CrysAlis^Pro User Guide

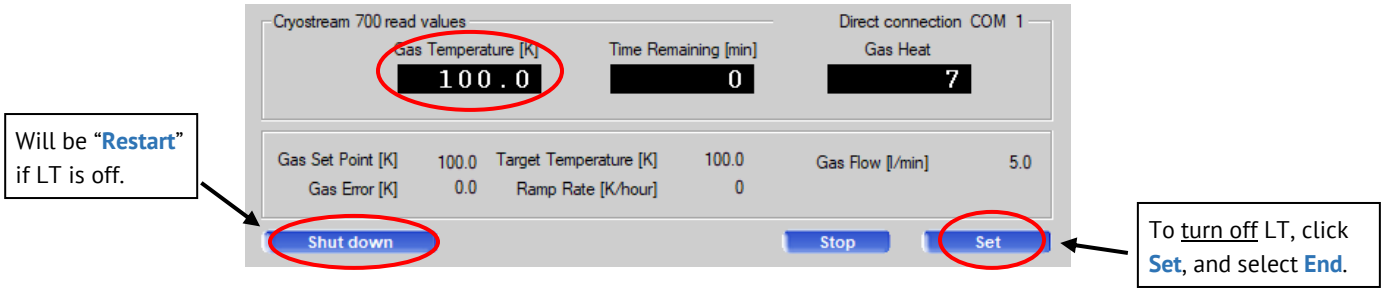
❖ Turning on LT Device and X-rays

1. Launch CrysAlisPro (online) with the desktop icon. 

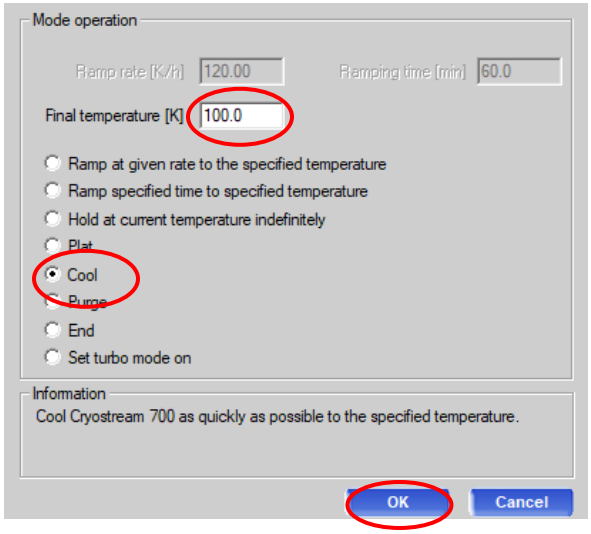
2. Wait for the instrument to initialize.

3. If the Oxford Cryosystem/LT unit is off, left click on **CRYO**.

   i. Click **Restart** if the LT is not turned on and wait for the Gas Temperature to show numbers instead of "???".
   ii. Click **Set**.

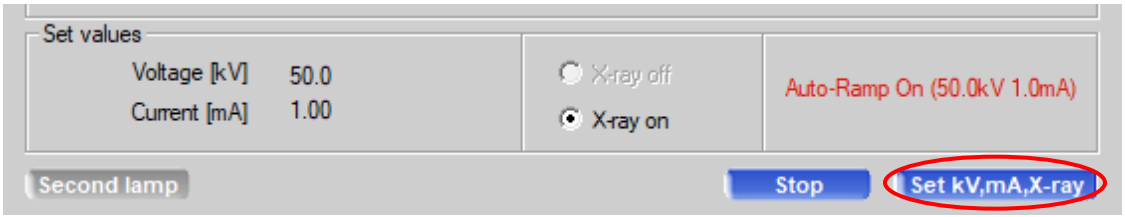


   iii. Change the mode to **Cool** and Final temperature to **100K**, click **OK** and close the Oxford Cryosystems window.

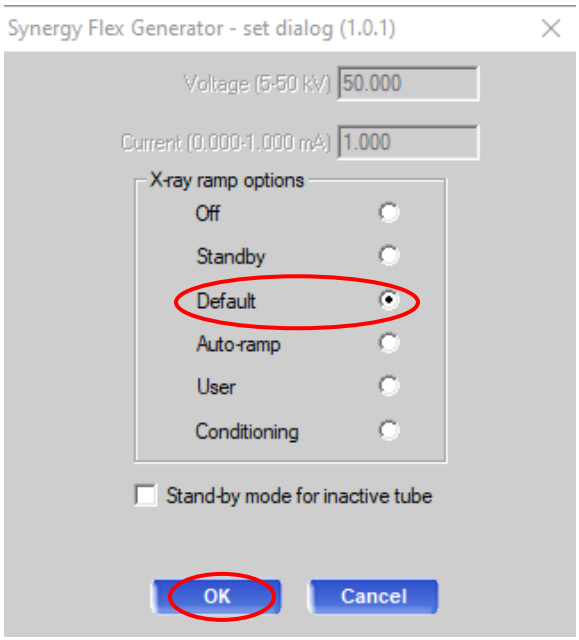


4. If the X-ray generator is off, left click on **X-RAY**.

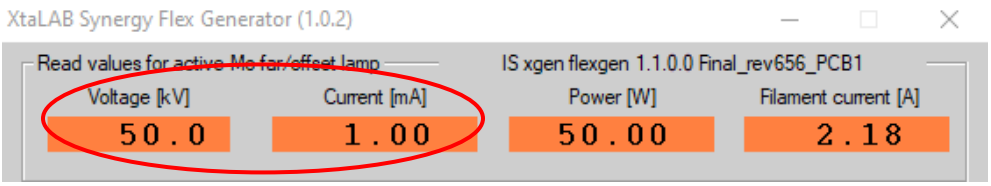
i. Click **Set kV, mA, X-ray**.



ii. Select **Default** and click **OK**.



iii. Close the window when Voltage is **50.0** and Current is **1.00**.



Note: The generator window cannot be closed while the X-rays are being ramped up.

5. X-ray generator will turn off automatically after 6 hours. If you'd like to turn them off manually, click **Set kV, mA, X-ray**, select Off and click **OK**.

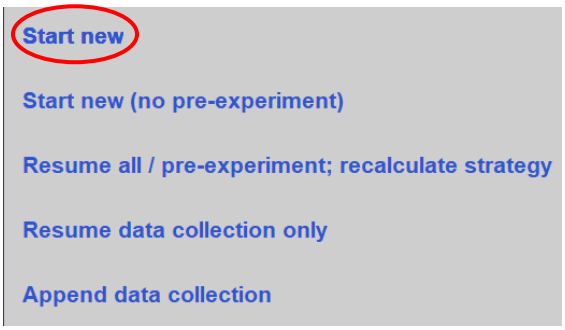
❖ GETTING STARTED (SETTING UP AN EXPERIMENT)

1. Click on **START/STOP**.

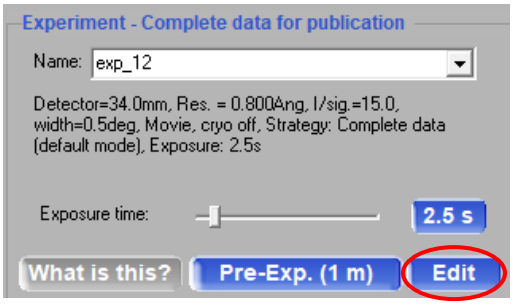
If you would like to switch the X-ray source, click on this button and select "Yes."

This button should read **X-ray Mo** or **X-ray Cu**.

2. Click on **Start new**.



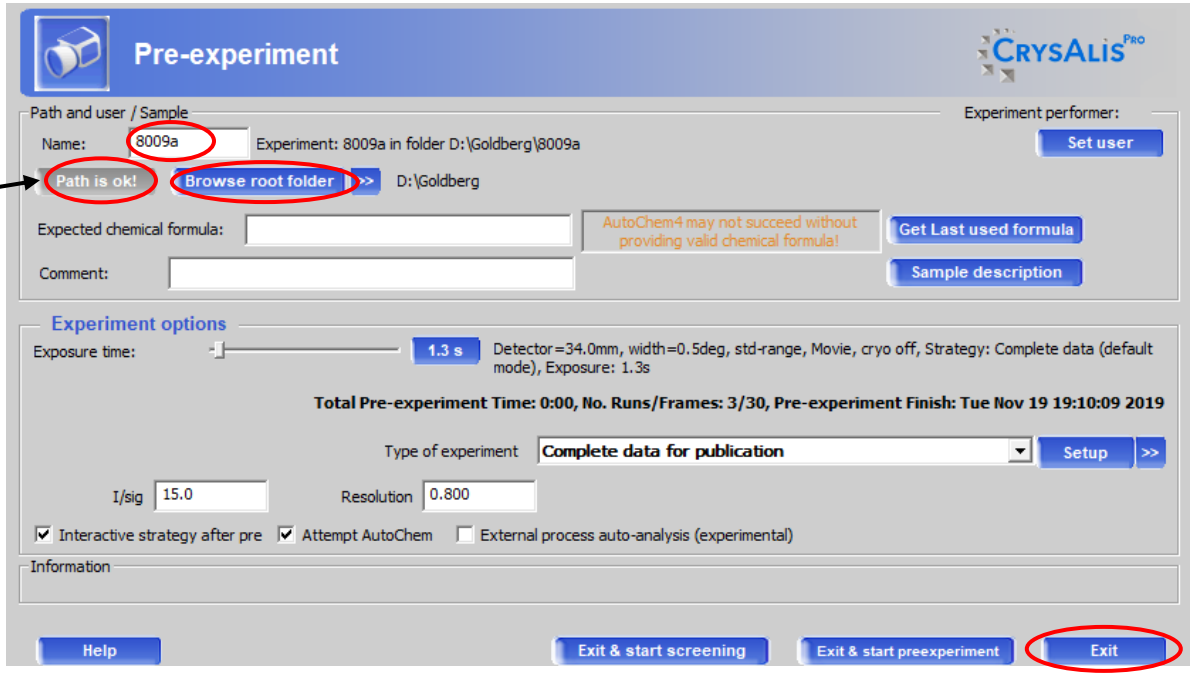
3. Click on **Edit** to setup experiment info.



4. Click on **Browse root folder** and locate your lab's folder in the D:\DATA drive and click **OK** (click **No** when asked to change root folder).

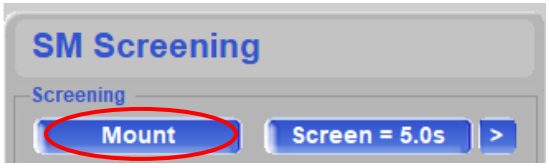
5. Change the name of the experiment (check Penn database or sign-up board) and click **Exit**.
   e.g. 6874a (# differs between labs, letter is used if multiple crystals of the same sample are being tested)

Will be "**Clear Folder**" if the name is taken. **Please do not overwrite anything**. Use the next letter.



❖ CENTERING THE CRYSTAL

1. Click **Mount** or press F12 on the keyboard, this will open the centering interface.

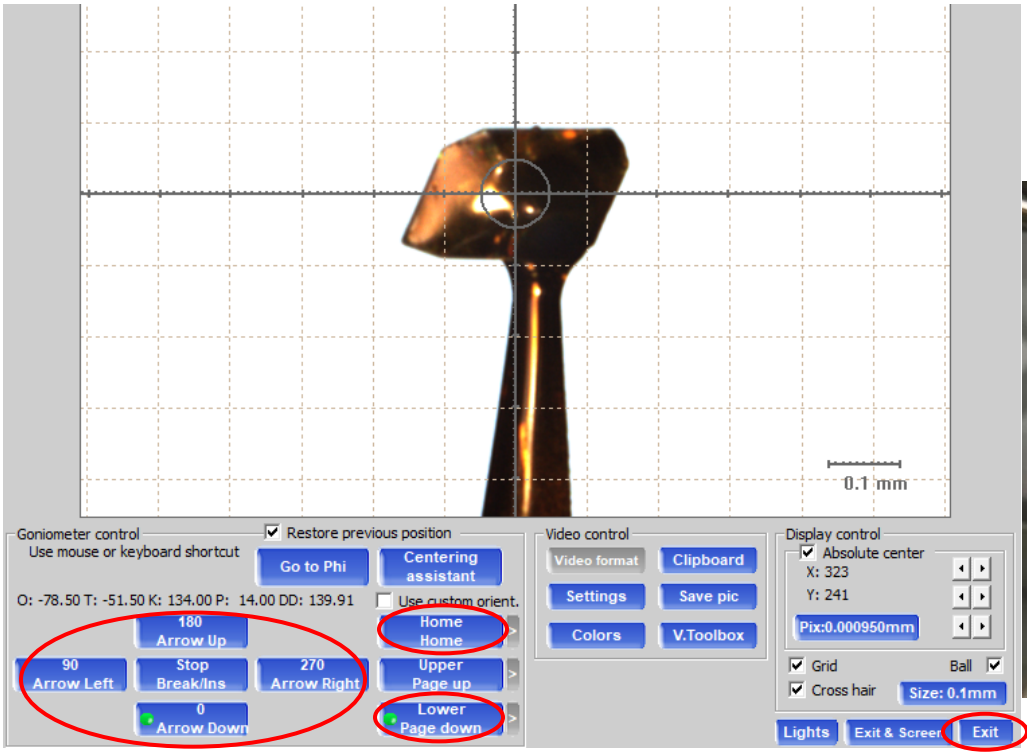


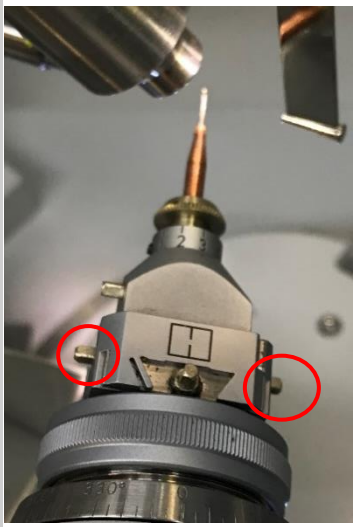
Note: If the door is open, press and hold **both** "**Motion Enable**" buttons in the enclosure or the goniometer will not move. Alternatively, you can close the door and press the "**Doors OK**" button on the bottom right panel.

2. Click **Home** (easiest position to put on the goniometer head) and screw on the goniometer head, do not overtighten. **Be careful** not to bump the beamstop, collimator, coldstream nozzle or detector face.

3. Click **Lower** to begin centering (**hold down** Motion Enable buttons to allow movement or close doors and press "Doors OK").

4. Cycle through **0**, **90**, **180** and **270** phi orientations while centering the crystal at each position. Use the knobs on the **left** or **right** side of the gonio head to adjust the position shown on the camera.



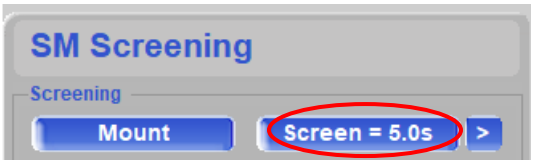


5. Once centered, click **Home**, **gently close the door** and press the **Doors OK** button. Then click **Exit.**
    a. **Icing** tends to occur in the Lower position, so spend as little time possible in that orientation.

Note: If you have trouble centering, the camera might be out of alignment, contact your crystallographer to check and adjust.

❖ Screening, Pre-Experiment and Strategy

1. Click the **Screen** button using default exposure time.

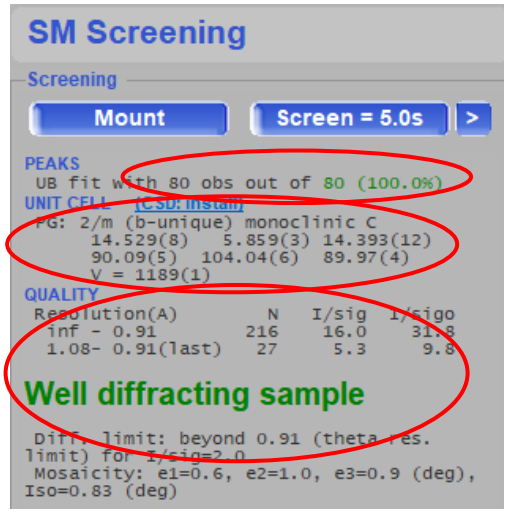


Note: If you have tried several crystals and they are all weak, you can increase the exposure time by clicking > and adjusting the value in the **Exposure time** box. It is beneficial to find a good sample BEFORE the pre-experiment.

2. Examine the images to ensure you have well-defined spots with no rings/streaks or overlaps. If you have trouble seeing spots, make sure the contrast is at **Level 1 (255)** and you can change the image color scheme as well.

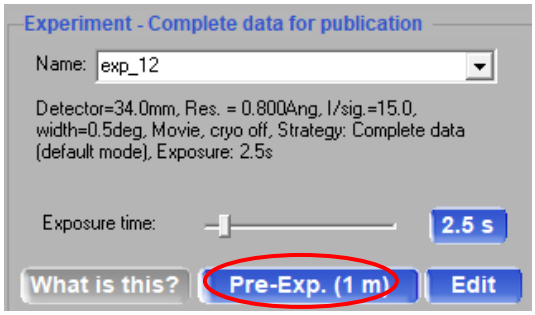
3. Once screening is complete, you should **check the results** before continuing to the Pre-Experiment.

   i. **High % fit** of predicted cell to observed spots
   ii. Reasonable unit cell (volume and lattice)
   iii. **Well diffracting sample** and good quality reflections



   a) If you have a 50-70% fit, then you might have a twinned crystal, try another and if it persists, you may continue.
   b) Check the density of your crystal sample (d = MW/V x 1.66 x Z), a reasonable density is 1.2 to 2.0 g/cm^3.
   c) If you have a **moderately diffracting** or **weakly diffracting** sample, try another crystal, if it persists but the reflections look well-defined, you may continue.

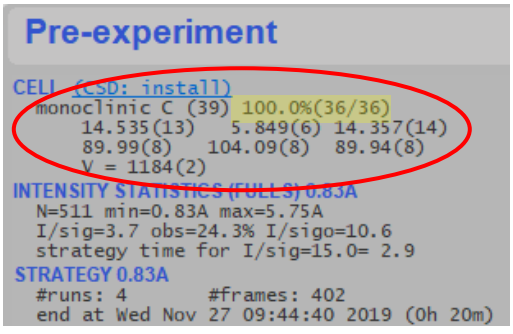
4. Click **Pre-Exp** to begin the pre-experiment with default exposure time (estimated from screening process).



Note: The pre-experiment is a more intensive screening where more frames are collected to better determine exposure times, unit cell and the orientation matrix.

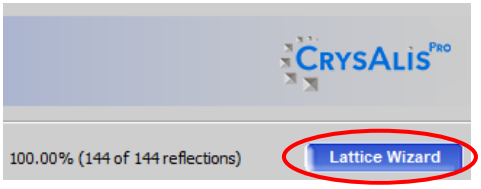
5. Once the pre-experiment is finished, the strategy module will open automatically.

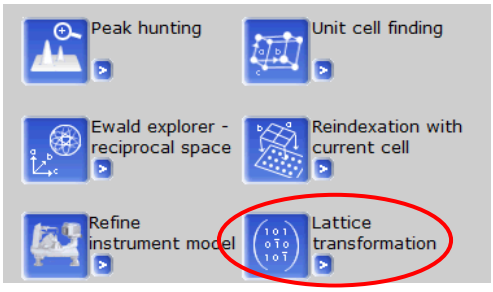
Note: Double check the pre-experiment results are consistent with the screening results. (% of fit reflection and unit cell)



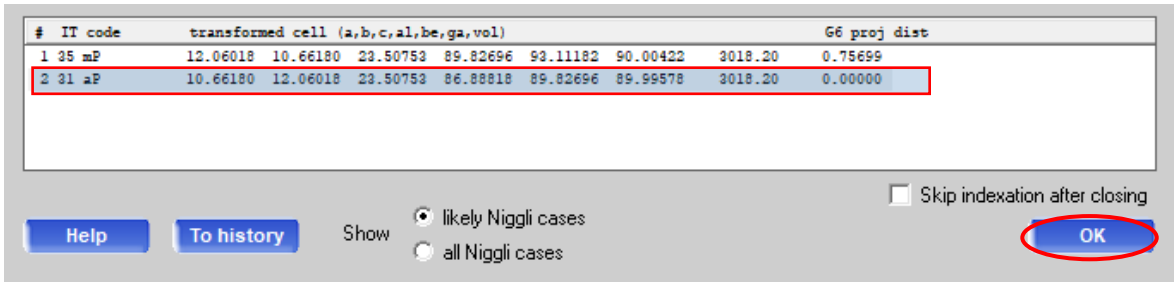
Note: If you closed the Experiment Strategy window, the only way to re-open it is to click **START/STOP**, then **Resume all/pre-experiment; recalculate strategy**, and then select the "**pre_xxxx.run**" file. If you want to **Screen** a different crystal, you must start over. Click **START/STOP**, then **Start New** and **change the name of the experiment** before continuing.

6. Click **Lattice Wizard** in the top right of the Experiment Strategy window.

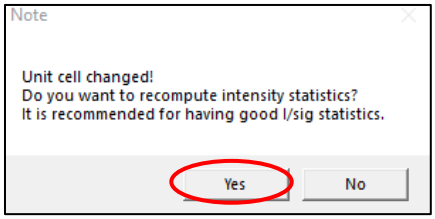




7. Click **Lattice transformation**.
8. Select **aP** for Triclinic P (should always be on the bottom) and click **OK**.



9. Click **Close** to exit the Lattice wizard. A window will pop up informing you of the unit cell change, click **Yes**. The strategy module will re-open.

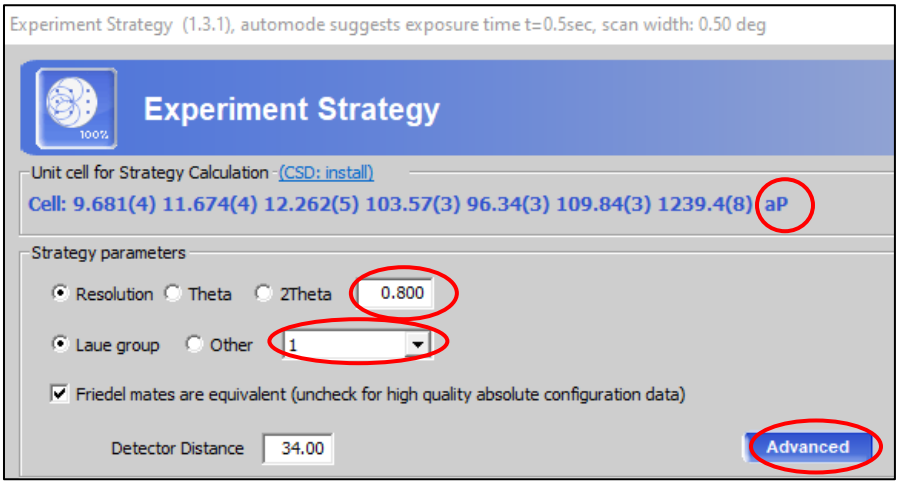


10. **aP** should be the new Cell and the Laue group should be **1**.

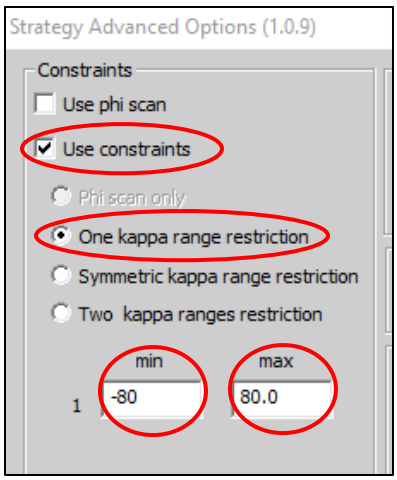
11. Under **Strategy parameters**, change the **Resolution** to **0.75 for Mo** and **0.80 for Cu**.

Note: Microfocus X-ray sources are very intense and should be able to reach good resolutions for both heavy and light atom compounds.

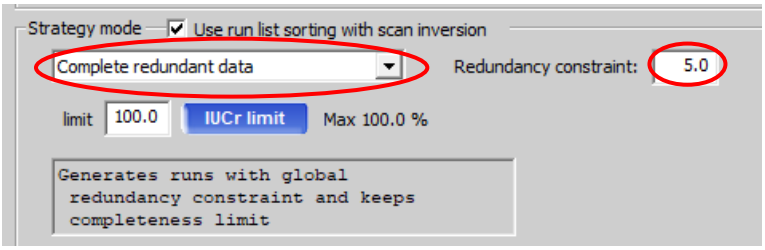
12. Click **Advanced** to open the Strategy Advanced Options window.



13. Select **Use constraints**, and **One kappa range restriction** and change the min to **-80** and max to **80**, click **OK**.
(kappa restriction prevents icing for long collections)



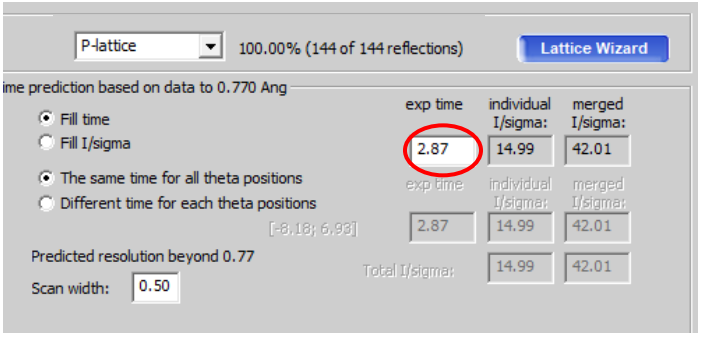
14. Under Strategy mode, use the dropdown menu to select **Complete redundant data** and change the value to **4** if the crystal may be higher symmetry than triclinic and **6** if the crystal can only be triclinic.



15. The checkbox for Friedel mates and Detector Distance value can be left as is. (the only reason to increase detector distance is if you have a twinned crystal with a lot of overlapping spots).

16. **Exp time** can be left as is or adjusted (we like to change it to a whole number, a half or an interval of 5).

Note: For collections done with **Cu**, you will have TWO different exposure times (low angle and high angle), this shortens collection time due to low angles having shorter exp time.

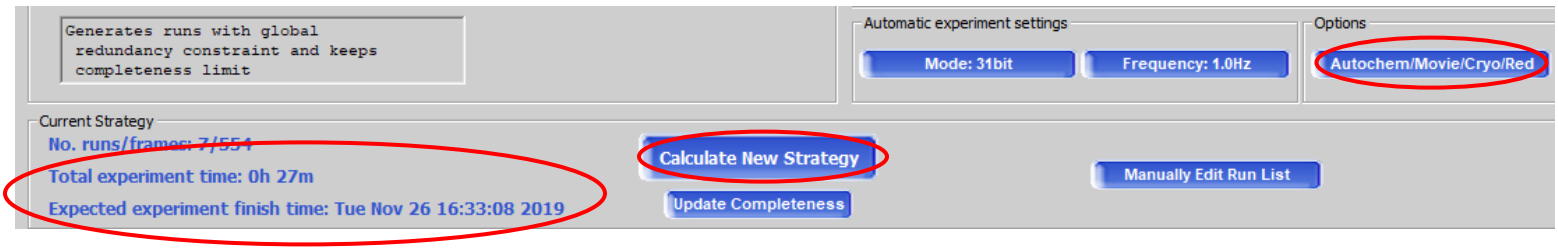


Note: If you have a **very weak crystal** (exp time longer that **60 seconds**), you can:
1) Change the I/sigma to 10
2) Reduce the resolution to 0.83Å.

17. Click **Calculate New Strategy** (might have to change exp time again).

18. Make note of the **total experiment time** and the **experiment finish time**.

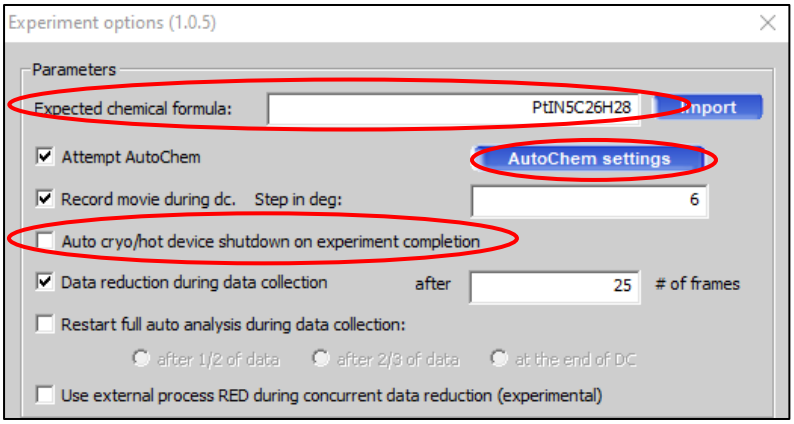
19. Click **Autochem/Movie/Cryo/Red**.



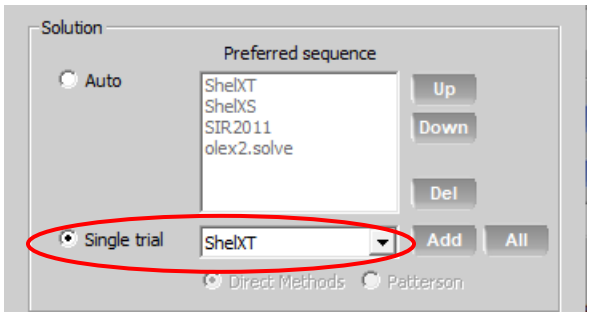
20. Enter the **Expected chemical formula**.

21. If the experiment will finish in the middle of the night or another user will not use the diffractometer after you, **check the Auto cryo device shutdown**.

22. Click **AutoChem settings**.



23. Select **Single trial** and use the dropdown menu to select **ShelXT**.



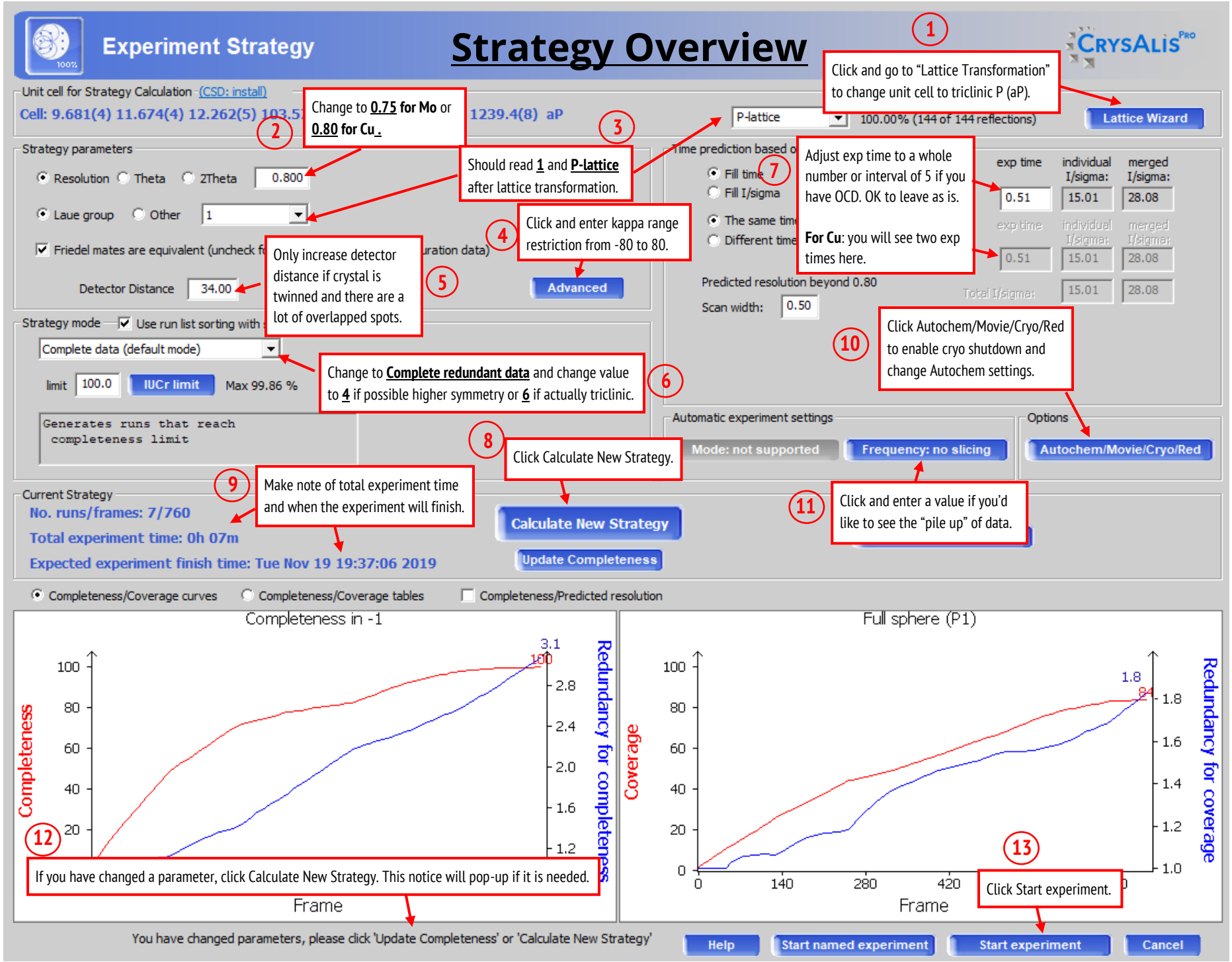
24. Click **OK**, then **OK** again to exit Experiment options.

25. Click **Start Experiment**.

26. CrysAlisPro will begin automatically processing the data every 25 frames and attempt a solution when the data completeness reaches ~40%.

Note: When the collection is finished allow it to run a final integration, scaling and AutoChem. You can find the AutoChem files (.res, .hkl and .cif_od) in the **struct** folder of your corresponding experiment.

- If the automatic data processing and solution are satisfactory, you can use the files in the **struct** to finish the structure. If you would like to manually process the data (we prefer to cutoff the resolution at 0.75Å or 0.83Å) then continue to the next section.


Experiment Strategy
Strategy Overview
CrysAlis PRO
1
Click and go to "Lattice Transformation" to change unit cell to triclinic P (aP).
Lattice Wizard
Unit cell for Strategy Calculation (CSD: install)
Cell: 9.681(4) 11.674(4) 12.262(5) 103.5
1239.4(8) aP
2
Change to 0.75 for Mo or 0.80 for Cu .
3
Should read 1 and P-lattice after lattice transformation.
P-lattice
100.00% (144 of 144 reflections)
Strategy parameters
Resolution
Theta
2Theta
0.800
Laue group
Other
1
Friedel mates are equivalent (uncheck f
uration data)
4
Click and enter kappa range restriction from -80 to 80.
Advanced
Detector Distance
34.00
5
Only increase detector distance if crystal is twinned and there are a lot of overlapped spots.
Strategy mode
Use run list sorting with
Complete data (default mode)
limit
100.0
IUCr limit
Max 99.86 %
6
Change to Complete redundant data and change value to 4 if possible higher symmetry or 6 if actually triclinic.
Generates runs that reach completeness limit
Time prediction based o
Fill time
Fill I/sigma
The same time
Different time
7
Adjust exp time to a whole number or interval of 5 if you have OCD. OK to leave as is.
For Cu: you will see two exp times here.
exp time
individual I/sigma:
merged I/sigma:
0.51
15.01
28.08
exp time
individual I/sigma:
merged I/sigma:
0.51
15.01
28.08
Total I/sigma:
15.01
28.08
Predicted resolution beyond 0.80
Scan width:
0.50
10
Click Autochem/Movie/Cryo/Red to enable cryo shutdown and change Autochem settings.
Automatic experiment settings
Mode: not supported
Frequency: no slicing
Options
Autochem/Movie/Cryo/Red
8
Click Calculate New Strategy.
9
Make note of total experiment time and when the experiment will finish.
11
Click and enter a value if you'd like to see the "pile up" of data.
Current Strategy
No. runs/frames: 7/760
Total experiment time: 0h 07m
Expected experiment finish time: Tue Nov 19 19:37:06 2019
Calculate New Strategy
Update Completeness
Completeness/Coverage curves
Completeness/Coverage tables
Completeness/Predicted resolution
Completeness in -1
Completeness
Redundancy for completeness
Frame
3.1
2.8
2.4
2.0
1.6
1.2
100
80
60
40
20
12
If you have changed a parameter, click Calculate New Strategy. This notice will pop-up if it is needed.
Full sphere (P1)
Coverage
Redundancy for coverage
Frame
1.8
1.8
1.6
1.4
1.2
1.0
0
140
280
420
13
Click Start experiment.
You have changed parameters, please click 'Update Completeness' or 'Calculate New Strategy'
Help
Start named experiment
Start experiment
Cancel

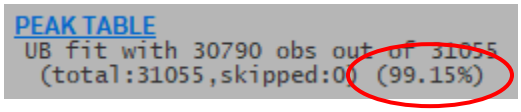
❖ MANUALLY PROCESSING THE DATA

1. Launch CrysAlisPro (red) with the desktop icon.

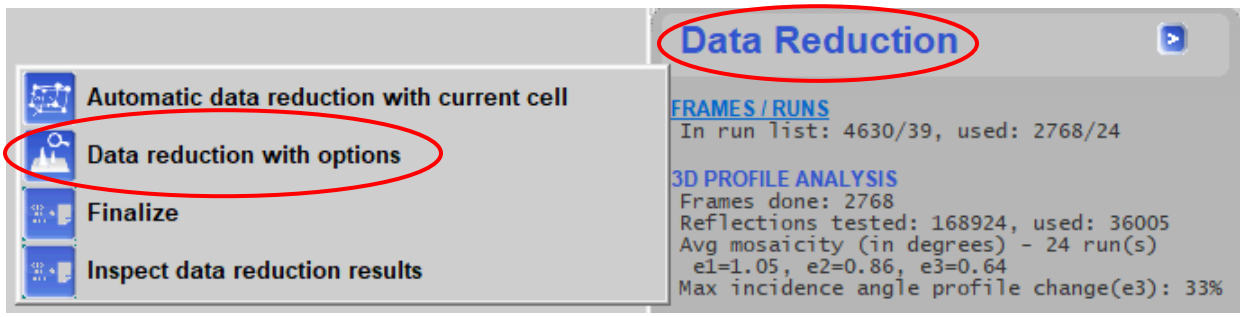
2. Locate the experiment you are trying to open. Select it and click **Open selected**. (If it's not automatically listed, click **Browse experiment**, locate the folder, click on the **xxxx.par** file, click **Open**, and then **Open selected**.)

3. Left-click or move cursor on **Crystal** RED, then click **Full auto unit cell finding**.

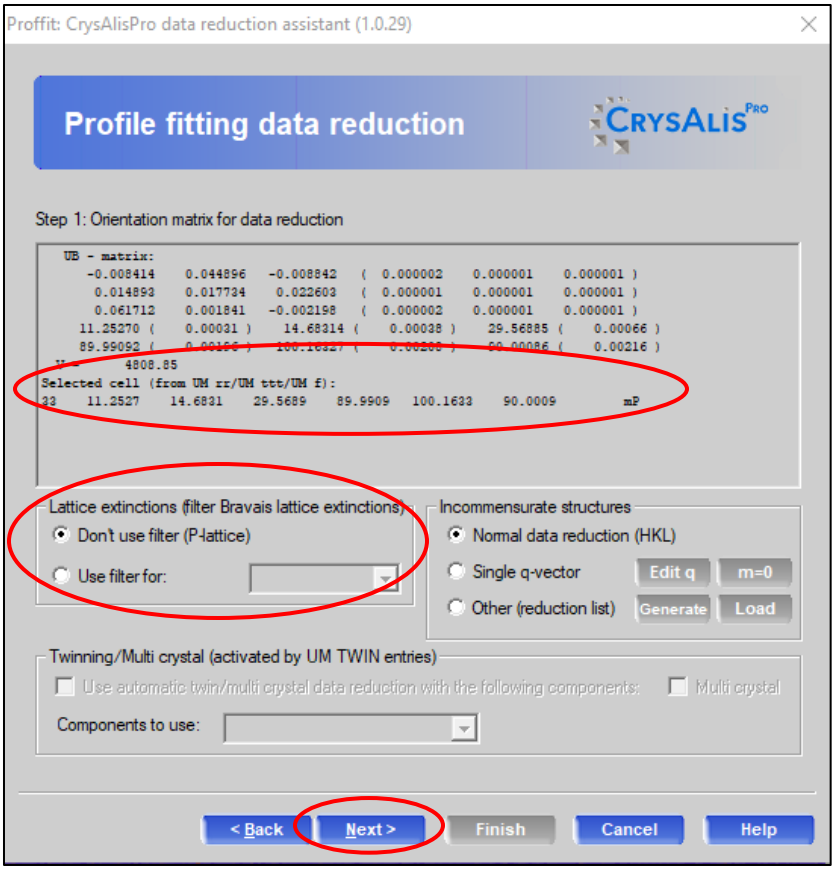
4. Check that the **PEAK TABLE** has a good fit (80-100%). If not, you might have a twin and will need to process the data as a twin. This is done using the **Ewald explorer – reciprocal space** interface in the **Lattice wizard**.



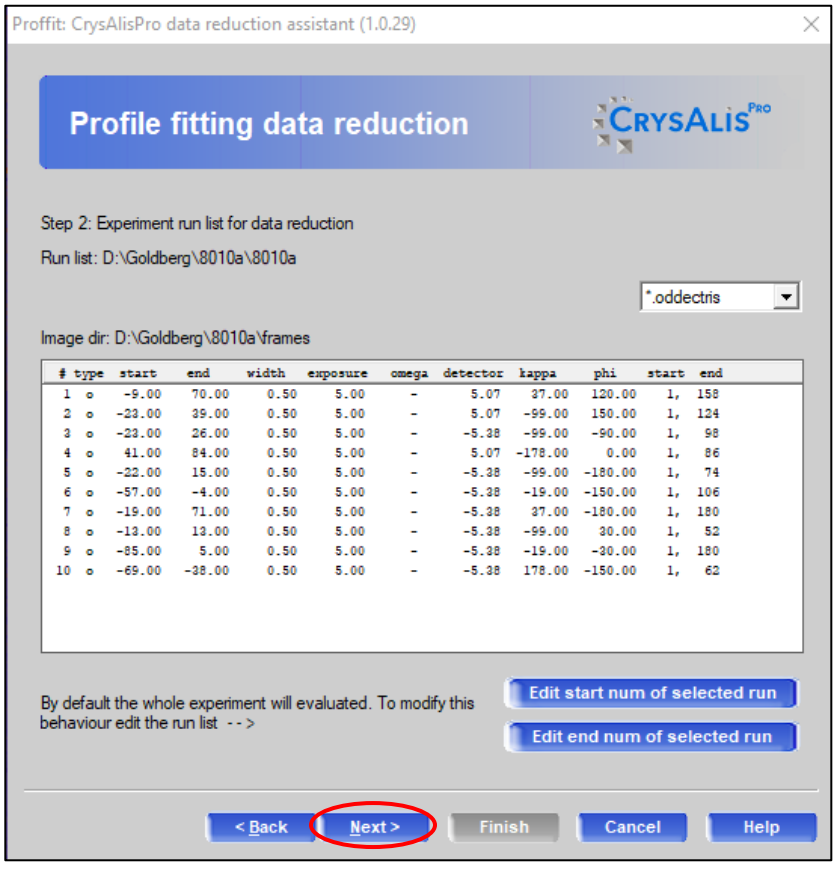
5. Click **Data Reduction**, then move cursor over **Data Reduction.** Then select **Data reduction with options**.



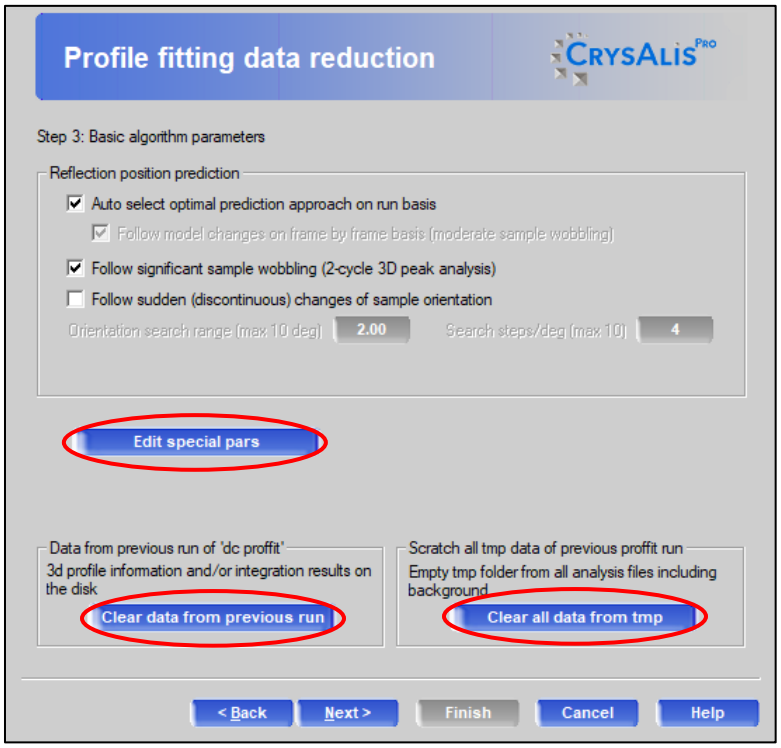
6. The Proffit interface will open. Check that the selected cell and lattice extinctions match. Click **Next >**.



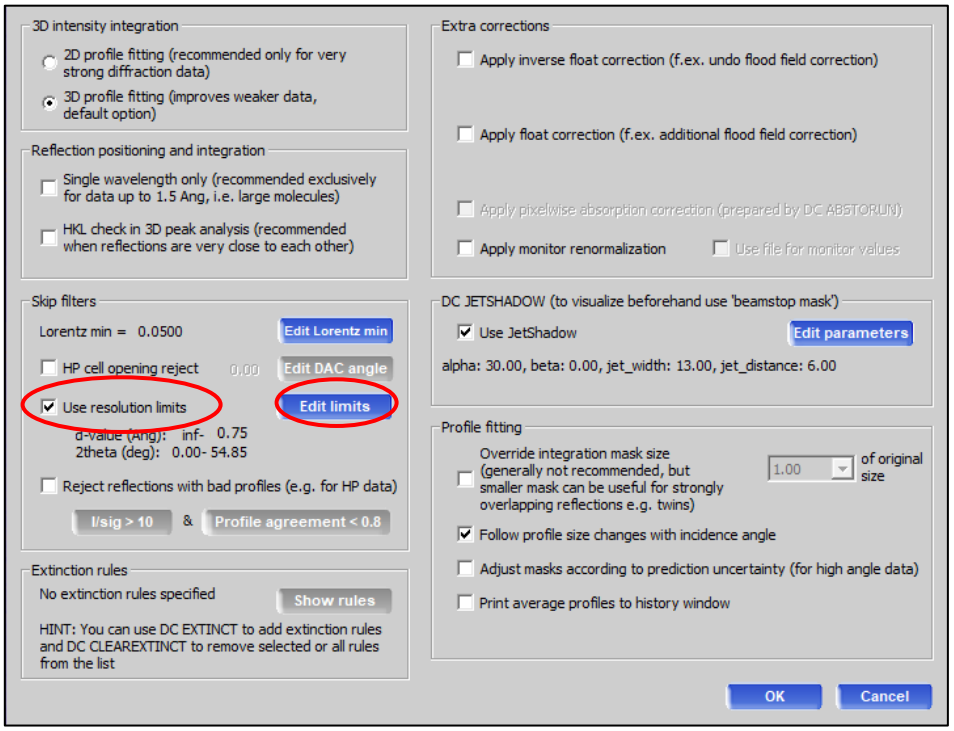
7. Step 2 is useful if you need to change the start or end frame (due to icing or other issues). Click **Next >**.



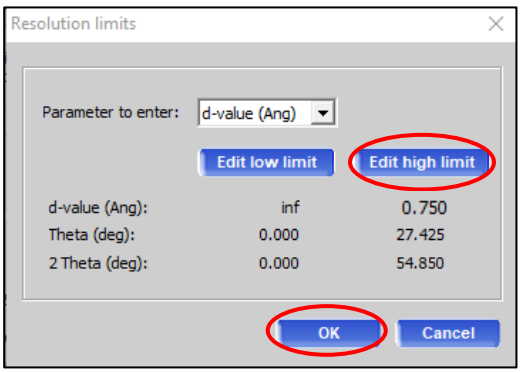
8. Click **Clear data from previous run** and **Clear all data from tmp** to delete all info from previous processing runs. Then click **Edit special pars**.



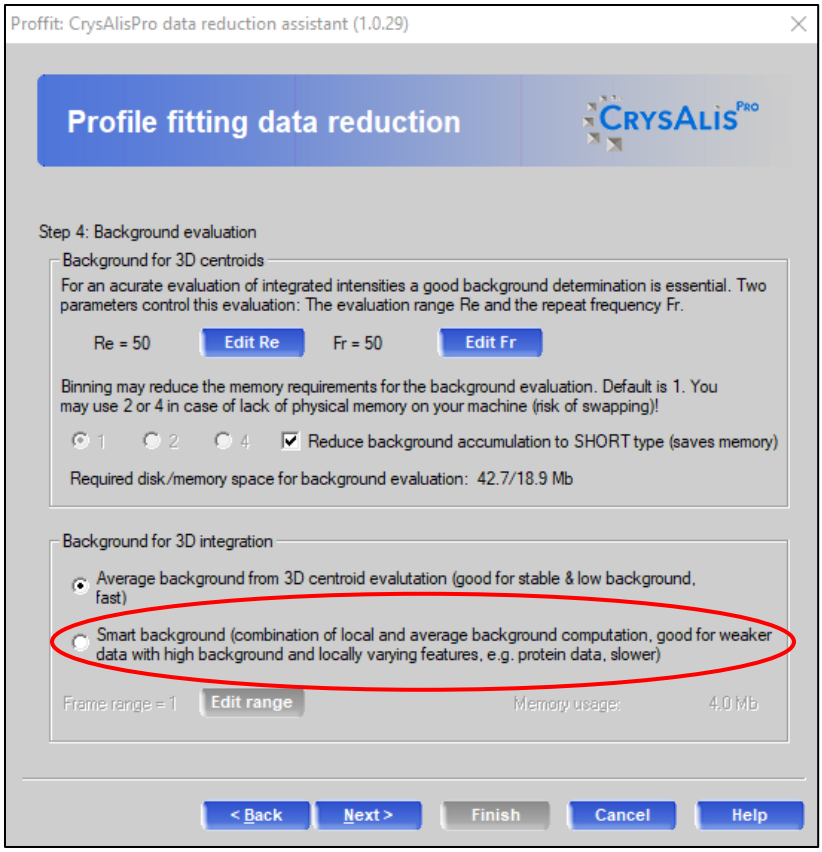
9. In the special parameters window, check **Use resolution limits** and click **Edit limits.**



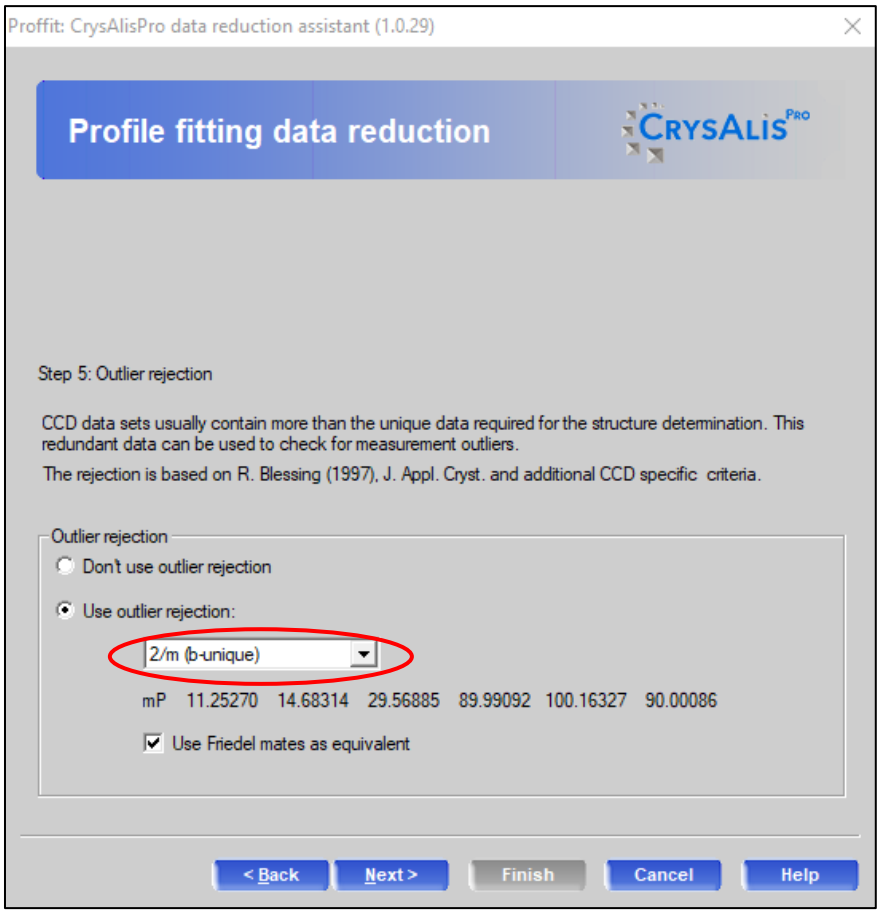
10. Click **Edit high limit** and change to **0.75**Å for heavy atom compounds and **0.83**Å for light atom compounds (no element heavier than Cl). Then click **OK** and **OK** in the special parameters window.



11. Select **Smart background** and click **Next >**. This is used if the X-ray background is high and/or irregular throughout the dataset. Automatic integration uses **Average background**.



12. Check the outlier rejection and make sure it matches previous lattice selections. Click **Next >**.



13. Click **Change output name** and enter a new output name (e.g. 8010a_v1), Click **OK**. This is important so no files will be overwritten.

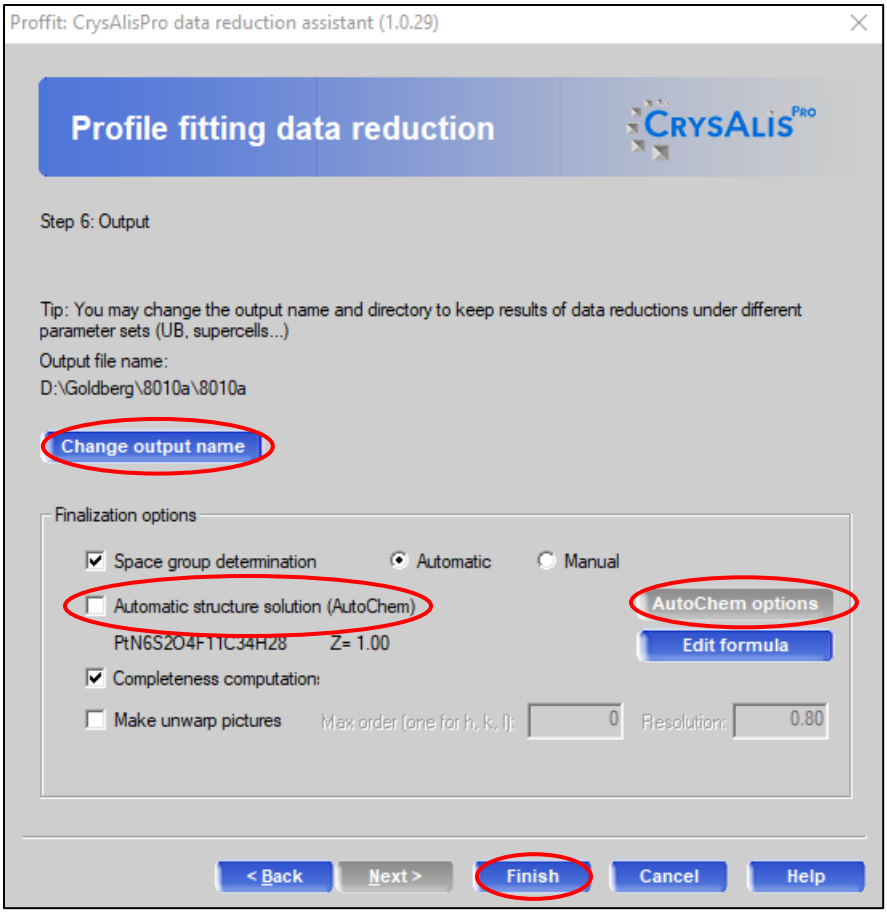
14. Check the **Automatic structure solution** box. This will generate a new **struct** folder with your new output .res and .hkl.

    Note: If you forget to select this, only an .ins and .hkl will be generated in the primary experiment folder. Open the .ins in Olex2 and SOLVE with ShelXT.

15. Click **AutoChem options** and select **Single Trial** and **ShelXT,** then click **OK**.

16. Click **Finish**. CrysAlis^{Pro} will integrate, determine the space group and scale.

17. If the resulting .res and .hkl files in the corresponding **struct** folder are satisfactory, you may finish refining the structure.



18. To further inspect the data collection and reduction results, click  in the left toolbar.

❖ USEFUL NOTES AND COMMANDS

- To go to Home position, click 

in the left toolbar. Make sure CCD tab is selected and type "gt a 0 0 0 0 180" (goto all omega# theta# kappa# phi# detector distance#)

- If you forget to select Auto cryo device shut down during the Strategy setup, you can do so after the experiment has started. Click **Data Collection** and click on "**Off: Autocryo Shutdown**". It should now read "On: Autocryo Shutdown."

- If you suspect a twin, use the **Ewald Explorer** in the **Lattice Wizard** to identify the twin components by clicking on **Activate twin/multicrystal** and selecting **Automatic Twin Finding**. Typical rotation should be 180 degrees for an actual twin and a reasonable percentage of leftover spots should be selected (if primary component indexes as only 60%, the twin should hopefully fit other 40%). Once this is done, you must manually process data, and after structure solution with ShelXT, refine using the HKLF5 file that has been generated.