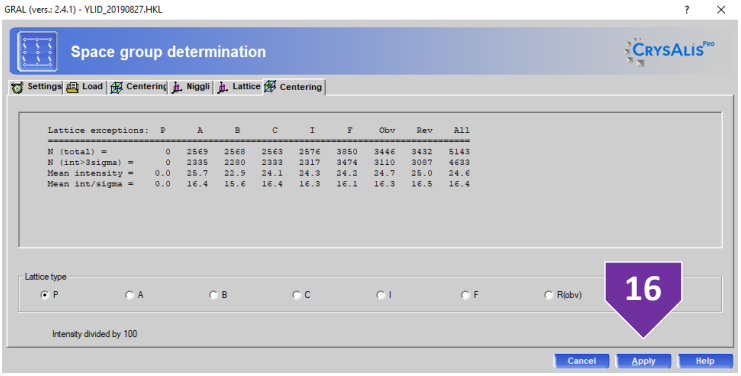


30. Investigate the centering absences. Recall that the first row corresponds to the total number of reflections you would expect to collect for the condition listed in the column. The second row indicates how many of the symmetry equivalent reflections that you collected violate that condition. This cell appears to be primitive. Click “Apply”.
31. The Niggli cell test will look for any unit cell transformation matrices that produce a reduced cell. Click “Apply”

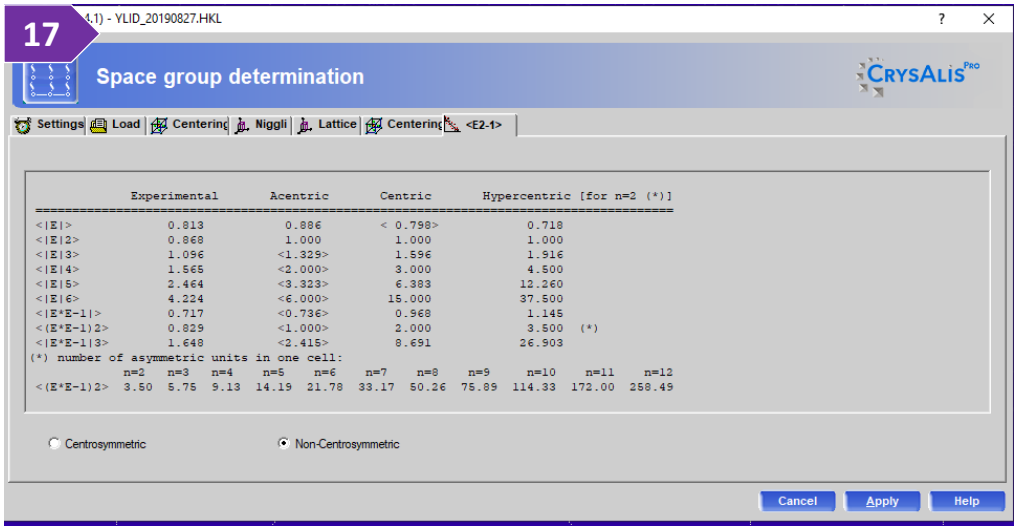
32. Pay close attention to the Rint value, recall that this value should be below 0.10 for good data. A low Rint value suggests that your integration strategy and unit cell assignments are correct. Click “Apply”.



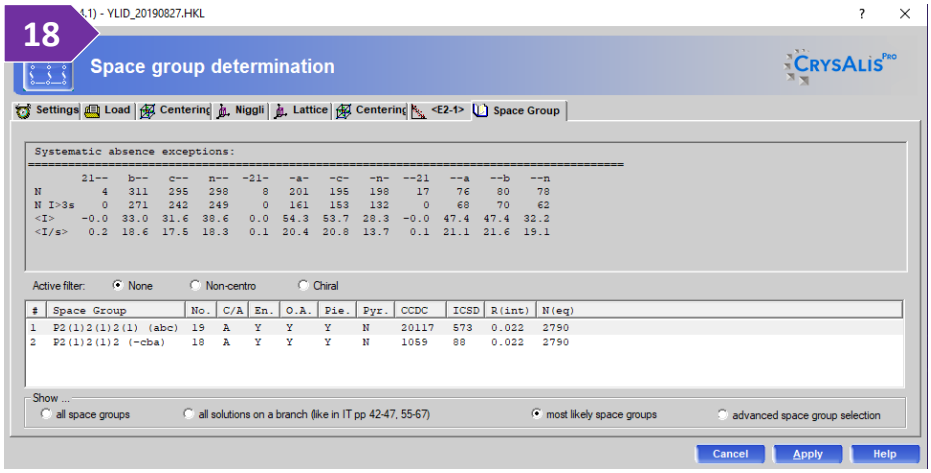
33. Now GRAL will search for higher metric symmetry and additional centering conditions. Click “Apply”.



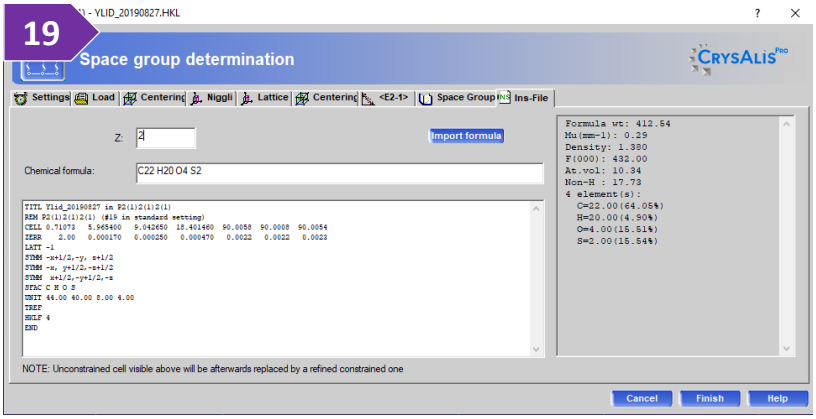
34. The |E2-1| analysis indicates a center symmetric structure. Look at the “Experimental” column, and the |E2-1| value. Click “Apply”



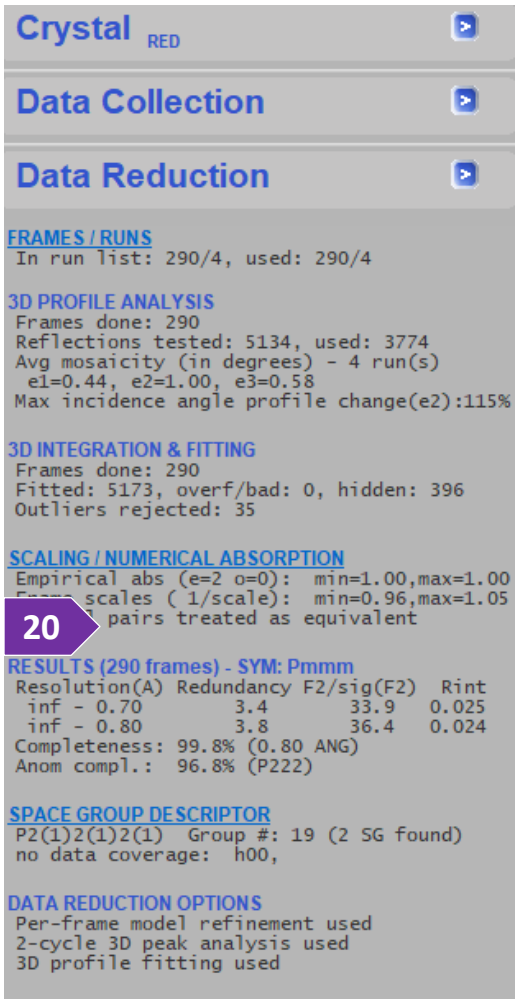
35. Space groups are presented.



36. Now GRAL will produce your HKL and .ins files. Click “Apply”.



37. Notice the side bar of CrysAlisPro: resolution, redundancy, intensity (F2/σ(F2), Rint, and completeness values are listed for the reflection list. As is mosaicity values (e1,e2,e3), the min/max Empirical abs and Frame scales values. Mosaicity gives an idea of peak widths and crystal quality. For a good quality organic sample, values <1 are normal. The frame scaling and the empirical absorption correction correct for inconsistencies in the X-ray beam, absorption, and other anomalies. Ideally, both the min/max empirical absorption correction and the frame scaling should be close to 1. If a crystal is of poor quality, or a crystal absorbs X-rays strongly and a more accurate absorption correction based on crystal shape and size is necessary, then these values will deviate more substantially from 1.



PUBLICATION

A. EXPERIMENTAL SECTION

Project-specific details and setup information are saved in the '*.cif_od' file located under your project folder. ***Modify the text below according to the setup and conditions you used during the measurement:***

"Electron intensity data of an electron transparent single crystal of ***(project name)*** were collected at ***XXX(Y)*** K. Sampel was mounted on a standard TEM grid on an XtaLAB Synergy-ED diffractometer equipped with **a** *200 kV LaB6 source* and a Hybrid Pixel Array Detector (HyPix) detector. Temperature of the crystal was controlled with a Gatan ELSA 698 cryo-holder. Data reduction was performed with the CrysAlisPro software. The structure was solved with the ***(ShelXT | ShelXD | ShelXS | etc.)*** structure solution program using ***(the Intrinsic Phasing | direct methods | Patterson | Dual space | charge flipping)*** solution method and by using ***(Olex2 | Jana2020 | Jana2006 | ShelXle | etc.)*** as the graphical interface. The model was refined with ***(ShelXL | Jana2006 | Jana2020 | etc.)*** using least squares minimization."

B. ACKNOWLEDGEMENT

"Funding from the Air Force Office of Scientific Research (AFOSR) through the Defense University Research Instrumentation Program, Grant FA9550-24-1-0058, which made the purchase and installation of the Rigaku XtaLAB Synergy-ED possible."

TROUBLESHOOTING

A. THE COMPUTER SCREEN WILL NOT TURN ON

Begin your reservation in NUcore to initiate access to the instrument

B. COMPUTER REQUIRES LOGIN AND A PASSWORD

The default 'StadiMP' user account should be logged in. In case the computer was restarted, the password for the 'Synergy-ED' account is . See 'Default instrument status' section for more details

C. THERE IS AN ERROR/PROBLEM WITH THE INSTRUMENT THAT IS NOT ADDRESSED UNDER THE TROUBLESHOOTING SECTION

If there is an error or problem with the instrument which is not addressed under the troubleshooting section, please report the issue by following at least one of the steps below:

1. If you have already started your reservation using NUcore, please end your reservation and select the error reporting option with a brief description about the issue. Place the 'Stop' sign near the instrument computer to notify users immediately after you. 'Stop' signs are located on the shelf above the computers in BG51
2. If you have not started your reservation using NUcore, please report problems with the instrument at http://imserc.northwestern.edu/contact-issue.html and place the 'Stop' sign near the instrument computer
3. Contact a staff member for instructions

REVISIONS

v1.00 2024/08/16	• Release of original version of the user manual for Synergy-ED using CrysAlisPro 44.65a