GΦL status

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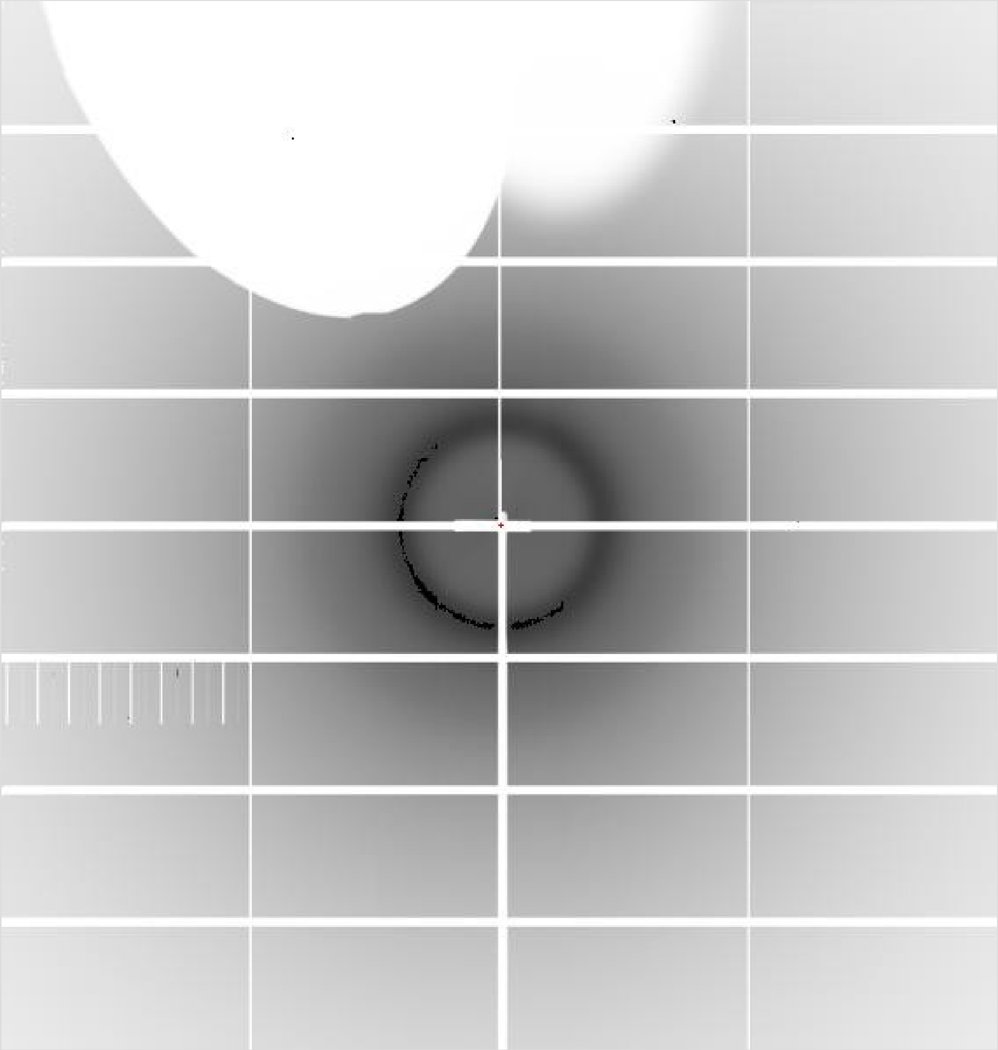
DESY: P14 Visit 6-7 May 2019 (1)

- Lessons:
  - We must always have access to the MxCUBE code running on the beamline before the visit
  - Detector positioning is handled in an unusual way at P14
    - Quick and dirty fix during the visit
    - The Abstract Beamline Interface contract was broken
  - Workflow-generated collection sequences were new to the beamline: images were lost
  - Initial configuration is still hard
    - the camera transforms the coordinate system
    - a software solution is possible
  - Pin shadowing is a problem

Unmodelled shadow



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DESY: P14 Visit 6-7 May 2019 (2)

- Achievements:
  - Translation calibration
  - Diffractometer calibration (using cubic insulin and thaumatin)
  - Shadowing images
- This is the furthest that we have got on the initial beamline visit so far.

ALBA: XALOC visit (28-29 May 2019)

- Diffractometer calibration
  - Our first use of germanate crystals within the MxCUBE collaboration (rather than DLS)
- First scientific use of the GΦL Workflows
  - Native strategies on ligand-soaked crystals

Diffractometer calibration (1)

- Wrong wavelength used
  - Too much absorption, led to crystal decay
    - Inexperience with handling germanate – next time it will be done better
- Crystal quality variable?
- Centring at many (κ,φ) values
  - We still have not cracked the retention of centring on a mini-Kappa at multiple orientations
    - Unsure of the relative contributions of pin shrinkage and mini-Kappa mechanics

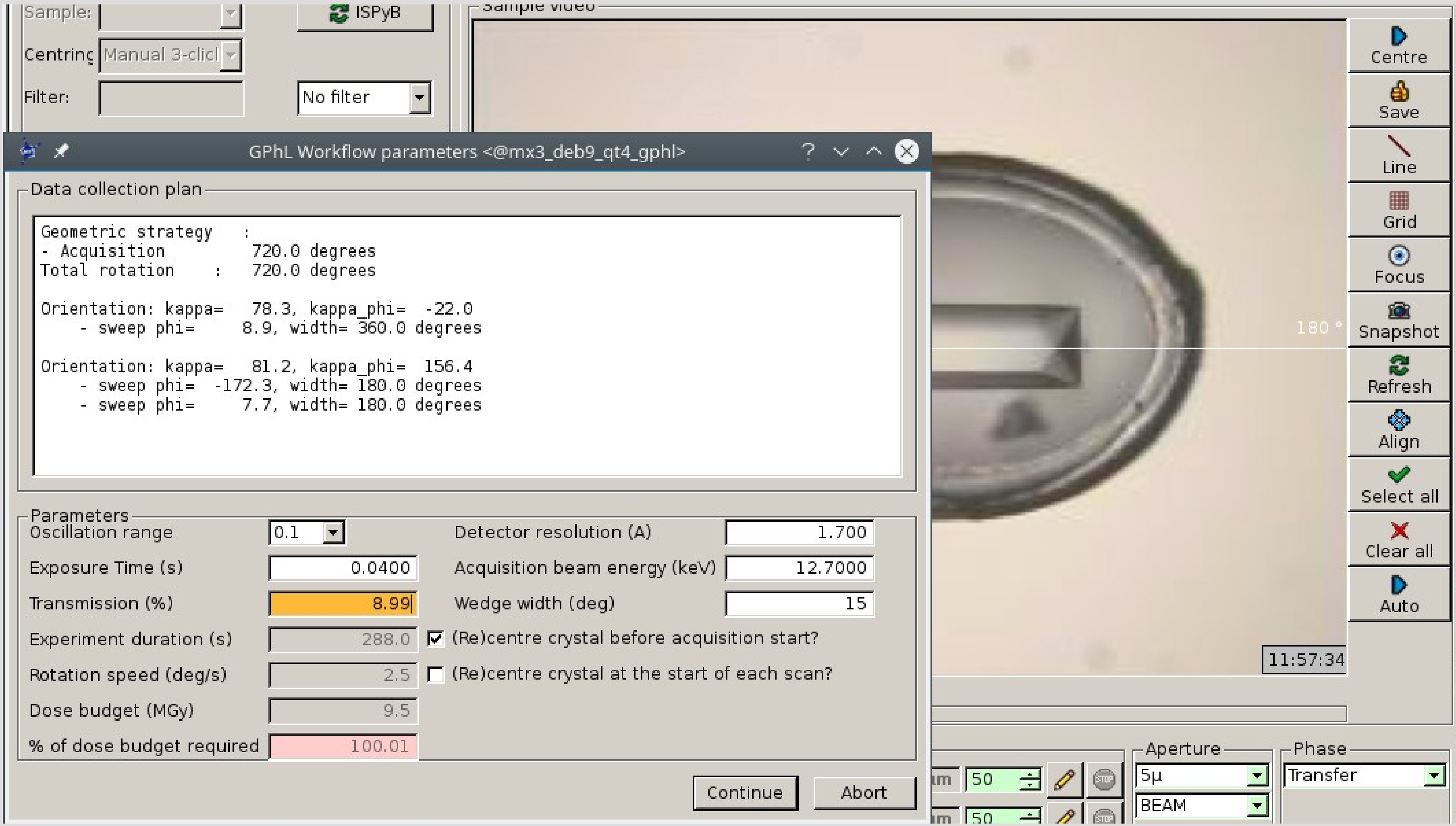
Diffractometer calibration (2)

- Shadowing still a problem
  - Pin shadowing more unpredictable than goniostat shadowing
    - Careful sample preparation needed
  - Beamstop shadowing also caused problems
    - Edge effects gave spurious spots
    - Had to be corrected by hand in processing
- Nevertheless, the processing succeeded
  - Some parameters needed to be adjusted to their most permissive values
- Improvements possible for next time

Native data collection strategies

- Strategies specifically designed for known symmetry and orientation of each sample
  - Dose calculation added to MxCUBE by Rasmus, with input from Leigh Carter (GΦL) and Gleb (EMBL-HH)
- Samples provided by José Márquez (EMBL-Grenoble)
  - 5 datasets collected: 2 apo, 3 on ligand-soaked crystals
  - One ligand-soaked crystal had approximately-doubled unit cell dimensions:
    - $a_2 = 2.0\ a_1$; $c_2 = 2.1\ c_1$

Dose budget in GΦL Workflow UI



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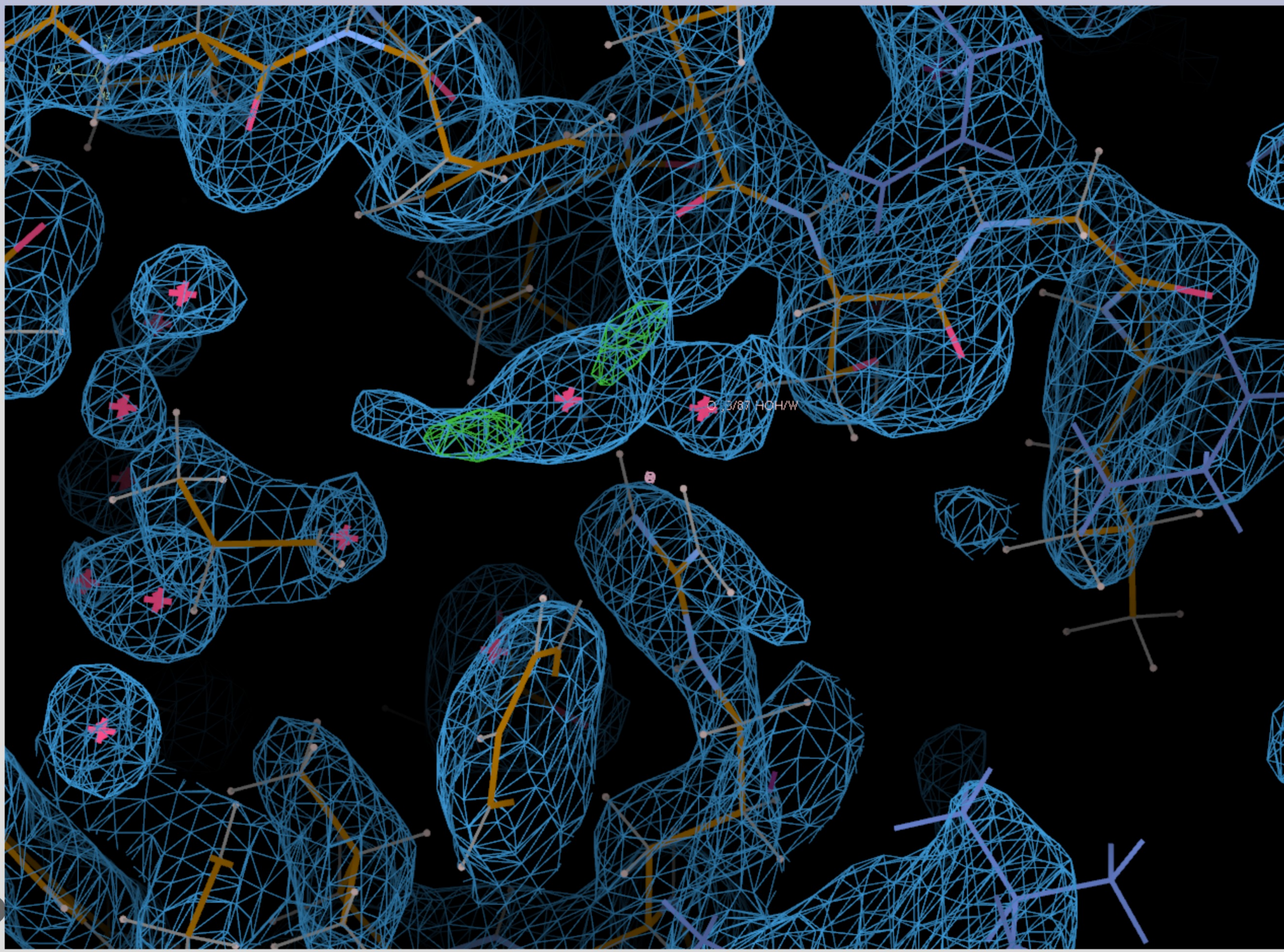
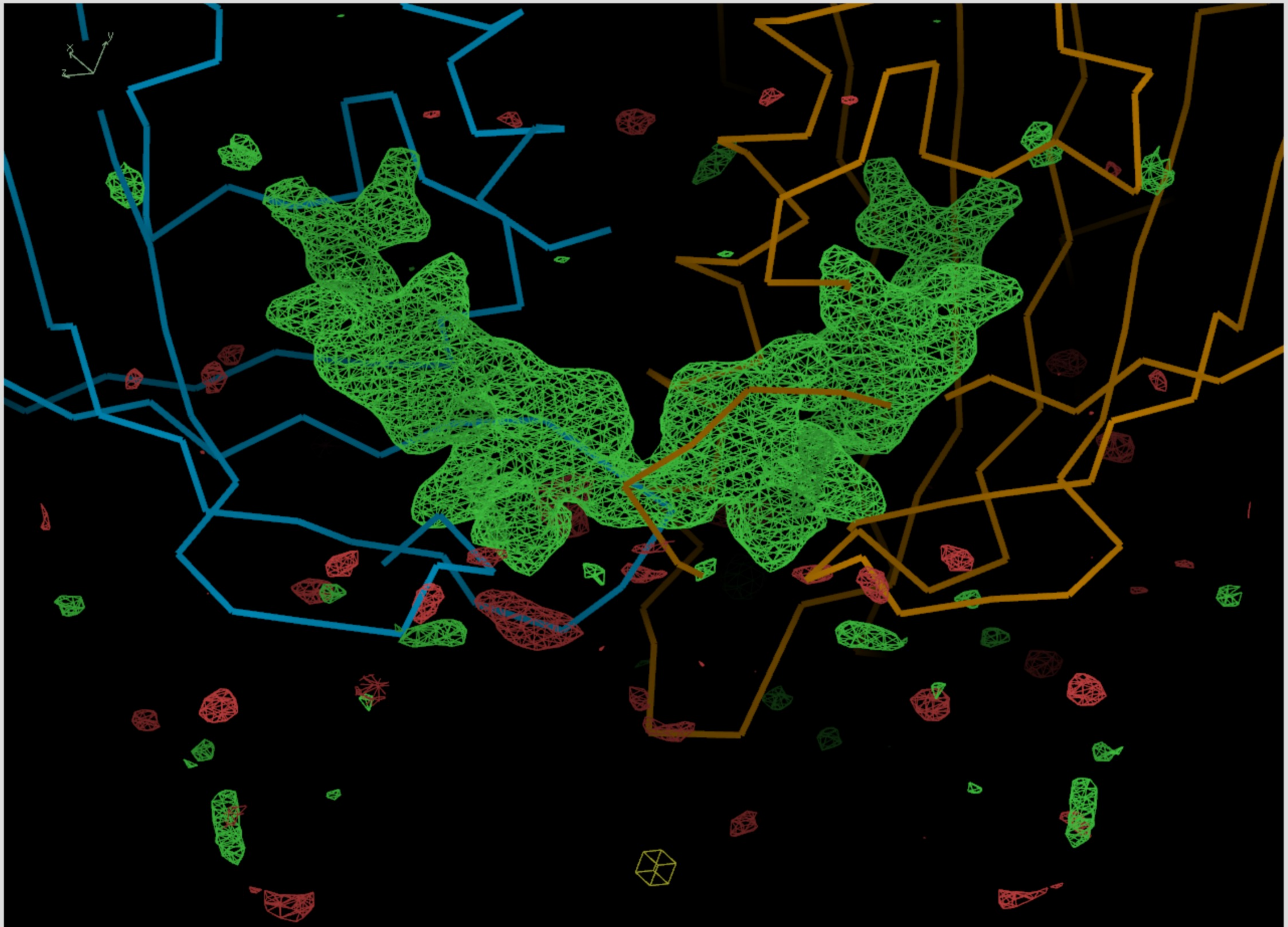
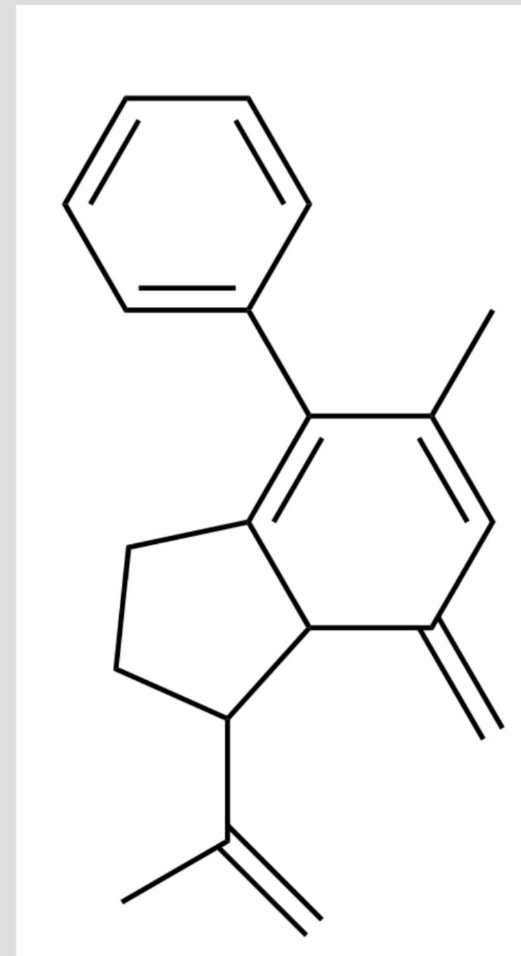
Good map quality



Ligand identification





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Diffraction Anisotropy

- The GΦL workflows have an initial characterisation data collection to derive the orientation matrix
  - Normally 6°, but increased to 12° for this ALBA visit
- With Ian Tickle, re-processed characterisation datasets with STARANISO
  - Can diffraction anisotropy be predicted before the crystal's dose budget has been used?

Diffraction Anisotropy

- An example:

Collection	Diffraction limits	B11, B33
Char	1.7, 2.2	22, 51
Main	1.5, 2.0	25, 52

•The symmetry in this case is high, so the direction of anisotropy is constrained.
•The strongest and weakest diffracting directions have been correctly identified from the characterisation dataset alone; for this symmetry that is all that is needed.

- More investigation with low-symmetry systems is needed.