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Things we might do better together
as prompted by the integration of GΦL workflows

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Global Phasing Ltd, Cambridge

MXCuBE meeting, ALBA
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- **Introduction**
- (Re)centring and calibration
- Other opportunities

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GΦL Workflows - automated expertise

- Make it quick and easy to acquire consistently high-quality data
  - Includes setting the (default) parameter values for you
- Calculation and execution of optimised multi-sweep acquisition strategies in real-time
- Uses full range of κ values, setting ω ranges to minimise goniostat shadows
- Bespoke processing that combines sweeps, and corrects for remaining shadows

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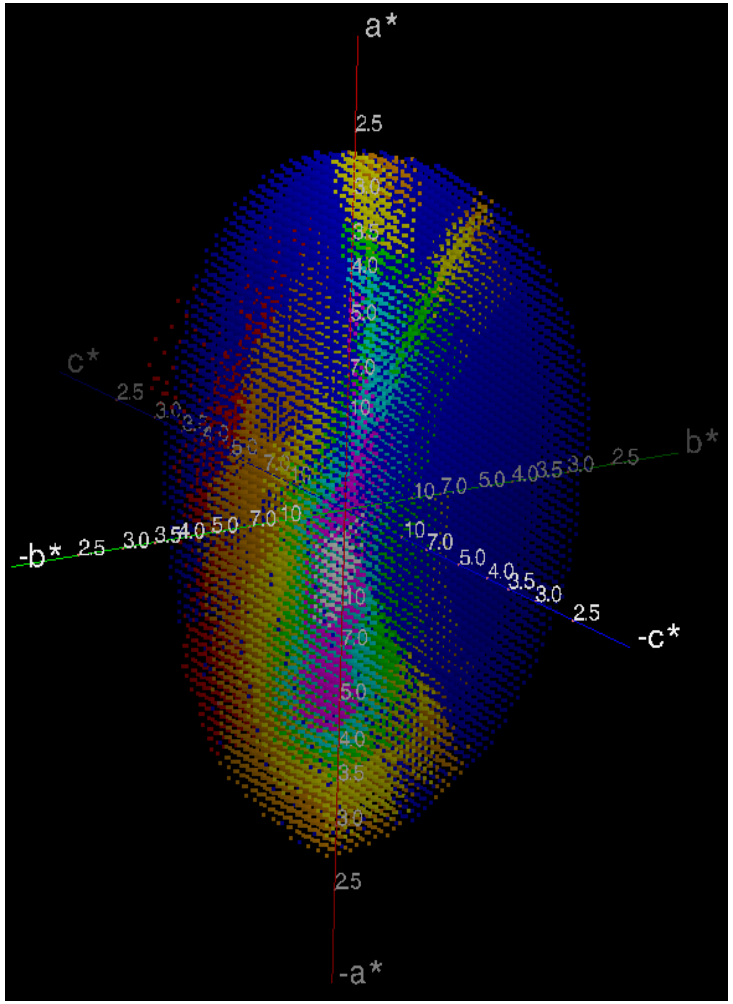
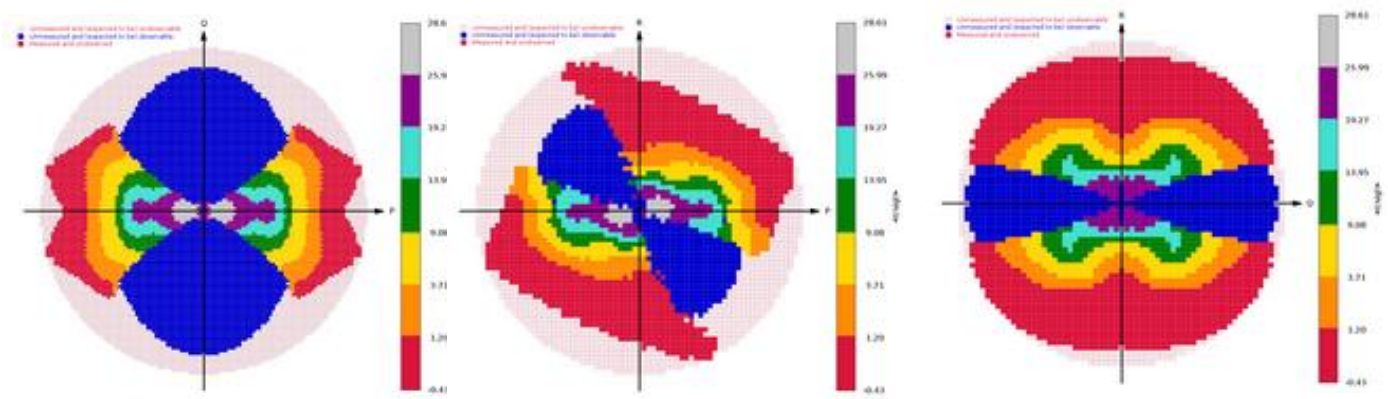
At a minimum to avoid this



Covid-related deposition (6W9C, 01/04/2020)

- C2, 2.7Å. 57% complete, 2.5 redundancy,
- High background, (40 counts), strong radiation damage.



Reflection I/σ – reciprocal space

Red = Unobservable reflections

Blue = Missed: not measured but expected to be observable

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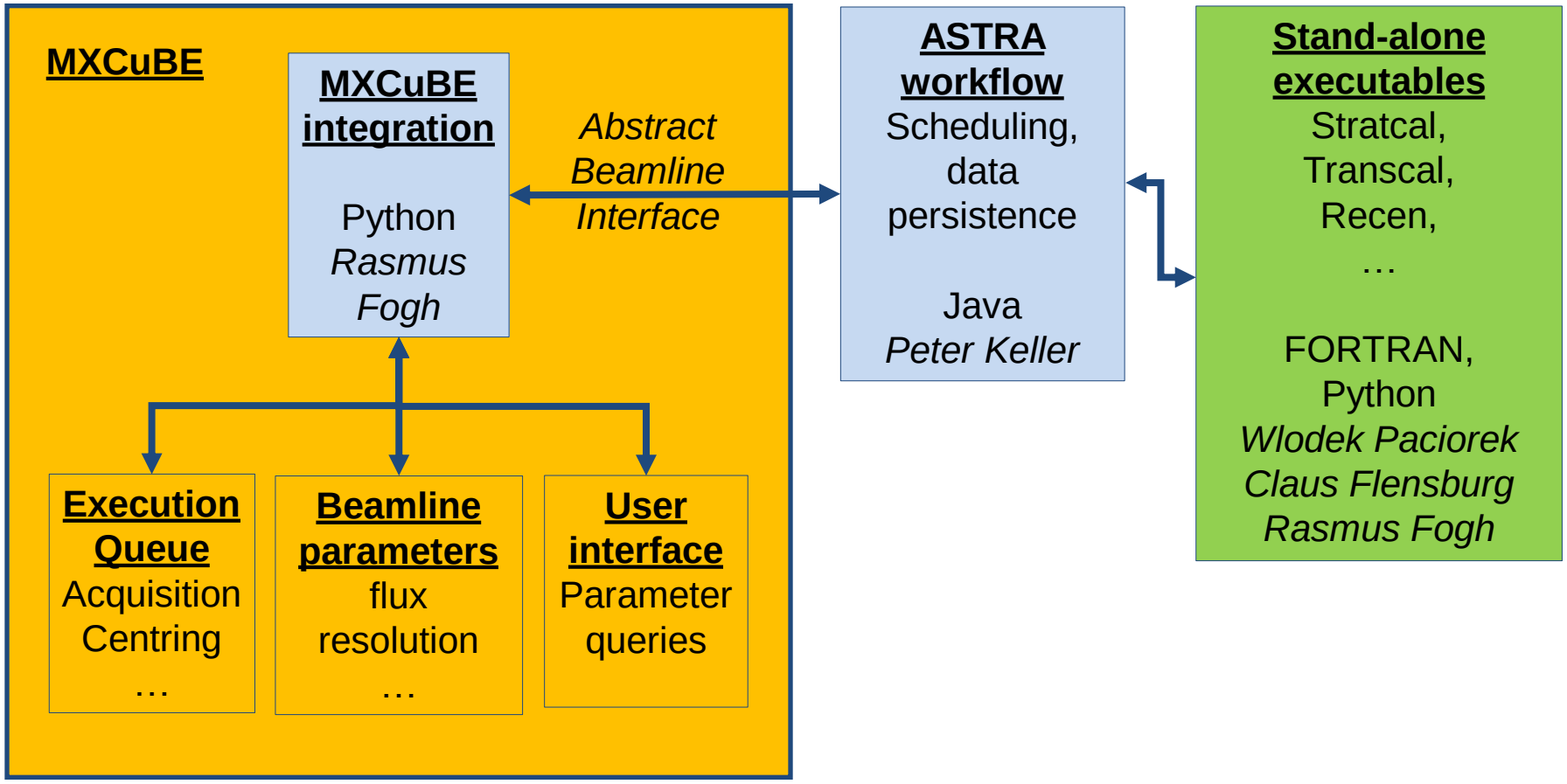
Program structure



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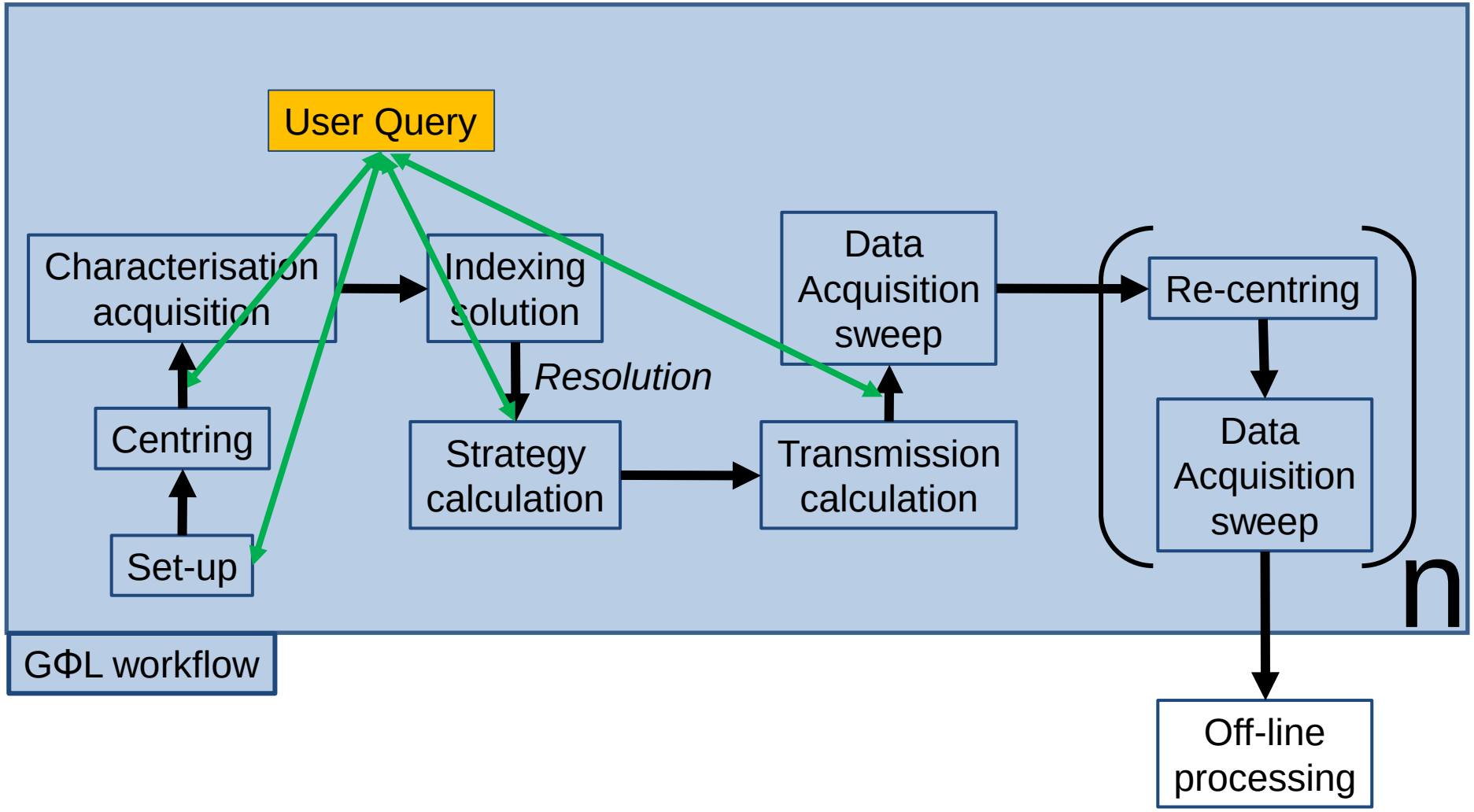
GΦL workflow actions



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Centring and re-centring

- The first centring is always needed to decide which part of the crystal to use
  - Various automation protocols are in use: tricky job.
- For multi-sweep experiments you need **re**-centring
- New centrings can be calculated – but mechanics of goniostat only allows 7-10µm reproducibility at best.
- Re-centring quicker than first centring, since you can start close, at calculated values

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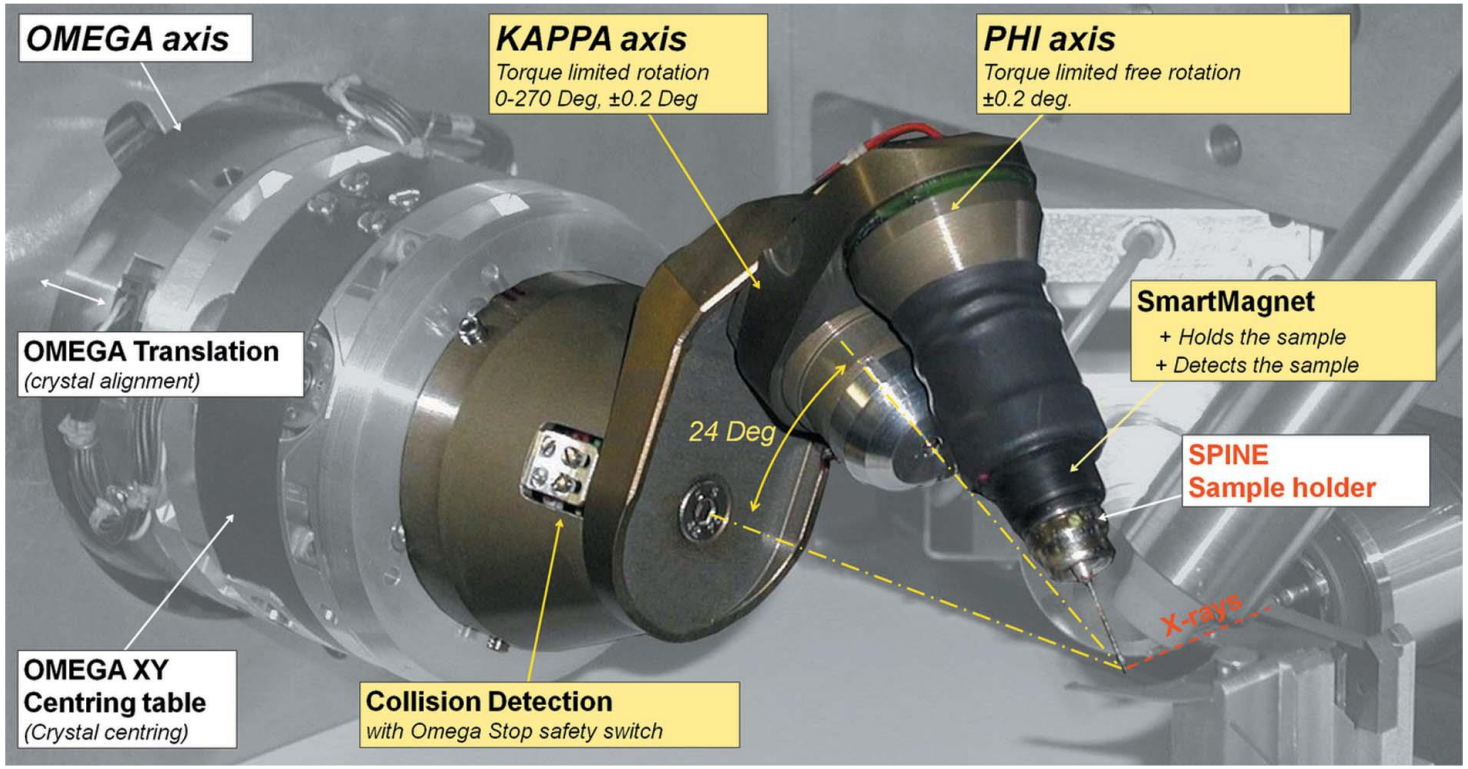
Centring changes with on κ and φ

- Centring puts the crystal on the ω axis and in the beam.
- New centring needed when κ and φ change, as κ and φ axes do not go through the crystal position
- Axis directions and offsets must be known and calibrated – (mis)alignment can change after goniostat head is taken on and off



From Brockhauser et al. (2011) without permission

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Recentring calculations

- EMBL-HH (Gleb Bourenkov, Ivars Karpics) have the MiniKappaCorrection procedure
  - Based on STAC
  - Uses goniostat coordinate system with nominal geometry
  - Has been MXCuBE code since 2015; can be executed automatically on reorientation
  - Still not universally adopted
- GΦL has its own system: transcal/diffractcal.
  - Includes calibration for both axis directions and detector geometry
  - Requires GΦL release
  - In laboratory coordinate system – necessary to calculate and to correct for goniostat shadows
- The two descriptions are mathematically equivalent. One set of translational correction parameters can be calculated from the other – axis directions differ.

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GΦL - calibration

- The GΦL release comes with calibration workflows and analysis procedures
- Calibrating axes and detector plane requires a robust high-quality high-symmetry reference crystal, and a long acquisition and processing
  - Germanate crystals are available (courtesy of Armin Wagner, DLS).
- Translational calibration requires 30-40 centrings on a test object
  - Tungsten pins with ball head are available (courtesy of SOLEIL)
  - With optical image analysis it is possible to do the translational calibration hands-off in a few minutes (Olof Svensson, MASSIF-1)
  - Could we share this implementation in MXCuBE?

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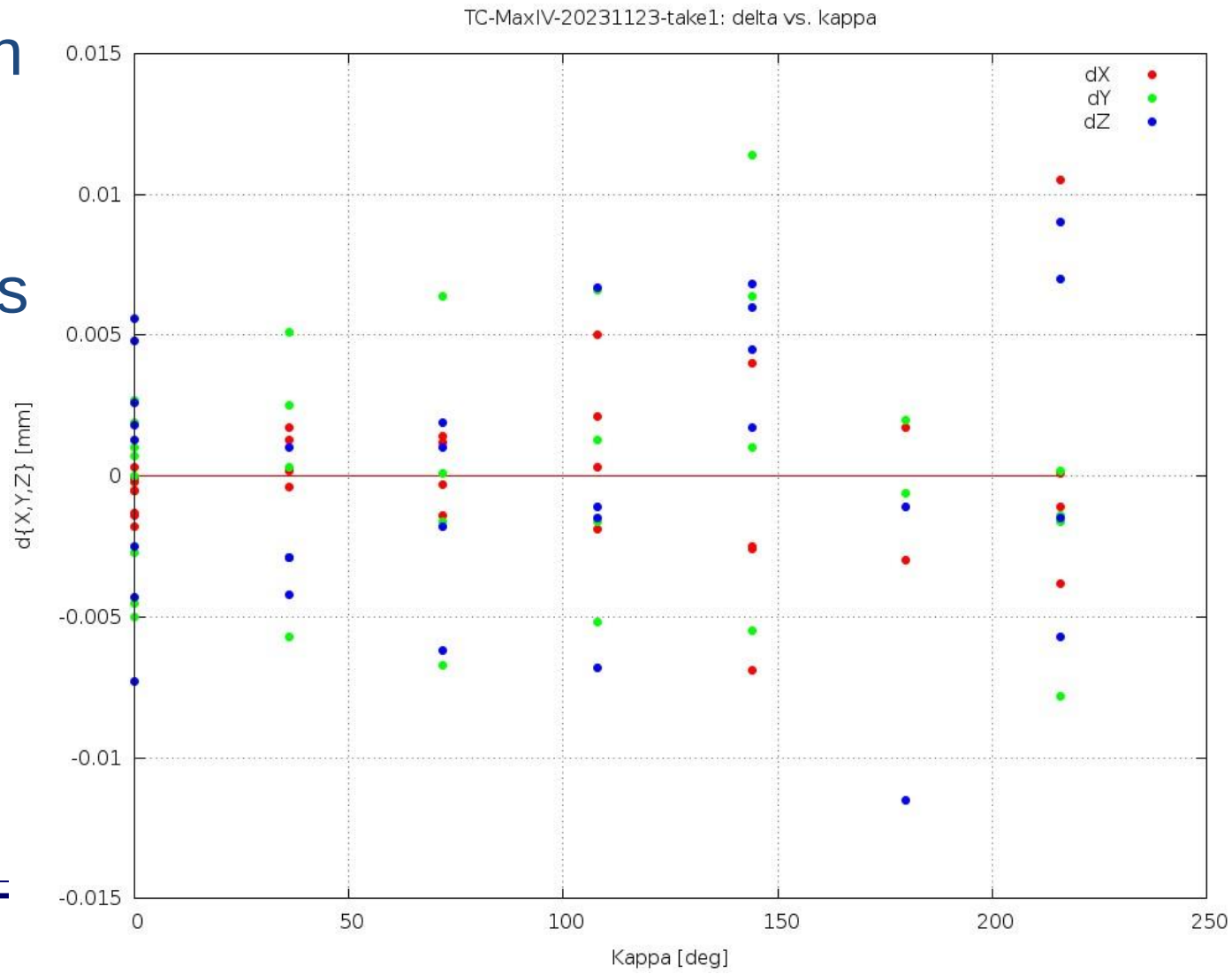
Translational calibration (Max IV)

Recent translational calibration at MAX IV (thanks to Jie Nan)

Analysis of 36 (8+28) centrings on a grid of κ,φ values

(residual/n_pts)^1/2:
RMSD(obs - pred) = 0.0072 mm



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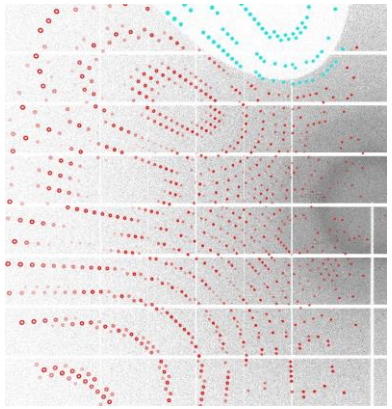
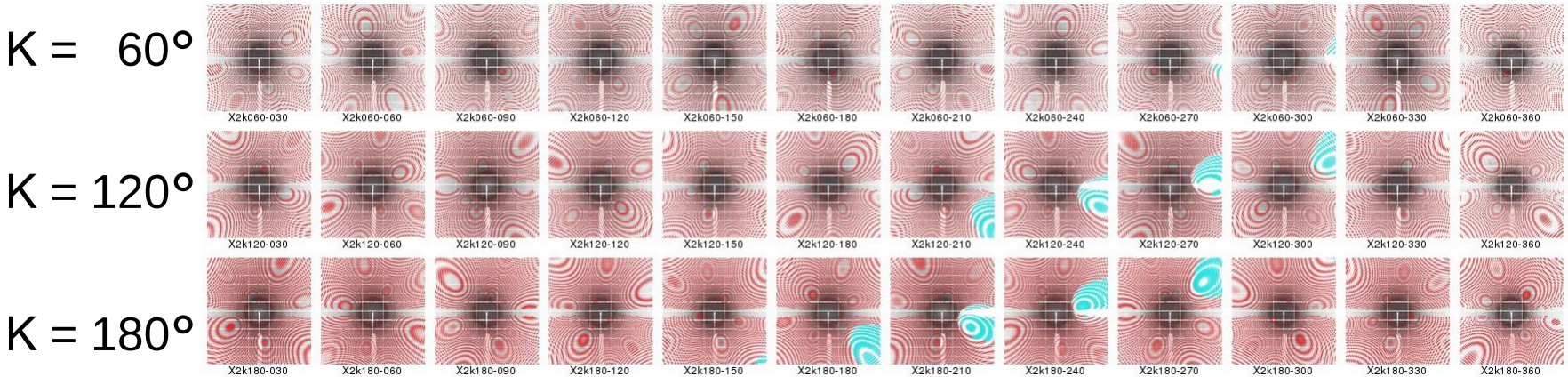
Goniostat shadows





ω: 30 ° - 360 °

- With high kappa angles and the detector close the goniostat casts shadows on the image
- Expected-but-missing reflections can lead to severe processing problems
- With GΦL recentring calculations, SimCal can predict dynamic shadows and autoPROC can mask out the missing reflections (in light blue)

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Setup - calibration

- Could we integrate the application of predicted recentring as standard behaviour for MXCuBE?
- To function reliably, either system requires calibration, which must be kept up to date on the beamline side. This is a matter of synchrotron procedures
- Could we agree on procedures?

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Automatic recentring procedures

- Recentring should be faster than centring, since you start close to the correct centring.
- X-ray recentring – you only need a small grid
- Optical recentring – ask Martin Savko
- MASSIF-1 use their workflow (MXPress) for X-ray recentring

- Could we agree on a standard interface, so you could use the same calls at any beamline with a bit of configuration?

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Estimating workflow parameters

- All parameters should be set automatically
  - in full automation this is required
  - in manual operation to simplify operation (better starting values)
- From current/default values: Image width, exposure time, wavelength, …
- Calculated: Transmission, dose budget, indexing solutions

- Currently only resolution, centring (and MAD wavelengths) are mandatory
- Could we estimate resolution reliably for each sample?

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Resolution from Dozor?

- Dozor is installed on most beamlines
  - But differently on each?
- GΦL thick characterization (5 x 1.2°) should be enough for a reliable resolution estimate
- Could we make a standard DOZOR integration, so that the results (including estimated resolution) could be queried by MXCuBE?

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HDF5 and multi-trigger

- Detectors must be re-armed between sweeps
  - which takes time and creates a new master file each time
- Multi-trigger mode allows multiple sweeps without re-arming
  - which saves time for (GΦL) multi-sweep characterisation, inverse-beam, and wavelength interleaving
- Multi-trigger present at multiple sites
  - but not apparently standardised
- Could we make a standard multi-trigger implementation?

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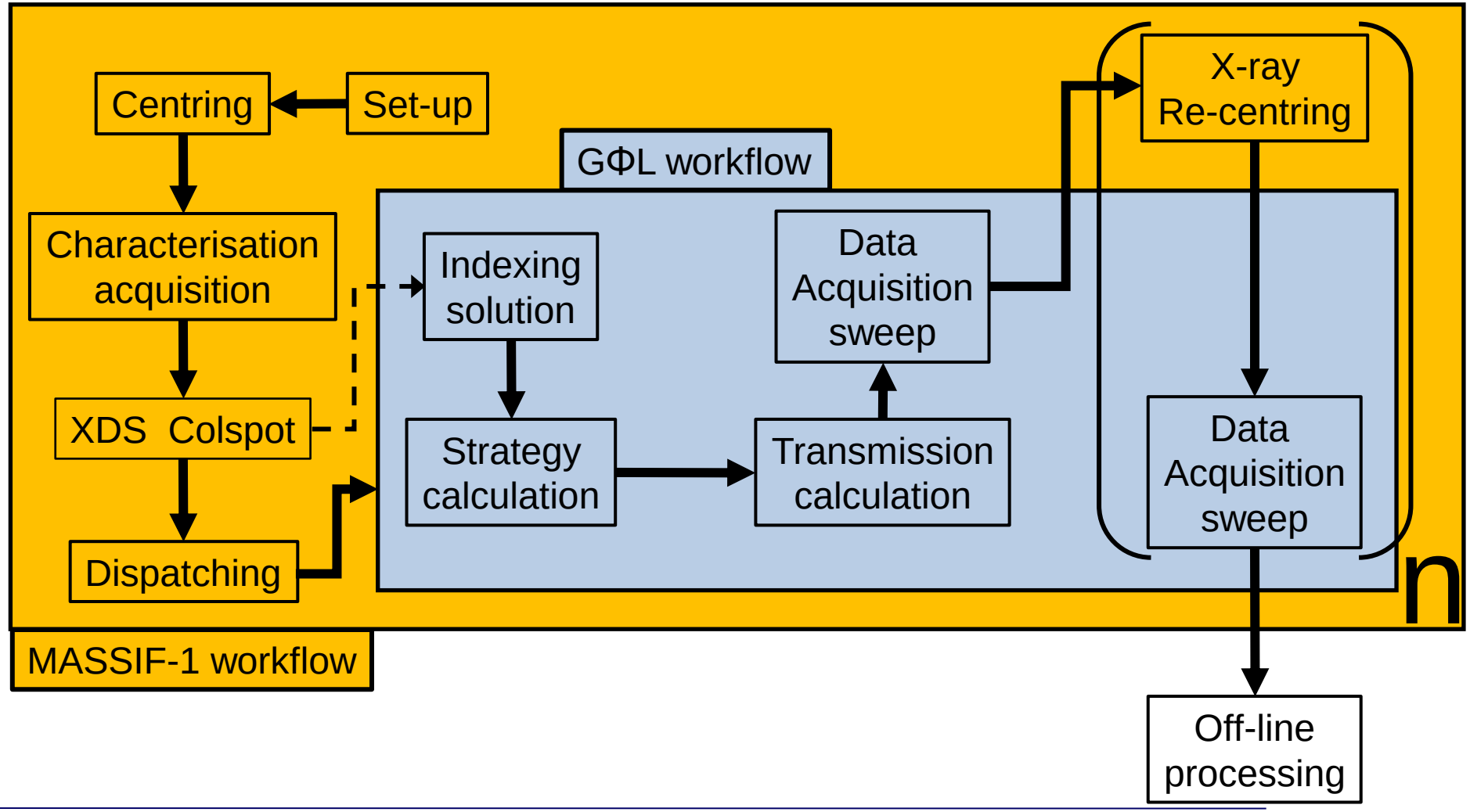
GΦL workflow on MASSIF-1



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Unattended operation

- The MXCuBE queue can run by itself once loaded
- GΦL workflows will work in full automation mode from a single input parameter dictionary
  - The main missing capabilities are resolution and centring (previously addressed)
- Can we (some of us?) collaborate on how to load and schedule the jobs for unattended operation?

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Acknowledgements

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