

Merging in 2D

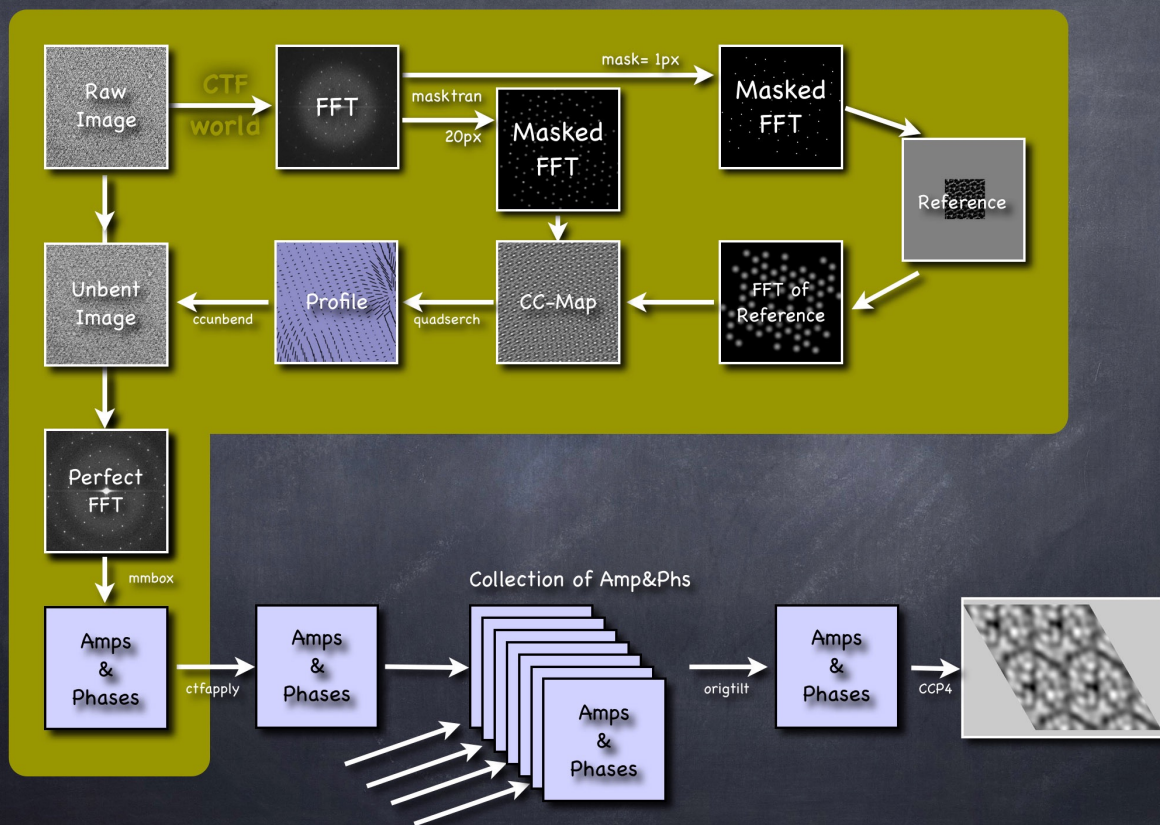
Henning Stahlberg,
Biozentrum, Uni Basel, Switzerland
c-cina.org

2dx Workshop
Basel, August 23-26, 2016

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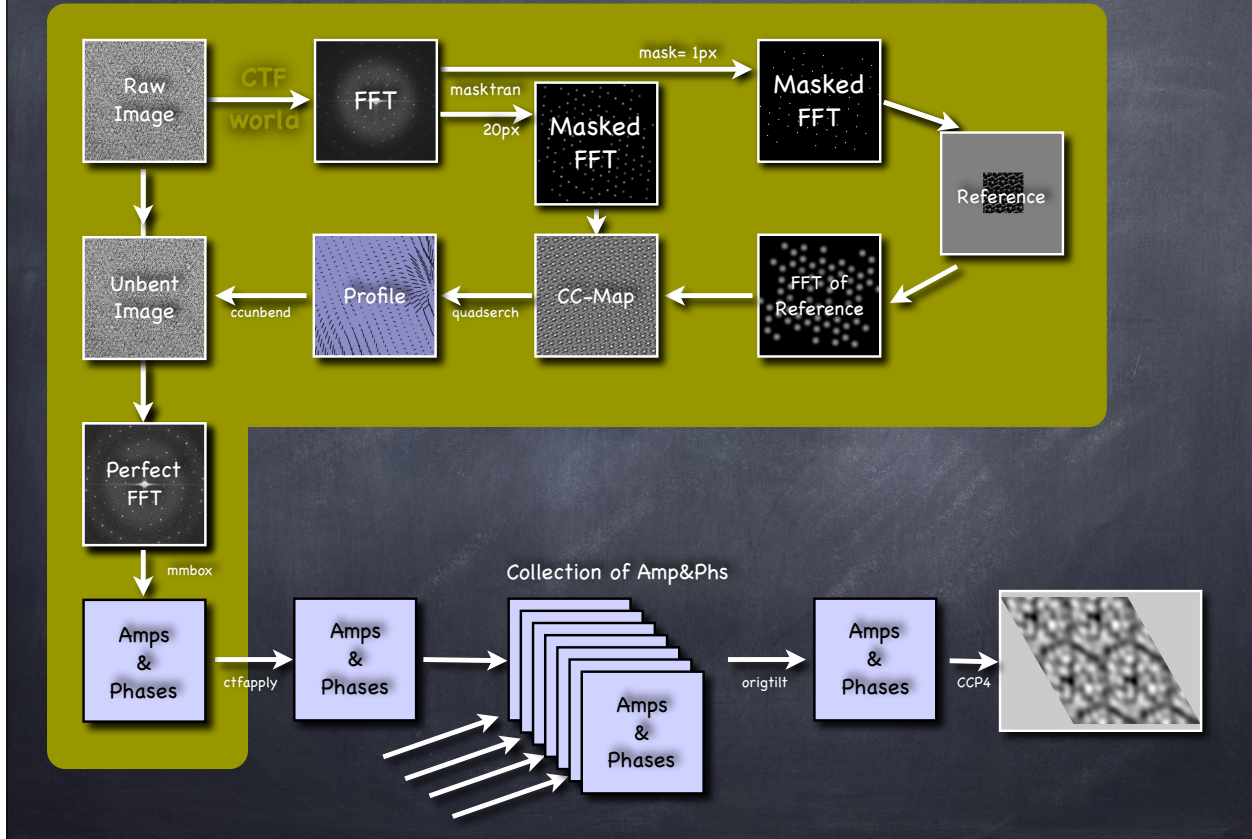
Algorithm Non-tilted



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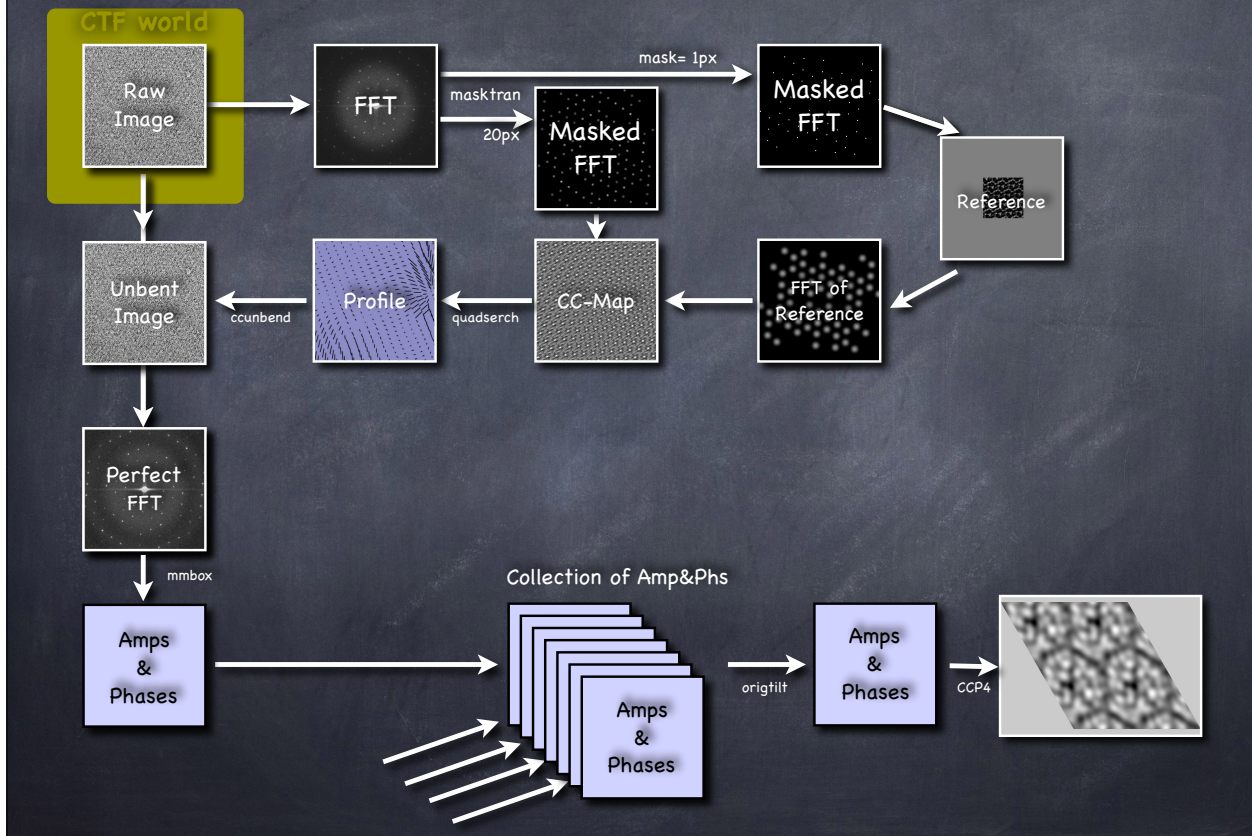
Algorithm Non-tilted



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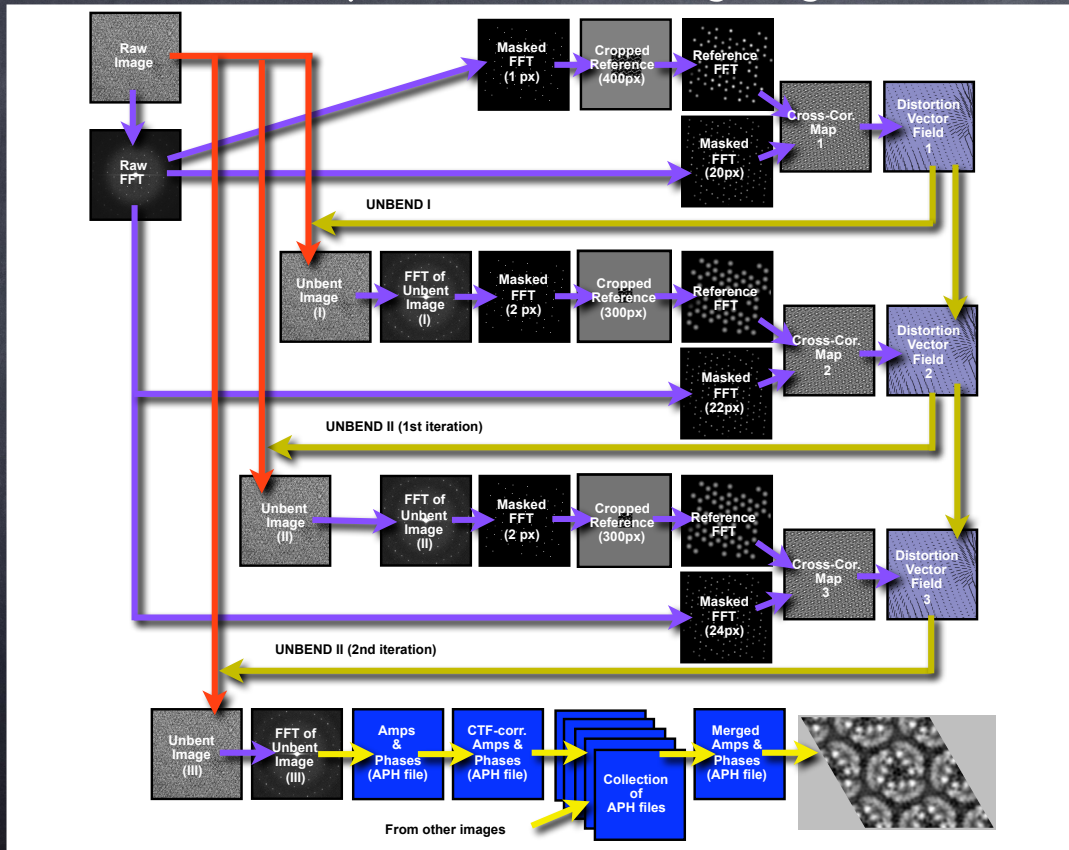
Algorithm for tilted or non-tilted data



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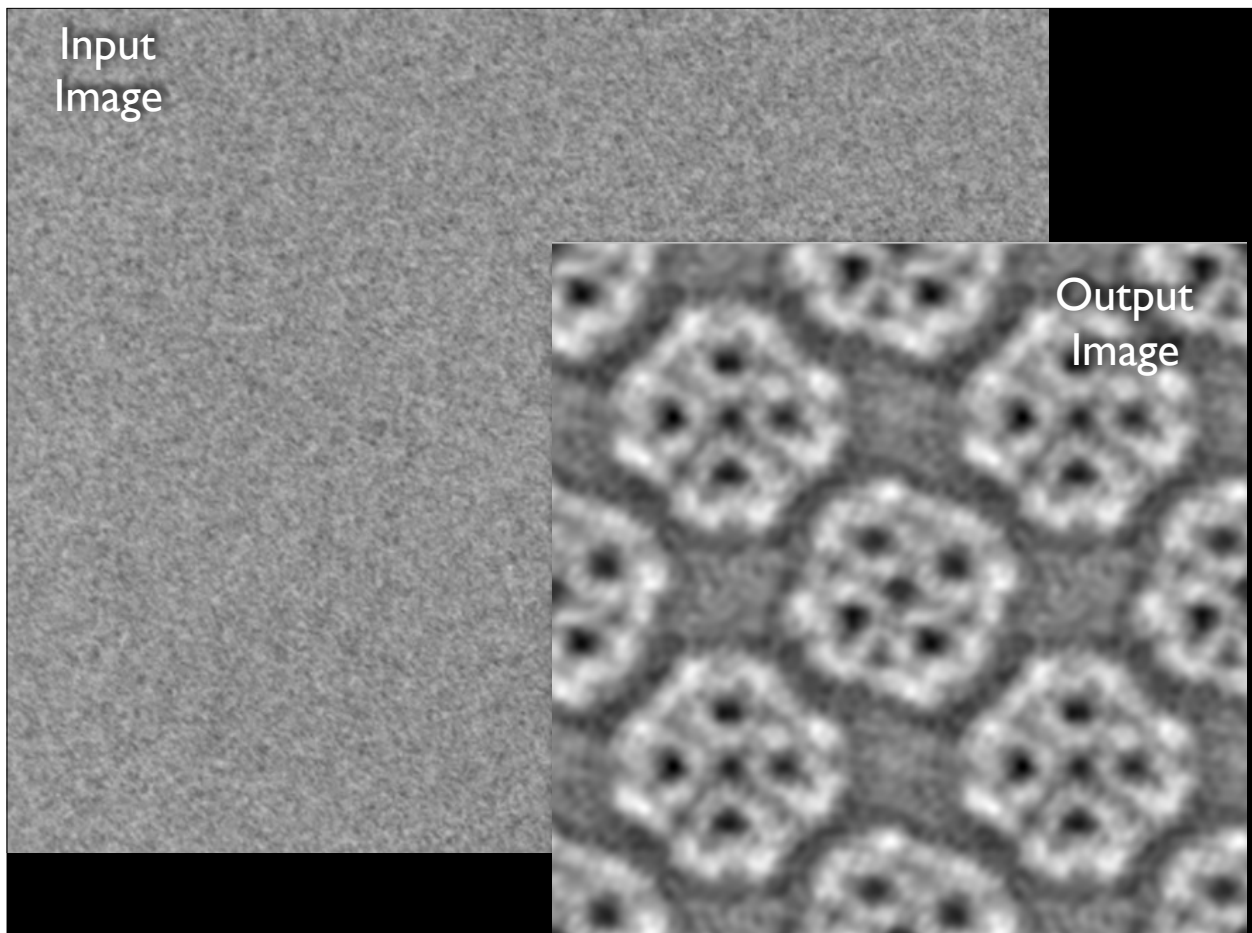
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More Complete Unbending Algorithm



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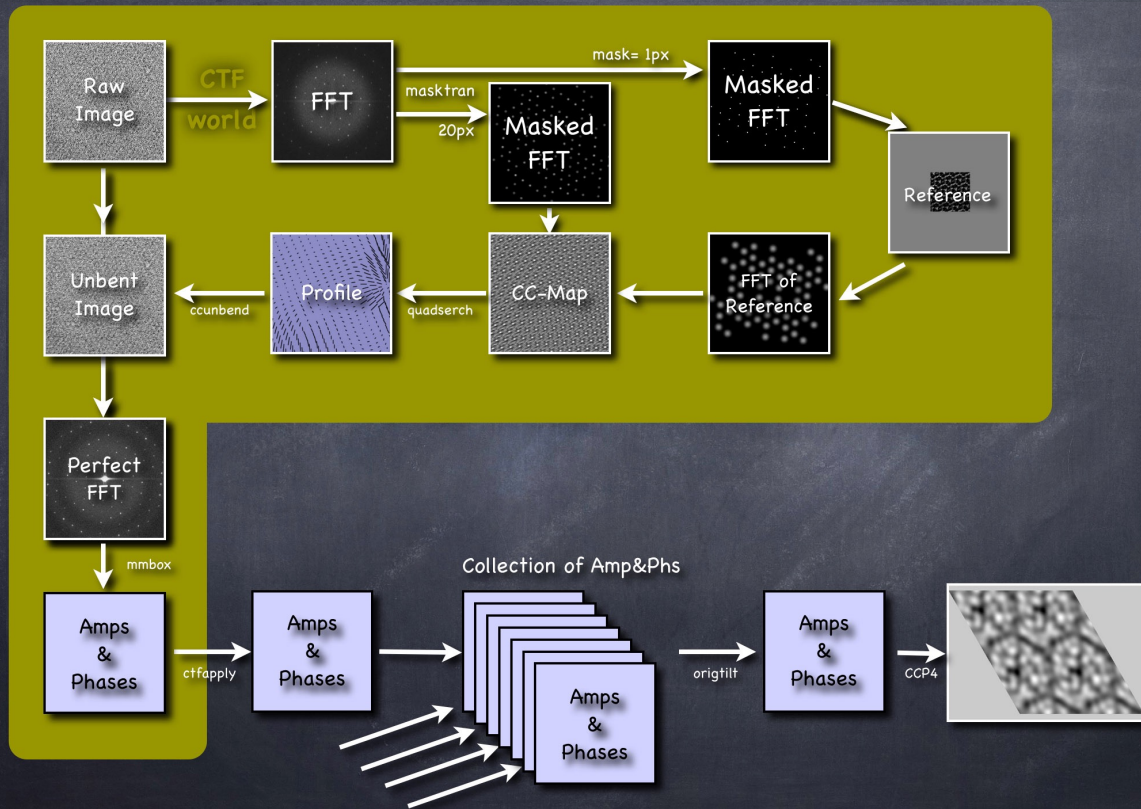
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Algorithm Non-tilted



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Project Management

- Launch *2dx_gui*
- Import only **ONE** image
- Open and process it, save database as project-default
- Import all other images into *2dx_gui*
- Process them all automatically (This will need finetuning...)
- Merge in 2D
- Merge in 3D

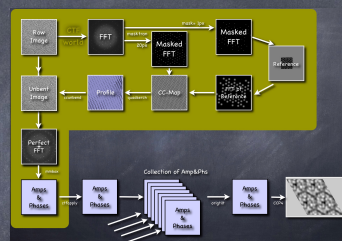
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File Formats and Flow

mmbox => ctfapply => origtilt => avramphs => f2mtz

mmbox evaluates the Fourier transform of the unbent crystal image and produces a list of Amplitudes, Phases, and Background Amplitudes.



mmbox:

- takes FFTIR/cor\${imagenam}.fft.mrc
- generates APH/\${imagenam}.fou.nolimit.aph

Filename: APH/\${imagenam}.fou.nolimit.aph

1: Header

2: H K AMP PHS IQ BCK CTF

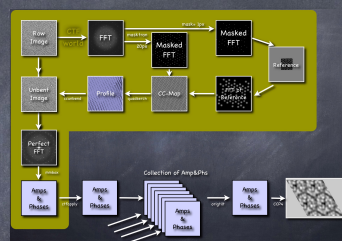
Example:

| 2: | H | K | AMP | PHS | IQ | BCK | CTF |
|--------|---|---|-------|-------|----|------|-----|
| 655201 | gf06552, Unbend2, Mon Jun 16 00:33:23 CEST 2008 | | | | | | |
| 0 | 1 | | 0.0 | 60.9 | 9 | 72.2 | 0.0 |
| 0 | 2 | | 165.8 | 330.8 | 3 | 49.3 | 0.0 |
| 0 | 3 | | 17.1 | 52.3 | 8 | 39.3 | 0.0 |
| 0 | 4 | | 151.8 | 123.1 | 2 | 36.5 | 0.0 |
| 0 | 5 | | 411.8 | 127.5 | 2 | 50.9 | 0.0 |

File Formats and Flow

mmbox => ctfapply => origtilt => avramphs => f2mtz

ctfapply calculates the CTF correction factor for each reflection, and produces a CTF-corrected APH file, where only the phases are potentially "flipped". This is one file for each image.



ctfapply:

- takes \${imagenam}.fou.nolimit.aph
- generates \${imagenam}.fou.ctf.nolimit.aph

Filename: \${imagenam}.fou.ctf.nolimit.aph

1: Header

2: H K AMP PHS IQ BCK CTF

AMP is not changed

PHS is already CTF-applied, possibly +180.

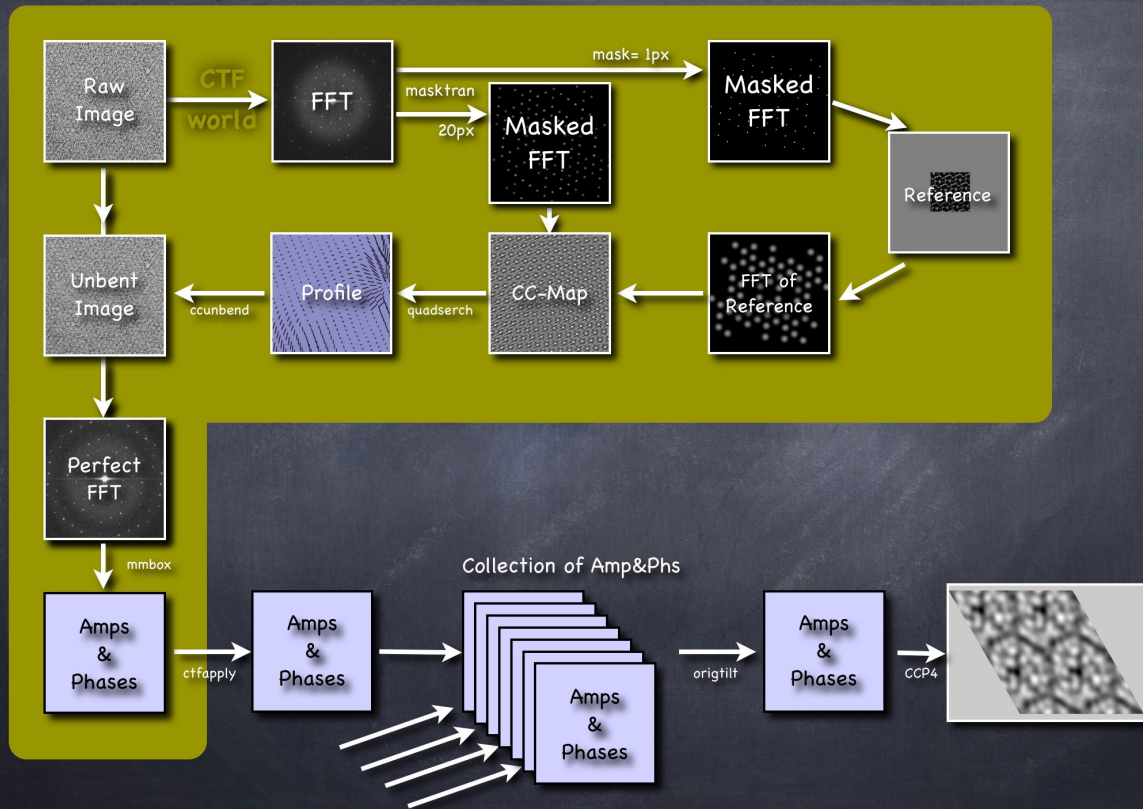
CTF lists the contrast.

Only phase (sign) was already applied.

Example:

| 2: | H | K | AMP | PHS | IQ | BCK | CTF |
|--------|--------------------------------------|---|-------|-------|----|------|--------|
| 655201 | gf06552, Thu Jul 3 22:49:19 PDT 2008 | | | | | | |
| 0 | 1 | | 0.0 | 240.9 | 9 | 72.2 | -0.096 |
| 0 | 2 | | 165.8 | 150.8 | 3 | 49.3 | -0.172 |
| 0 | 3 | | 17.1 | 232.3 | 8 | 39.3 | -0.295 |
| 0 | 4 | | 151.8 | 303.1 | 2 | 36.5 | -0.459 |
| 0 | 5 | | 411.8 | 307.5 | 2 | 50.9 | -0.648 |

Algorithm Non-tilted



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File Formats and Flow

mmbox => ctfapply => origilt => avramphs => f2mtz

origilt takes the several files from mmbox and combines them into one single large file merge.aph that contains AMP, PHS, original image number, and further data for each reflection. This is one single file for the entire project.

origilt:

- takes several APH/\${imagename}.fou.cor.aph
- generates APH/merge.aph, which now has for each spot potentially many entries

Filename: APH/merge.aph

1: Header

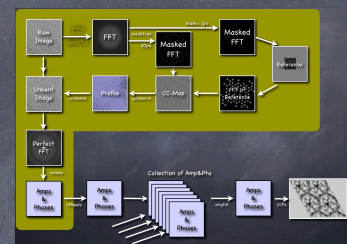
2: H K Z* AMP PHS NO IQ WEIGHT BACK CTF

AMP is the raw measured value, not yet changed

PHS is CTF phase flipped, and has also the PhaseOrigin applied to it

Example:

| | | | | | | | | | | |
|------|---|---------|---------|----------|--------|----|----------|---------|--------|--|
| 1001 | | | | | | | | | | |
| 1 | 0 | -0.0000 | 70.8670 | -88.1100 | 65551 | -6 | 0.078805 | 47.8734 | -0.083 | |
| 1 | 0 | 0.0000 | 112.300 | -21.6200 | 6543 | 6 | 0.192624 | 74.5000 | -0.093 | |
| 1 | 0 | 0.0000 | 25.0594 | -131.430 | 65551 | 8 | 0.027867 | 49.3105 | -0.088 | |
| ... | | | | | | | | | | |
| 2 | 0 | -0.0000 | 120.801 | -160.000 | 6557 | 4 | 0.192729 | 48.7613 | -0.165 | |
| 2 | 0 | -0.0000 | 156.540 | 169.480 | 665202 | -2 | 0.276022 | 34.6150 | -0.154 | |
| 2 | 0 | -0.0000 | 202.253 | 170.080 | 655201 | -3 | 0.375352 | 51.5399 | -0.167 | |
| ... | | | | | | | | | | |
| 2 | 0 | 0.0013 | 411.287 | -178.300 | 7032 | 1 | 0.440457 | 50.2215 | -0.383 | |
| 2 | 0 | 0.0013 | 313.198 | -178.300 | 8032 | 2 | 0.307053 | 40.1020 | -0.385 | |
| 2 | 0 | 0.0016 | 298.834 | -161.900 | 5685 | 2 | 0.360523 | 61.4043 | -0.198 | |



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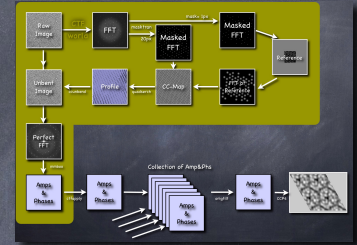
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File Formats and Flow

mmbox => ctffapply => origtilt => avramphs => f2mtz

avramphs takes the merge.aph file that contains for each reflection several measurements, and averages these into one AMPLitude and PHAsE value for each reflection.

This is one single file for the entire project.



avramphs:

- uses zminmax = -0.025...0.025, to make sure only non-tilted data participate.
- takes APH/merge.aph
- generates APH/avrg2D.hkl

Filename: APH/avrg2D.hkl

1: merging number

2: H K L

AMP

PHS

FOM

AMP is an average of several spots, and the values are now amplitude-CTF corrected (without inverting the sign).

PHS is an average of several spots, which are already CTF and phaseorigin corrected

FOM is between 0 and 100 and quantifies the reliability of the spots.

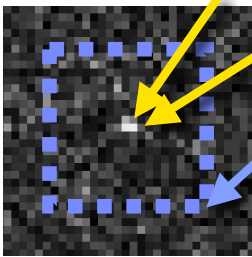
Example:

| merging number | H | K | L | AMP | PHS | FOM |
|----------------|---|---|---|---------|----------|---------|
| 1001 | | | | | | |
| 1 | 0 | 0 | | 196.833 | -64.0629 | 96.9797 |
| 1 | 1 | 0 | | 1018.19 | -6.10076 | 99.9556 |
| 1 | 2 | 0 | | 162.905 | -177.142 | 98.7745 |
| 1 | 3 | 0 | | 262.806 | -179.891 | 99.5678 |
| ... | | | | | | |
| 2 | 0 | 0 | | 453.627 | -177.303 | 99.8751 |
| 2 | 1 | 0 | | 109.832 | 4.30570 | 94.4169 |
| 2 | 2 | 0 | | 1278.94 | 177.975 | 99.9585 |

How to average Amplitudes

FT Readout:

H, K, Z*, Amplitude, Phase, BackgroundAmplitude



For each reflection H,K,z* :

$$SNR = Amp / Back$$

$$FOM = \frac{I_1(SNR)}{I_0(SNR)}$$

Simple Approach:

$$AvgAmp = \frac{\sum Amp}{n}$$

Better:

$$AvgAmp = \frac{\sum Amp * FOM}{\sum FOM}$$

In 2dx now:

$$AvgAmp = \frac{\sum \frac{Amp * CTF}{Back^2}}{\sum \left(\frac{CTF^2}{Back^2} + \frac{1}{Amp^2} \right)}$$

How to average Phases

R. Henderson et al. / Structure of purple membrane from Halobacterium halobium

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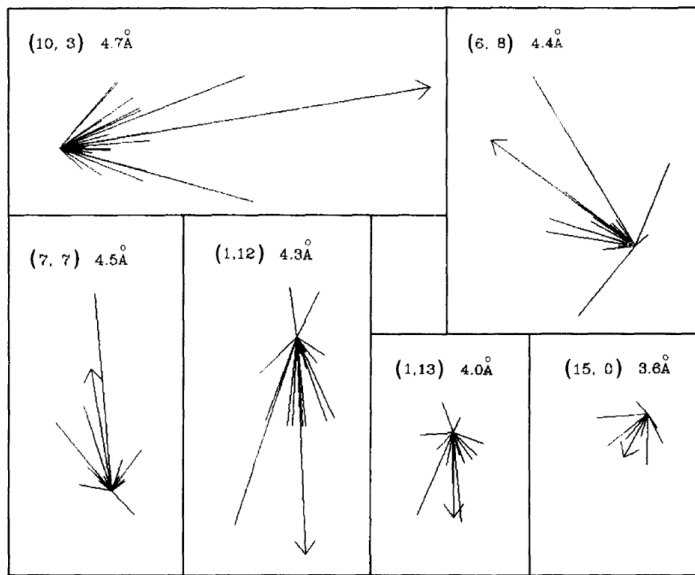


Fig. 7. Graphical comparison of all phases determined for six spots with resolution beyond 4.7 Å. Phases are plotted as vectors on a polar diagram with the length of each vector being proportional to $1/IQ^2$. Thus the strongest spots show up as longer vectors. The result of the summation of all the vectors is also shown as a vector, but this is plotted at 1/4 of its true length. The direction of this vector is our best estimate of the phase of the Fourier component in the structure, and its length can be used to provide an estimate of the error.

$$AvgPhs = \tan^{-1} \left(\frac{\sum \cos(Pha) * SNR(FOM)}{\sum \sin(Pha) * SNR(FOM)} \right)$$

Averaging of data from different images in the MRC/2dx software is done in Fourier space by dealing with each reflection individually:

Amplitudes and Phases are averaged, taking $1/IQ^2$ (or now FOM) as weight.

On the left, the Phase averaging is shown graphically.

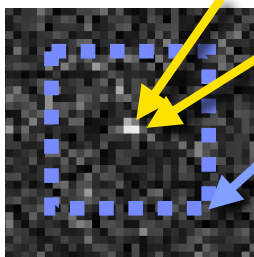
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How to average Figures of Merit (FOM)

FT Readout:

H, K, Z*, Amplitude, Phase, BackgroundAmplitude



For each reflection H,K,z* :

$$SNR = Amp / Back$$

$$FOM = \frac{I_1(SNR)}{I_0(SNR)}$$

$$FOM = lut(SNR)$$

$$lut(IQ) = 0.990, 0.982, 0.939, 0.870, 0.763, 0.630, 0.505, 0.124$$

$$FOM = \exp\left(-\frac{\sigma_{Phase}^2}{2}\right)$$

$$FOM = \cos(\max(\sigma_{Phase}, 90))$$

Use Lookup Table to switch between FOM and XARG

$$AvgFOM = \frac{I_1(AvgSNR)}{I_0(AvgSNR)}$$

I = modified Bessel Function of the first kind

$$AvgSNR = \sqrt{\left(\sum \cos(Pha) * SNR(FOM)\right)^2 + \left(\sum \sin(Pha) * SNR(FOM)\right)^2}$$

G. Sim, Acta Cryst. 13, 511 (1960)

D. Blow & F. Crick, Acta Cryst. 12, 795 (1959)

G. Sim, Acta Cryst. 12, 813 (1959)

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Filename: APH/merge.aph

1: Header

2: H K Z* AMP PHS NO IQ WEIGHT BACK CTF
AMP is the raw measured value, not yet changed
PHS is CTF phase flipped, and has also the PhaseOrigin applied to it

Example:

| | H | K | Z* | AMP | PHS | NO | IQ | WEIGHT | BACK | CTF |
|------|---|---------|----|---------|----------|--------|----|----------|---------|--------|
| 1001 | | | | | | | | | | |
| 1 | 0 | -0.0000 | | 70.8670 | -88.1100 | 65551 | -6 | 0.078805 | 47.8734 | -0.083 |
| 1 | 0 | 0.0000 | | 112.300 | -21.6200 | 6543 | 6 | 0.192624 | 74.5000 | -0.093 |
| 1 | 0 | 0.0000 | | 25.0594 | -131.430 | 65551 | 8 | 0.027867 | 49.3105 | -0.088 |
| ... | | | | | | | | | | |
| 2 | 0 | -0.0000 | | 120.801 | -160.000 | 6557 | 4 | 0.192729 | 48.7613 | -0.165 |
| 2 | 0 | -0.0000 | | 156.540 | 169.480 | 665202 | -2 | 0.276022 | 34.6150 | -0.154 |
| 2 | 0 | -0.0000 | | 202.253 | 170.080 | 655201 | -3 | 0.375352 | 51.5399 | -0.167 |
| ... | | | | | | | | | | |
| 2 | 0 | 0.0013 | | 411.287 | -178.300 | 7032 | 1 | 0.440457 | 50.2215 | -0.383 |
| 2 | 0 | 0.0013 | | 313.198 | -178.300 | 8032 | 2 | 0.307053 | 40.1020 | -0.385 |
| 2 | 0 | 0.0016 | | 298.834 | -161.900 | 5685 | 2 | 0.360523 | 61.4043 | -0.198 |

avramphs:

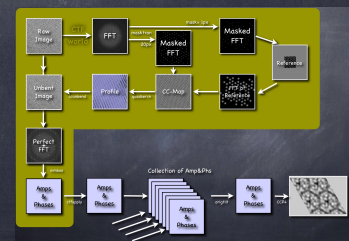
Filename: APH/avrg2D.hkl

1: merging number

2: H K L AMP PHS FOM
AMP is an average of several spots, and the values are now amplitude-CTF corrected (without inverting the sign).
PHS is an average of several spots, which are already CTF and phaseorigin corrected
FOM is between 0 and 100 and quantifies the reliability of the spots.

Example:

| | H | K | L | AMP | PHS | FOM |
|------|---|---|---|---------|----------|---------|
| 1001 | | | | | | |
| 1 | 0 | 0 | | 196.833 | -64.0629 | 96.9797 |
| 1 | 1 | 0 | | 1018.19 | -6.10076 | 99.9556 |
| 1 | 2 | 0 | | 162.905 | -177.142 | 98.7745 |
| 1 | 3 | 0 | | 262.806 | -179.891 | 99.5678 |
| ... | | | | | | |
| 2 | 0 | 0 | | 453.627 | -177.303 | 99.8751 |
| 2 | 1 | 0 | | 109.832 | 4.30570 | 94.4169 |
| 2 | 2 | 0 | | 1278.84 | 177.975 | 99.9585 |



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File Formats and Flow

mmbox => ctfapply => origtilt => avramphs => f2mtz

centric and **hklsym** apply boundary conditions to reflection values.
Centric enforces for certain symmetries that reflections of non-tilted crystals must have phases of either 0° or 180° .
Hklsym averages symmetry-related reflections.

f2mtz transforms the AMP&PHS file into a binary MTZ file.

f2mtz:

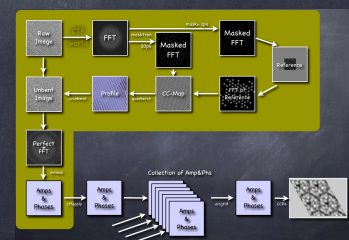
- takes APH/sym2D.hkl
- generates merge2D.mtz

Filename: merge2D.mtz

SYMMETRY \${CCP4_SYM}

ABOUT H K L F PHI FOM

CTYPOUT H H H F P W



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File Formats and Flow

mmbox => ctfapply => origilt => avramphs => f2mtz

cent and hklsym apply boundary conditions and reflection values.
Centrifuges for cryo-EM systems are designed to minimize non-tilt
crystals must have phases of either 0° or 180°.
Hklsym averages symmetry-related reflections.
f2mtz transforms the AMP&PHS file into a binary MTZ file.

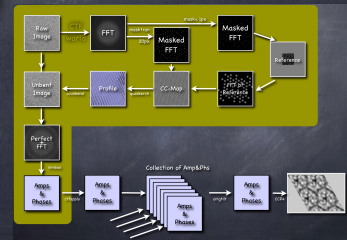
File Workflow is described on:

f2mtz:

- takes APH/sym2D.hkl
- generates merge2D.mtz

```
Filename: merge2D.mtz
SYMMETRY ${CCP4_SYM}
ABOUT H K L F PHI FOM
CTYPOUT H H H F P W
```

2dx.org



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2dx Software
2dx User Manual (.pdf)
Manual
Explanation of the 2dx GUI
Beginning an Image Processing Project
Status Pane
Processing an individual image
The Full-Screen Image Navigator
Refining a lattice
Refining the CTF
Spot Selection
Tilt Geometry Definition
ML algorithm applied to 2d crystals

Data Flow
Processing in 2dx_image of ONE image
Merging in 2dx_merge in 2D
Merging in 2dx_merge in 3D

Data Flow

by Admin — last modified Nov 14, 2011 11:40 AM

These pages describe the flow of data through the different programs. File names, formats and contents is listed. 2dx and the underlying MRC programs process and then evaluate images, and extract values for amplitudes and phases of Fourier spots. These are then handed over from one program to the next one, where each program does something to those values (e.g. correcting for the CTF, or merging several values together). Each program thereby unfortunately needs a different type of data that is therefore often in a different format, and the whole thing quickly gets very confusing. Most of these files are text files (ASCII), though, so that you can always open them with a text browser (vi, edit, SimpleText, or even Word (but don't save it back as "Word" file.doc file). We have here compiled a list of all the programs that are called in sequential order, and have listed the input and output files for these programs. For the output files we have then included a short description of the file format, followed by an example of how that should look like. This is only for reference purposes, in case somebody tries to follow the flow of data from one program to the next one.

Processing in 2dx_image of ONE image

This processing deals only with one single image and is therefore two-dimensional processing.

[Read More...](#)

Merging in 2dx_merge in 2D

This processing merges several images into one dataset. This here describes the merging of several non-tilted images into a two-dimensional dataset.

[Read More...](#)

Merging in 2dx_merge in 3D

This processing merges several images into one dataset. This here describes the merging of several tilted images into a three-dimensional dataset.

[Read More...](#)

2D Merging

- Select best image only
- Run “Merge Selected Images” to create reference
- Select all images with low sample tilt (< 7 deg)
- Run “Refine PhaseOrigins Once” to align against reference
- Run “Merge & Refine” 5 times to refine PhaseOrigins
- Run “Final Merge”, look at final map.
- Run “Quality Evaluation”
- Run “Generate Image Maps”, check phase origins of images.

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Final Merge in 2D

2D

- Initialization
- Merge Selected Images

3D

- Refine PhaseOrigins Once
- Merge & Refine (Iterative)
- Final Merge
- Quality Evaluation
- Generate Image Maps

DEPENDENT SCRIPTS

- 2dx_origtilt_merge.com
- 2dx_origtilt_aph_to_mtz.com
- 2dx_sym2spcgrp_sub.com
- 2dx_merge_makedirs
- initialize

Final Merge

Setup Level: Simplified

Search Parameters

Common Image Processing

Lower Resolution Limit (RESMIN) 90.0

Upper Resolution Limit (RESMAX, in Angstroms) 4.5

Merging resolution limit

0-Use global resolution limit

Resolution of the merged dataset for reference 4.5

Output (Double click for logbrowser) Verbosity Level: Low

```
==== 2dx_avrgamphs - to calculate statistics
==== Symmetry statistics here are only good in 2D.
==== Using therefore zminmax=-0.0025,0.0025.
====
```

| PHASE RESIDUALS IN RESOLUTION RANGES | | | | | | | | | | | | | |
|--------------------------------------|--------|------|------|------|------|------|------|------|------|------|------|------|-------------------------|
| # | DMIN | DMAX | IQ= | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | all | IQs |
| 1 | 1000.0 | 14.2 | 9.0 | 13.6 | 20.0 | 29.3 | 36.8 | 43.3 | 52.3 | 63.7 | 73.5 | 23.5 | |
| 1 | | | 1667 | 2727 | 1402 | 647 | 405 | 304 | 243 | 829 | 8224 | | |
| 2 | 14.2 | 10.1 | 10.1 | 13.3 | 18.3 | 25.5 | 32.7 | 38.2 | 46.4 | 53.7 | 26.7 | | |
| 2 | | | 332 | 1257 | 806 | 524 | 396 | 295 | 200 | 749 | 4559 | | |
| 3 | 10.1 | 8.2 | 8.5 | 14.0 | 21.4 | 29.3 | 39.3 | 43.1 | 54.9 | 59.7 | 40.1 | | |
| 3 | | | 24 | 313 | 532 | 531 | 388 | 318 | 248 | 1033 | 3387 | | |
| 4 | 8.2 | 7.1 | 0.0 | 11.4 | 21.7 | 30.3 | 45.1 | 48.8 | 57.5 | 66.6 | 53.4 | | |
| 4 | | | 0 | 36 | 108 | 263 | 298 | 268 | 195 | 1053 | 2221 | | |
| 5 | 7.1 | 6.4 | 0.0 | 22.1 | 34.8 | 41.5 | 43.4 | 56.9 | 60.1 | 72.0 | 62.3 | | |
| 5 | | | 0 | 1 | 44 | 150 | 214 | 215 | 197 | 1039 | 1860 | | |
| 6 | 6.4 | 5.8 | 0.0 | 0.0 | 16.3 | 48.0 | 52.2 | 58.4 | 65.0 | 74.2 | 67.0 | | |
| 6 | | | 0 | 0 | 15 | 98 | 168 | 181 | 183 | 980 | 1625 | | |
| 7 | 5.8 | 5.4 | 0.0 | 0.0 | 38.5 | 47.4 | 53.1 | 54.0 | 68.8 | 72.1 | 66.0 | | |
| 7 | | | 0 | 0 | 26 | 88 | 176 | 181 | 190 | 1042 | 1703 | | |
| 8 | 5.4 | 5.0 | 0.0 | 0.0 | 57.5 | 63.0 | 56.5 | 67.6 | 76.3 | 78.0 | 73.3 | | |
| 8 | | | 0 | 0 | 10 | 58 | 139 | 142 | 143 | 744 | 1236 | | |
| 9 | 5.0 | 4.7 | 0.0 | 5.8 | 65.2 | 61.2 | 63.5 | 69.7 | 78.5 | 75.7 | 73.4 | | |
| 9 | | | 0 | 3 | 11 | 85 | 103 | 138 | 144 | 953 | 1437 | | |
| 10 | 4.7 | 4.5 | 0.0 | 0.0 | 82.5 | 61.0 | 69.0 | 74.4 | 76.4 | 79.8 | 76.8 | | |
| 10 | | | 0 | 0 | 10 | 69 | 98 | 124 | 138 | 735 | 1174 | | |
| | | | | | 9.2 | 13.5 | 20.7 | 33.4 | 44.1 | 51.8 | 61.9 | 69.5 | |
| | | | | | 2023 | 4337 | 2964 | 2513 | 2385 | 2166 | 1881 | 9157 | |
| Overall: | | | | | | | | | | | | | |
| | | | | | | | | | | | | | Phaseresidual = 43.489 |
| | | | | | | | | | | | | | Number of spots = 27426 |

The last column shows the average phase residual in that resolution range, which is the mean weighted squared distance of the phase values from the averaged ψ (90 deg = random)

Results

| Parameter | Value |
|---------------------------|--------|
| JREFL | 54832 |
| JREFL_2D | 27426 |
| initialization_executable | y |
| initialization_reset | y |
| overall_phase_residual_2D | 43.489 |

Images

- CSH: First (01) merging script
- LOG: First (01) origtilt A output
- CSH: Last (12) merging script
- LOG: Last (12) origtilt A output
- LOG: Console output from merging
- APH: merge.aph
- merge.aph [H,K,Z,A,P,#,IQ,W,Bk,CTF]
- LOG: 2dx_avrgamphs output for 2D run
- TXT: Phase residual table
- APH: FOMSTATS HKL output
- TXT: FOMSTATS statistics
- APH after hklsym4 [H,K,L,FPFOM,1.0]
- PS: Resolution Circle Plot of non-tilted data
- MTZ: Merged full reciproc. space 2D data
- MTZ: Merged full reciproc. space 2D data (copy for ref)
- MRC: FINAL p4212 symmetrized 2D map

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14-2dx-2016-Merging-2D.key - 24 Aug 2016

Final Merge in 2D

MloK1-cAMP | 2dx (4.0.0)
Level: Simplified

2D

- Initialization
- Merge Selected Images

3D

- Refine PhaseOrigins Once
- Merge & Refine (Iterative)
- Final Merge**
- Quality Evaluation
- Generate Image Maps

DEPENDENT SCRIPTS

- 2dx_origtilt_merge.com
- 2dx_origtilt_aph_to_mtz.com
- 2dx_sym2spcgrp_sub.com
- 2dx_merge_makedirs
- initialize

Final Merge

Setup

Search Parameters

Common Image Processing

Lower Resolution Limit (RESMIN) 90.0

Upper Resolution Limit (RESMAX, in Angstroms) 4.5

Merging resolution limit

0-Use global resolution limit

Resolution of the merged dataset for reference 4.5

Output (Double click for logbrowser) Verboosity Level: Low

| | | | | | | | | | | | |
|----|-----|-----|-----|------|------|------|------|------|------|------|------|
| 8 | 0 | 0 | 10 | 58 | 139 | 142 | 143 | 744 | 1236 | | |
| 9 | 5.0 | 4.7 | 0.0 | 5.8 | 65.2 | 61.2 | 63.5 | 69.7 | 78.5 | 75.7 | 73.4 |
| 9 | 0 | 3 | 11 | 85 | 103 | 138 | 144 | 953 | 1437 | | |
| 10 | 4.7 | 4.5 | 0.0 | 0.0 | 82.5 | 61.0 | 69.0 | 74.4 | 76.4 | 79.8 | 76.8 |
| 10 | 0 | 0 | 10 | 69 | 98 | 124 | 138 | 735 | 1174 | | |
| | | | | 9.2 | 13.5 | 20.7 | 33.4 | 44.1 | 51.8 | 61.9 | 69.5 |
| | | | | 2023 | 4337 | 2964 | 2513 | 2385 | 2166 | 1881 | 9157 |

Overall: Phaseresidual = 43.489 Number of spots = 27426

The last column shows the average phase residual in that resolution range, which is the mean weighted squared distance of the phase values from the averaged ψ (90 deg = random)

```
==== fomstats - to calculate FOM statistics =====
FOMSTATS output:
N RESOLUTION NDAT PHASERR(0/180, 45 is random)
1 14.3 42 21.9
2 10.1 38 13.0
3 8.2 38 19.0
4 7.1 36 16.8
5 6.4 40 22.8
6 5.8 36 25.9
7 5.4 38 25.3
8 5.0 33 34.2
9 4.7 41 34.6
10 4.5 33 31.3
THE OVERALL ERROR FOR 375 REFL. WAS: 24.3 DEG

The last column shows the average distance of the averaged phase values from the symmetry-constrained target values of 0 or 180 degrees. (45 deg = random)

==== 2dx_centric2 - to correct phases to 0 or 180 for 2D run =====
==== 2dx_hklsym4 - to apply symmetry to APH file for 2D run =====
==== 2dx_plotreska - to plot the powerspectrum with resolution circles =====
==== f2mtz - to transform APH file into MTZ file for 2D run =====
==== 2dx_processor.exe - to create a MAP file from HKL =====
==== 2dx_merge normal end. =====
#####2dx_finalmerge2D finished.#####
```

Results

| Parameter | Value |
|---------------------------|--------|
| JREFL | 54832 |
| JREFL_2D | 27426 |
| initialization_executable | y |
| initialization_reset | y |
| overall_phase_residual_2D | 43.489 |

Images

- CSH: First (01) merging script
- LOG: First (01) origtilt A output
- CSH: Last (12) merging script
- LOG: Last (12) origtilt A output
- LOG: Console output from merging
- APH: merge.aph
- merge.aph [H,K,Z,A,P,#,IQ,W,Bk,CTF]
- LOG: 2dx_avgamphs output for 2D run
- TXT: Phase residual table
- APH: FOMSTATS HKL output
- TXT: FOMSTATS statistics
- APH after hklsym4 [H,K,L,F,FOM,1.0]
- PS: Resolution Circle Plot of non-tilted data
- MTZ: Merged full reciproc. space 2D data
- MTZ: Merged full reciproc. space 2D data (copy for ref)
- MRC: FINAL p4212 symmetrized 2D map

Quality Evaluation in 2D

MloK1-cAMP | 2dx (4.0.0)
Level: Simplified

2D

- Initialization
- Merge Selected Images

3D

- Refine PhaseOrigins Once
- Merge & Refine (Iterative)
- Final Merge
- Quality Evaluation**
- Generate Image Maps

DEPENDENT SCRIPTS

- 2dx_sym2spcgrp_sub.com
- 2dx_merge_makedirs
- initialize
- get_max_defocus.py

Quality Evaluation

Setup

Search Parameters

Lower Resolution Limit (RESMIN) 90.0

Upper Resolution Limit (RESMAX, in Angstroms) 4.5

Resolution of the merged dataset for reference 4.5

ALAT (Z-dimension of unit cell to reconstruct) 200

Output (Double click for logbrowser) Verboosity Level: Low

```
#####2dx_QualityEvaluation2D#####
==== Sourcing sym2spcgrp_sub.com
==== get_max_defocus.py - to extract defocus and TANGL maxima. =====

2D Reconstruction Parameters
-----
Crystal plane group symmetry: p4212
Crystal unit cell parameters: a=135.0A, b=135.0A
                             gamma=90.0deg
-----
Number of images: 35
Range of defocus [micrometers]: -0.750 ... 1.937
Number of observed reflections: 27426
IQ range used: 1 ... 8
-----
Resolution cut-off [Angstroms]: 4.5
-----
Overall phase residual [deg]: 43.5
-----
19 images in MloK1-II
5 images in MloK1-K2-2014-05-26-MloK1 cAMP
2 images in MloK1-K2-2014-07-29-MloK1 cAMP
2 images in MloK1-K2-2014-09-08-MloK1 cAMP
5 images in MloK1-K2-2014-09-16-MloK1 cAMP
1 images in MloK1-K2-2014-11-17-MloK1 cAMP
1 images in automatic

-----
PHASE RESIDUALS IN RESOLUTION RANGES
-----
# DWIN DMAX IQ= 1 2 3 4 5 6 7 8 all IQs
1 1000.0 14.2 9.0 13.6 20.0 29.3 36.8 43.3 52.3 63.7 23.5
1 1667 2727 1402 647 405 304 243 829 8224
2 14.2 10.1 10.1 13.3 18.3 25.5 32.7 38.2 46.4 53.7 26.7
2 332 1257 806 524 396 295 200 749 4559
3 10.1 8.2 8.5 14.0 21.4 29.3 39.3 43.1 54.9 59.7 40.1
3 24 313 532 531 388 318 248 1033 3387
4 8.2 7.1 0.0 11.4 21.7 30.3 45.1 48.8 57.5 66.6 53.4
4 0 36 108 263 298 268 195 1053 2221
5 7.1 6.4 0.0 22.1 34.8 41.5 43.4 56.9 60.1 72.0 62.3
```

Results

| Parameter | Value |
|---------------------------|-------|
| initialization_executable | y |
| initialization_reset | y |

Images

- TXT: Summary_Statistics_2D
- TXT: Phase residual table
- PS: Resolution Circle Plot of non-tilted data