

# ***Phenix tools for crystallography and cryo-EM***

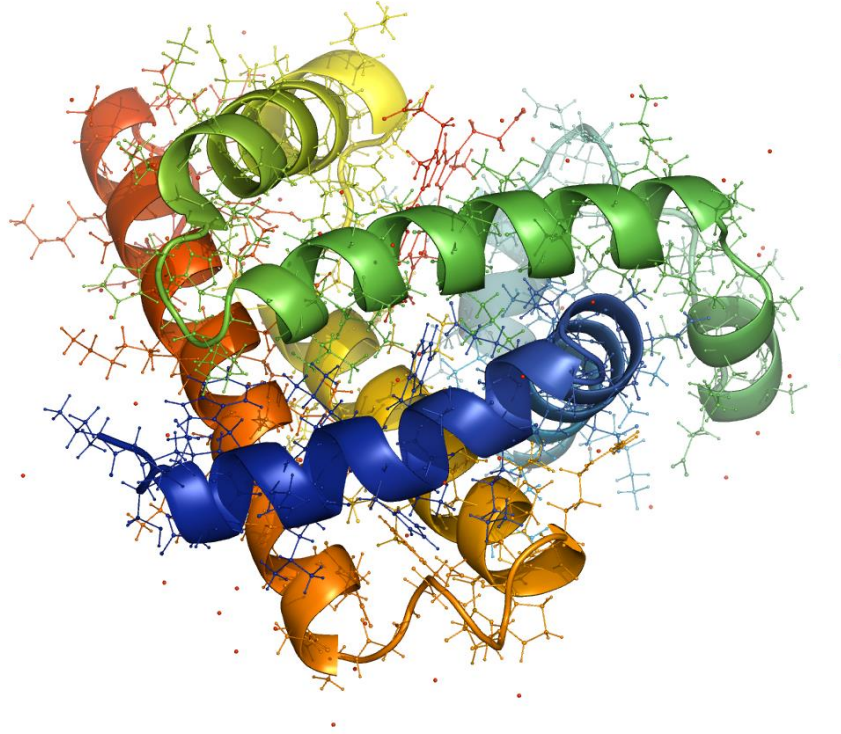
**Pavel Afonine**

**Phenix team**

**Lawrence Berkeley National Lab, California, USA**

**May, 2025  
MCCS, Madrid**

# A structure

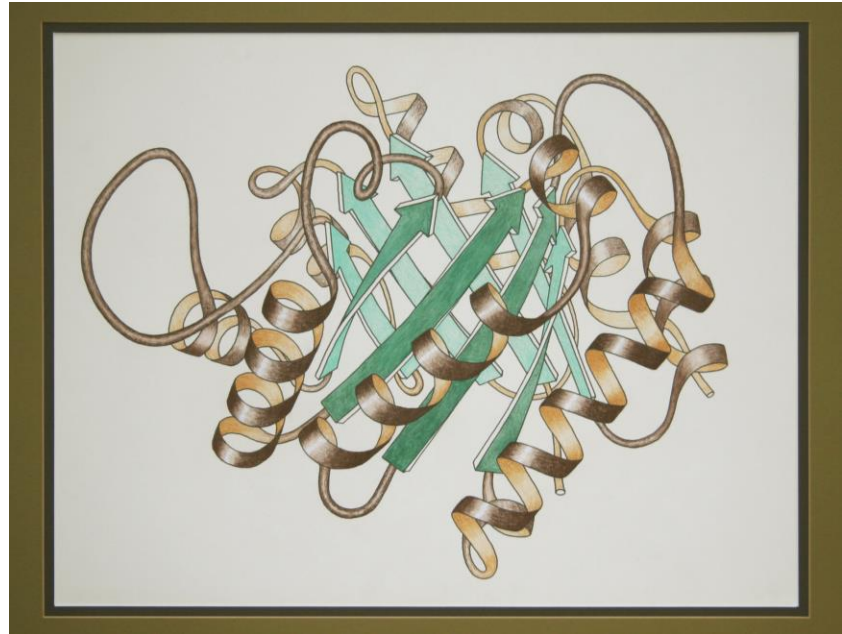


**Q:** Who invented this way of drawing a structure (ribbon diagram)?

# Ribbon diagram



Ribbon schematic of triose P  
isomerase monomer (PDB: 1TIM)



Hand-drawn by **J. Richardson**, 1981

# The Phenix Project

## Lawrence Berkeley Laboratory

Paul Adams, Pavel Afonine,  
Dorothee Liebschner, Nigel  
Moriarty, Billy Poon,  
Christopher Schlicksup,  
Oleg Sobolev



## University of Cambridge

Randy Read, Airlie McCoy,  
Rob Oeffner



## Los Alamos National Laboratory New Mexico Consortium

Tom Terwilliger, Li-Wei Hung



## UTHealth

Matt Baker



## Duke University

Jane & David Richardson,  
Christopher Williams,  
Vincent Chen



An NIH/NIGMS funded  
Program Project

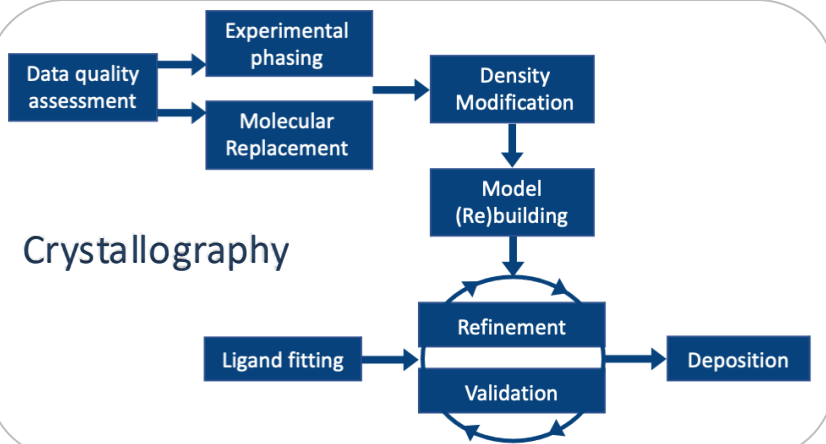
Liebschner D, *et al.*, Macromolecular structure determination using  
X-rays, neutrons and electrons: recent developments in *Phenix*.  
*Acta Cryst.* 2019 **D75**:861–877



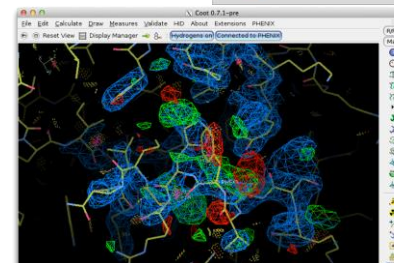
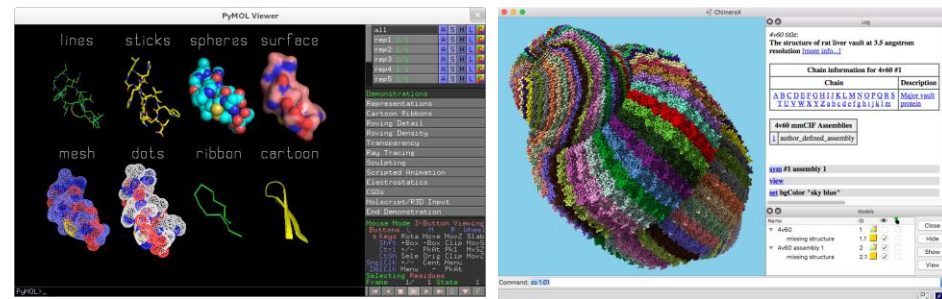
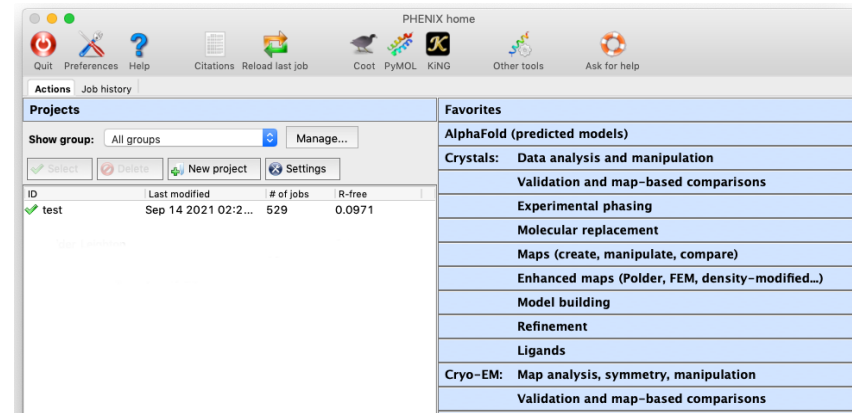
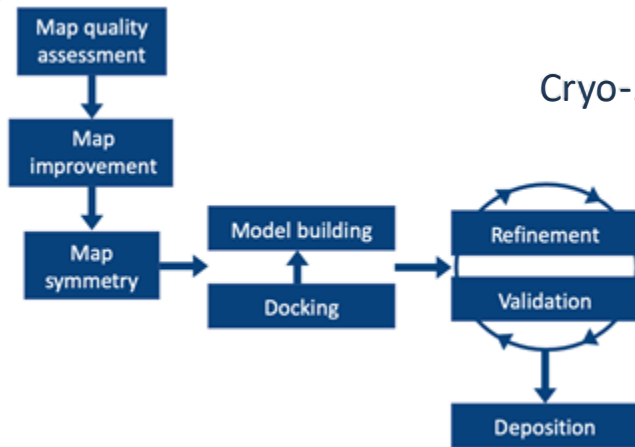
# Phenix now

- Complete system for crystallography and cryo-EM structure solution
- Available at scripting, command line and GUI levels
- Integrated with popular molecular graphics programs: Coot, PyMol and ChimeraX
- Integrated with structure prediction (AlphaFold)

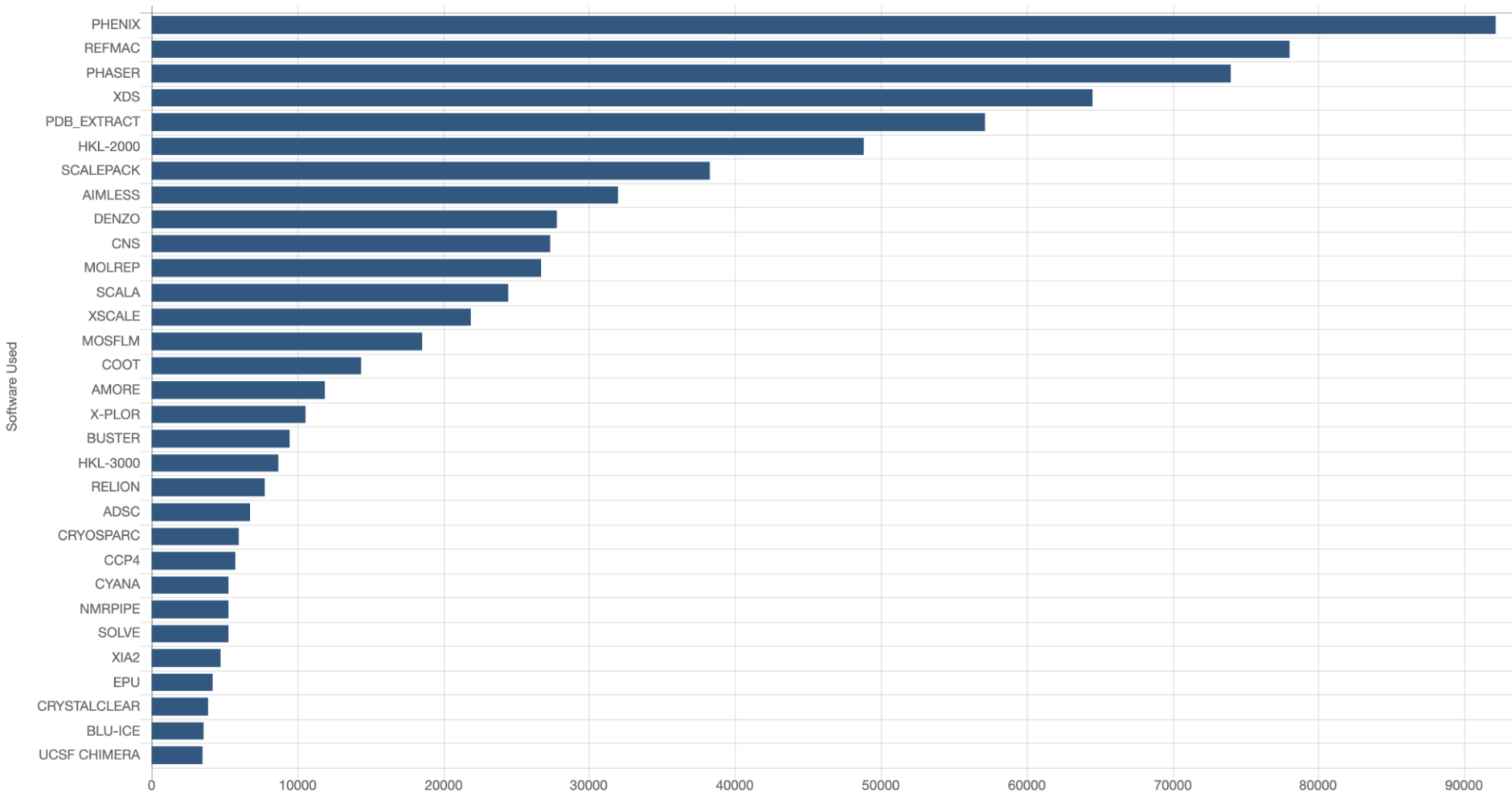
## Crystallography



## Cryo-EM



# Phenix PDB statistics



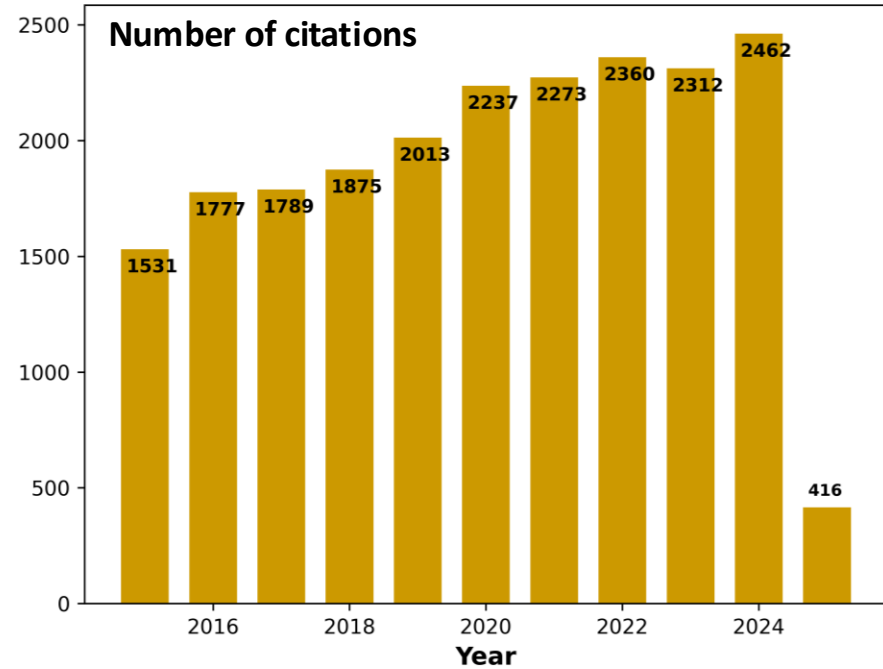
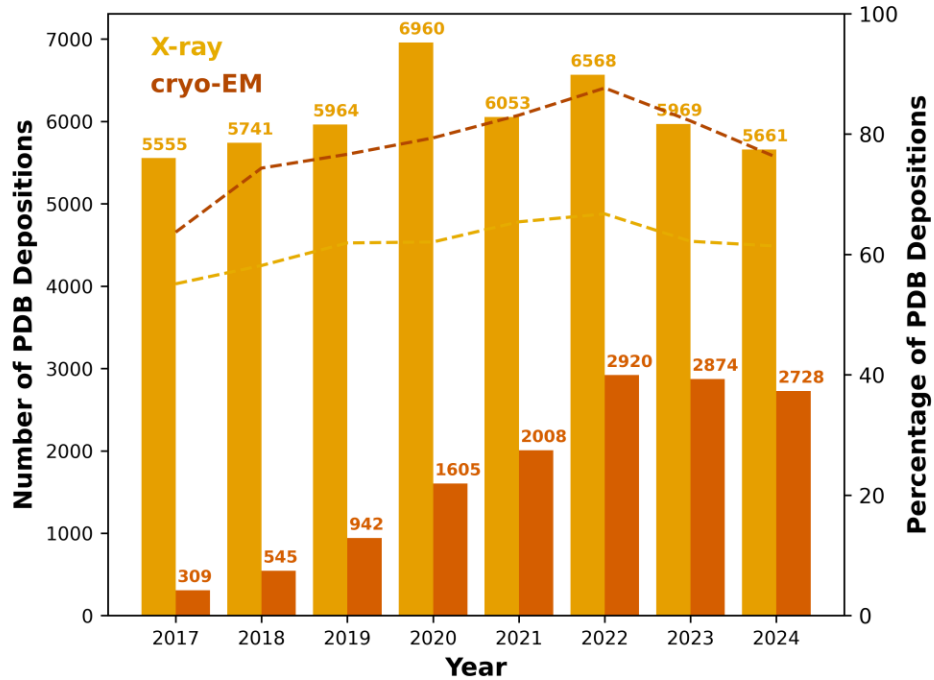
# Phenix metrics

220 publications | 18447 citations | 82101 models in PDB | 35000 users | 33 countries

About 70 industry users



# Phenix metrics



# Keep Up With Trends

- **YouTube** channel: 63 videos

The screenshot shows the YouTube channel page for "Phenix Tutorials". The channel has 560 subscribers and is categorized under "Science & Technology". The page displays a grid of 12 video thumbnails, each with a title, description, view count, and upload date. The videos are arranged in two rows of six. The first row includes videos on real-space refinement, secondary structure restraints, multiple refinement strategies, SAD experiments, map-to-model interpretation, and scaling/merging data. The second row includes automated map sharpening, ligandfit, Wilson plots, twinning, translational NCS, and checking data quality with Xtriage. The channel banner features the text "Phenix Tutorials" in a large, blue, 3D font.

Phenix Tutorials  
560 subscribers

SUBSCRIBE 560

HOME VIDEOS PLAYLISTS CHANNELS ABOUT

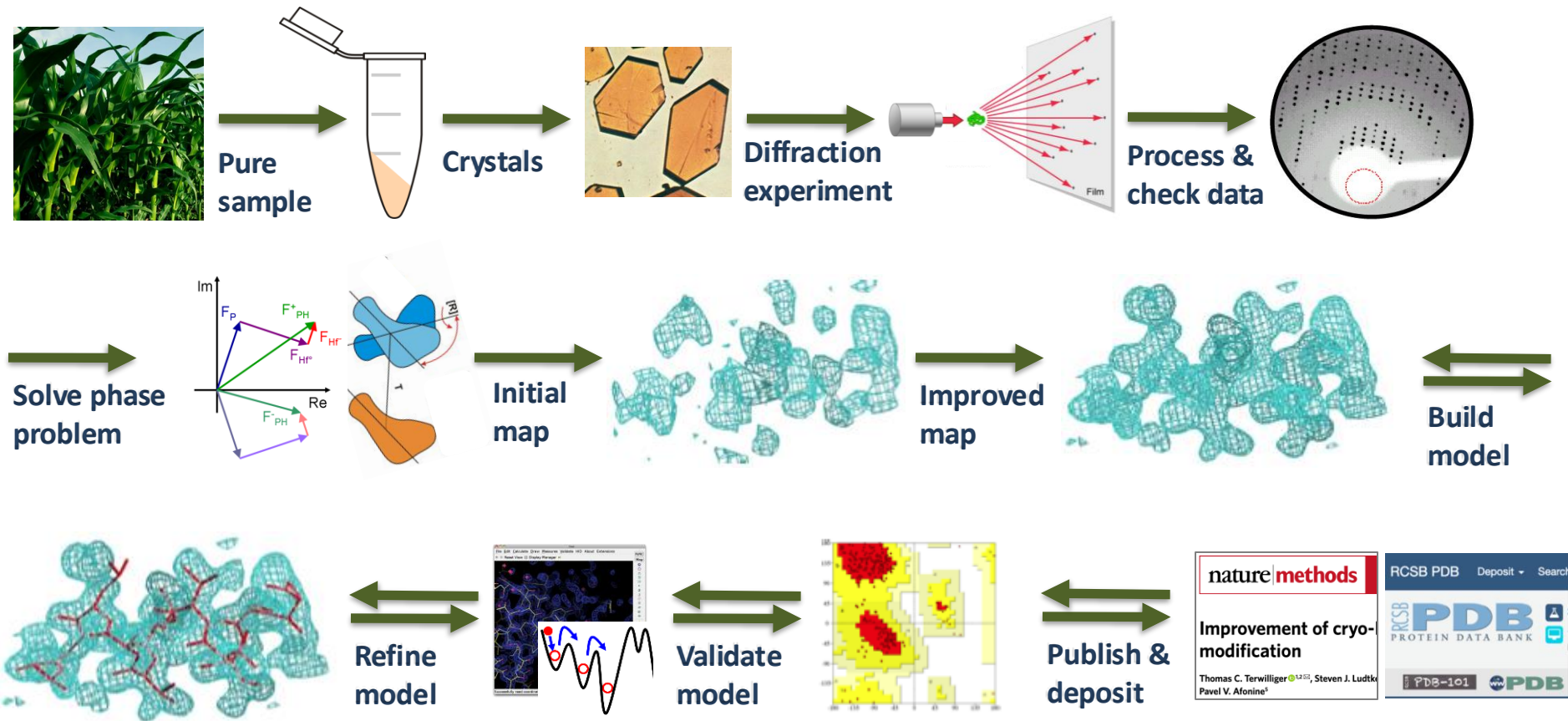
Uploads PLAY ALL SORT BY

Video Title	Duration	Views	Upload Date
real_space_refine Tutorial	5:27	763 views	7 months ago
Secondary Structure Restraints Tutorial	6:23	399 views	8 months ago
Multiple refinement strategies Tutorial	5:46	281 views	8 months ago
Planning a SAD experiment Tutorial	6:00	483 views	1 year ago
Map-to-model Tutorial	5:33	1.3K views	1 year ago
Scale-and-merge Tutorial	7:31	387 views	1 year ago
Automated map sharpening Tutorial	6:05		
Ligandfit Tutorial	5:48		
Wilson plots and space group identification phenix.xtriage	8:23		
Twinning phenix.xtriage	6:08		
Translational NCS phenix.xtriage	4:55		
Checking data quality with Xtriage	6:49		

# **Crystal structure solution**

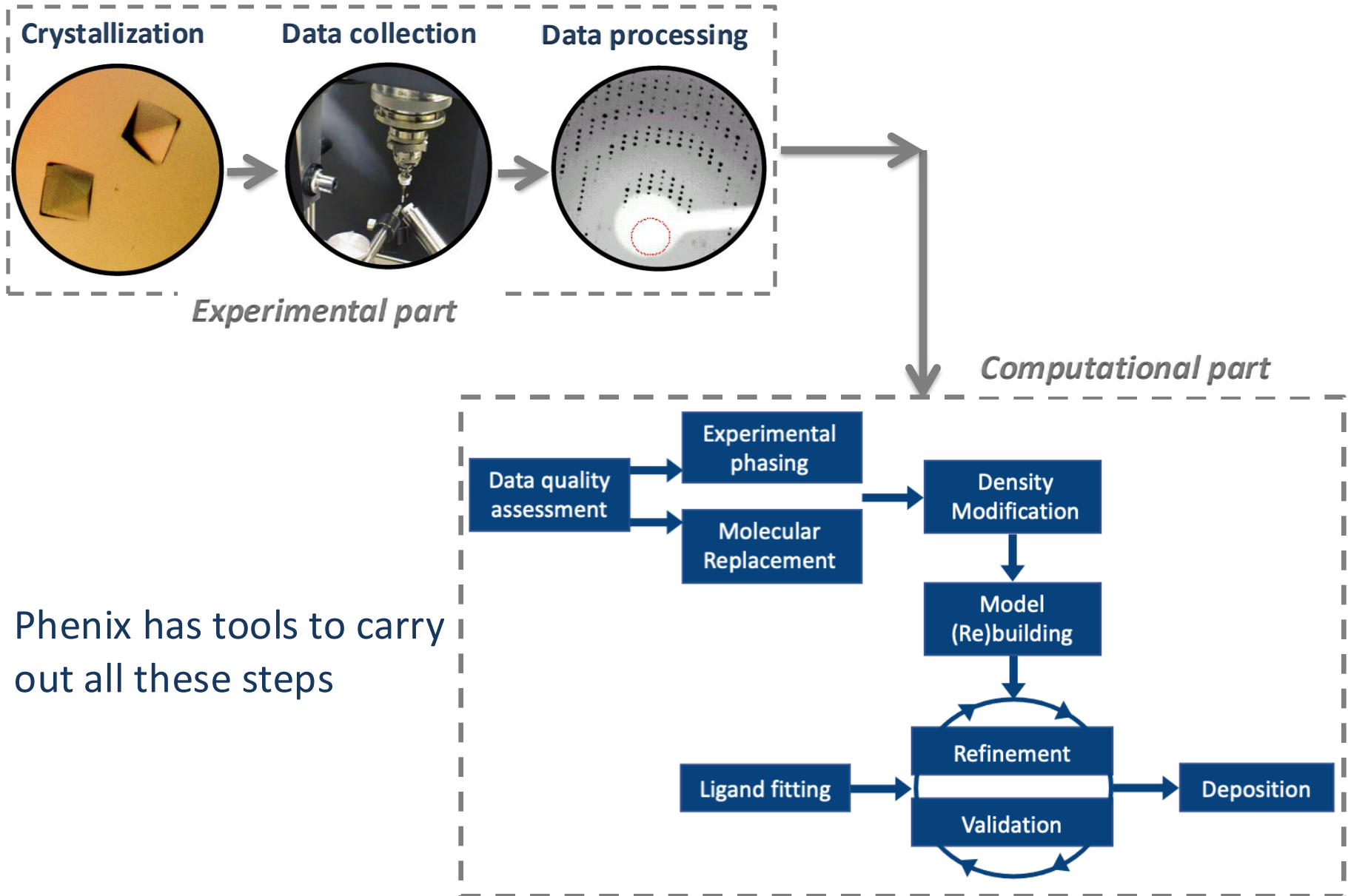


# Steps to solve structure by crystallography



- Process is not as 'linear' as shown
- Each step has numerous sub-steps
- Each step can take from days to years
- Each step can fail

# Crystal structure determination workflow



# Phenix tools for crystallography: *GUI and command line*

The screenshot displays the PHENIX home graphical user interface. At the top, there is a menu bar with icons for Quit, Preferences, Help, Citations, ChimeraX, Coot, PyMOL, KING, Other tools, and Ask for help. Below this is a toolbar with buttons for Actions, Job history, and a search bar. The main window is divided into two panes. The left pane, titled 'Projects', shows a list of projects with columns for ID, Last modified, # of jobs, and R-free. The right pane, titled 'Crystals', shows a list of tools for data analysis and manipulation, including Validation and map-based comparisons, Experimental phasing, Molecular replacement, Maps (create, manipulate, compare), Enhanced maps (Polder, FEM, density-modified...), Model building, Refinement, and Ligands. The 'Crystals' pane is highlighted with a red border. Below the project list, there is a 'Current directory' field showing the path /Users/pafonine/63\_goska/polder and a 'Browse...' button. The status bar at the bottom indicates the PHENIX version (dev-svn-000) and the current project (63G).

**PHENIX home**

Quit Preferences Help Citations ChimeraX Coot PyMOL KING Other tools Ask for help

**Actions** Job history

**Projects**

Show group: All groups Manage...

Select Delete New project Settings

ID	Last modified	# of jobs	R-free
63G	Jun 23 2022 05:26...	1	---
JAKE	Jun 01 2022 05:44...	1	---
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FEM	May 26 2022 03:02...	5	---
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48test	Jan 10 2022 01:30 ...	6	0.1540
1vqw	Jan 06 2022 05:59 ...	2	0.2323
real-space-refin...	Jan 06 2022 02:12 ...	4	---
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34_maria	Oct 13 2021 01:21...	0	---
30debug	Oct 05 2021 02:02...	2	0.1522

**Crystals:** Data analysis and manipulation

Validation and map-based comparisons

Experimental phasing

Molecular replacement

Maps (create, manipulate, compare)

Enhanced maps (Polder, FEM, density-modified...)

Model building

Refinement

Ligands

**Cryo-EM:** Map analysis, symmetry, manipulation

Validation and map-based comparisons

Map improvement

Docking, model building and rebuilding

Refinement

**Models:** Superpose, search, compare, analyze symmetry

Modification, minimization and dynamics

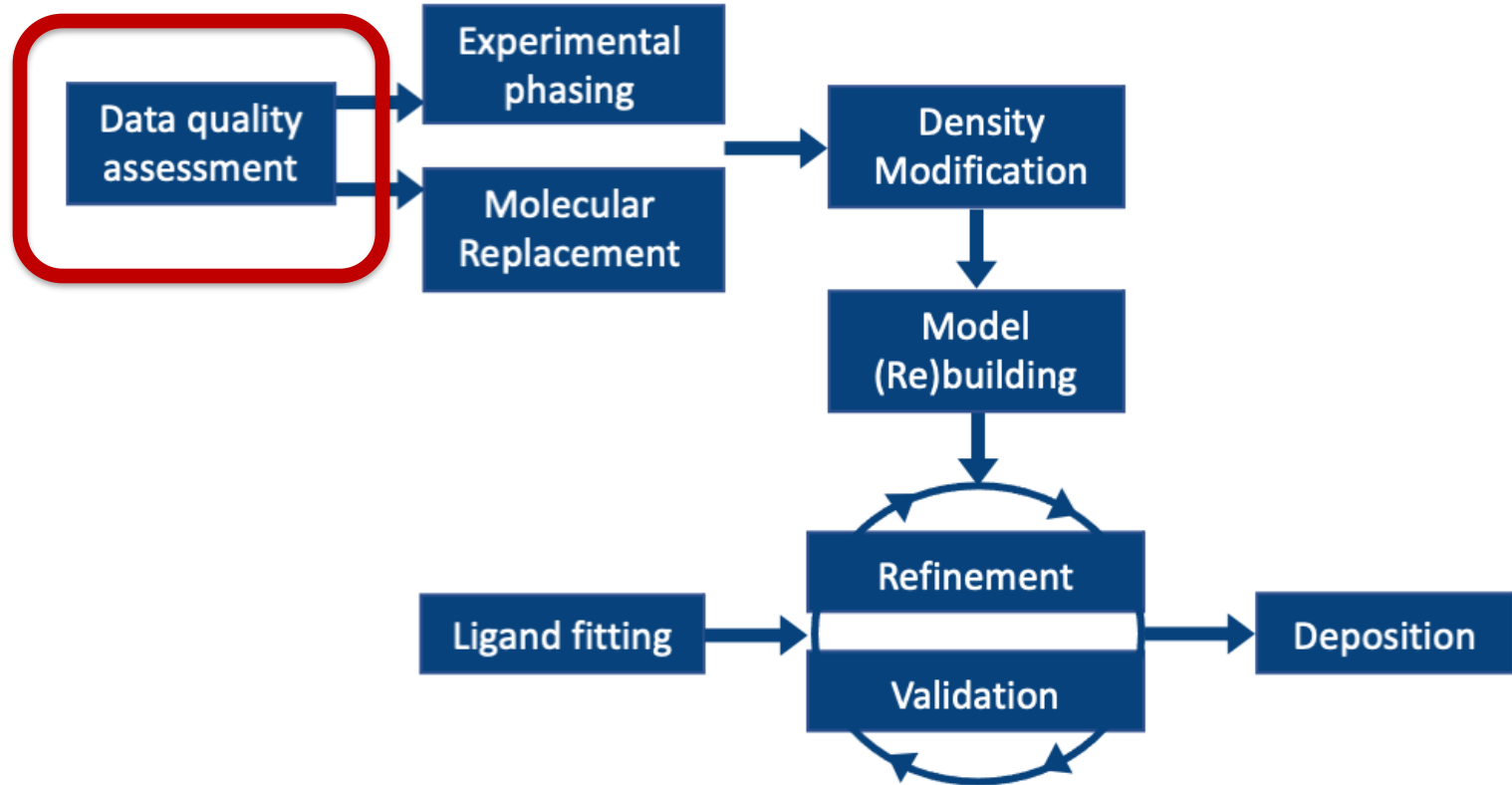
**PDB Deposition**

**Program search**

Current directory: /Users/pafonine/63\_goska/polder Browse...

PHENIX version dev-svn-000 Project: 63G

# Experimental data quality

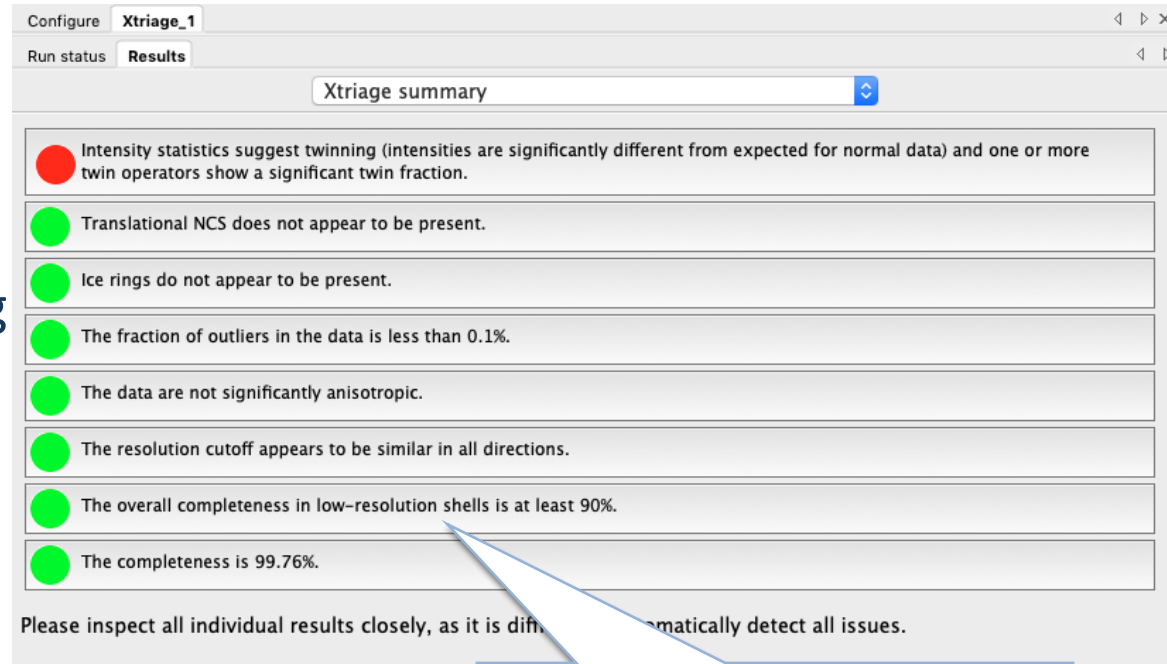


## Macromolecular crystals can have pathologies:

- Twinning: two or more crystals are intergrown (orientations are related by a twin operation)
- tNCS: more than one copy of a molecule is in a similar orientation in the asymmetric unit (ASU)

# Experimental data quality: Xtrriage

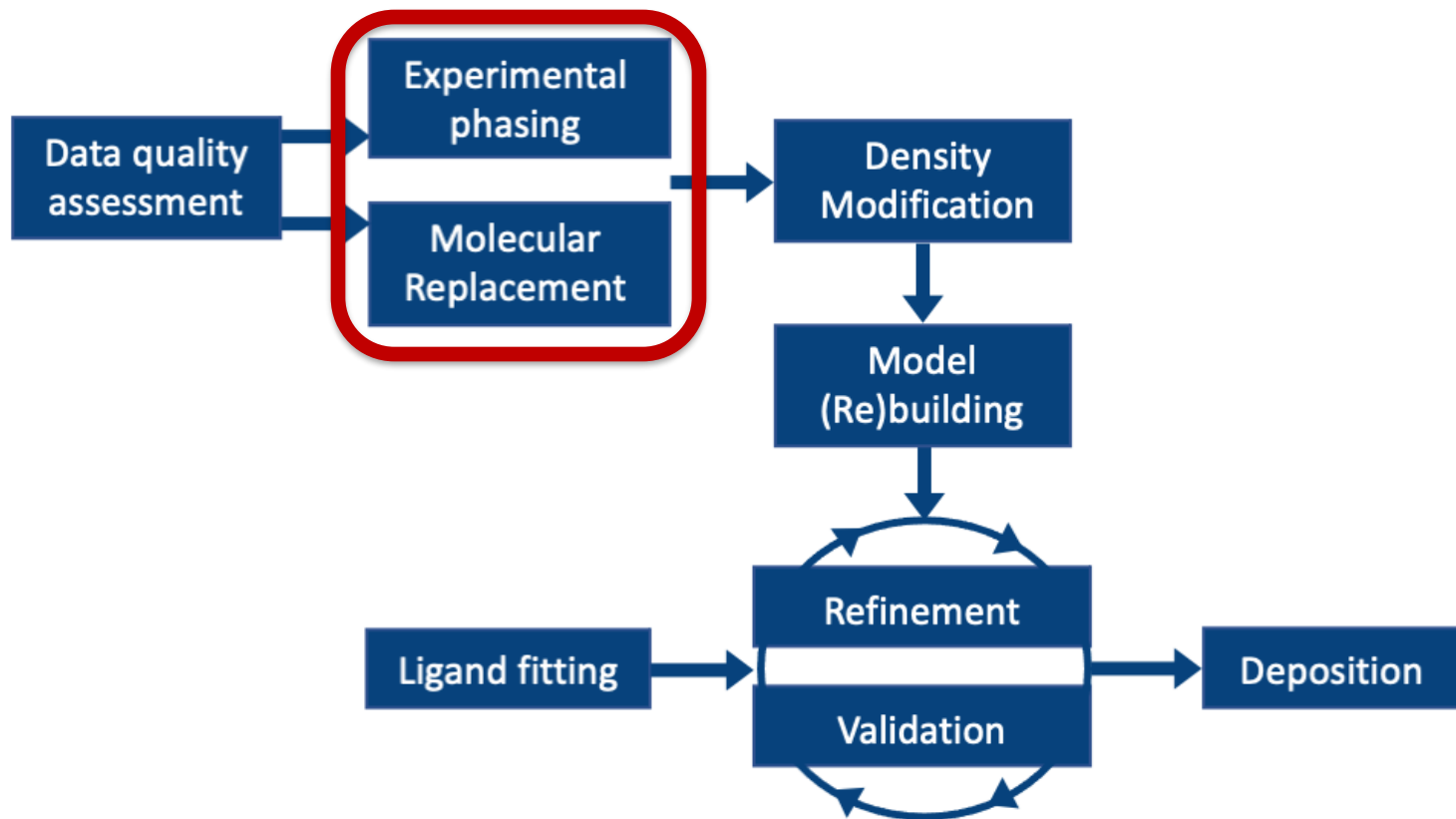
- Data anomalies can prevent structure solution
- Important to check the data before doing anything else
- Xtrriage is the tool for this!



Click to explore the results and investigate problems

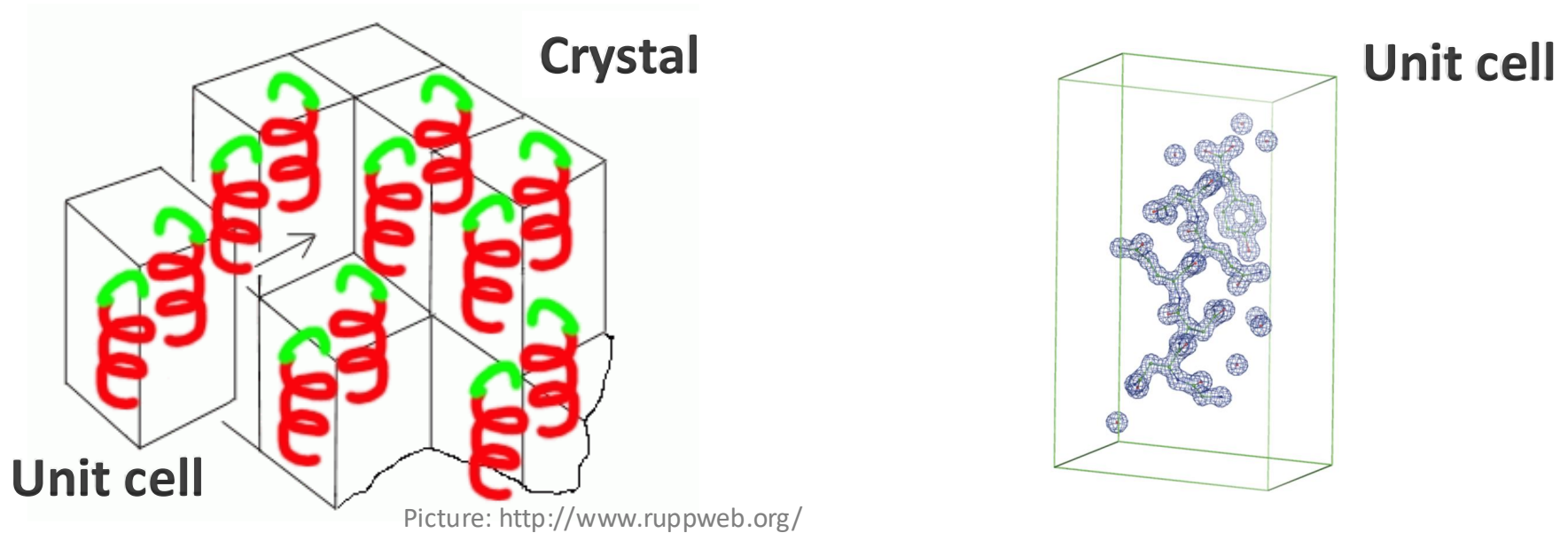
- Xtrriage comprehensively checks the data (Wilson plot, completeness, symmetry, tNCS, twinning, anomalous completeness and signal, and many more)

# Solving phase problem





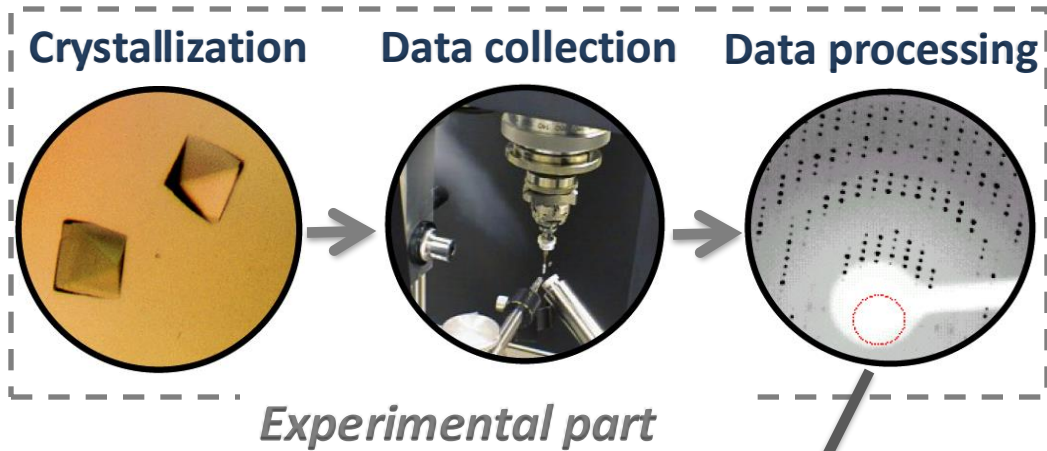
# Phase problem and finite completeness of the data



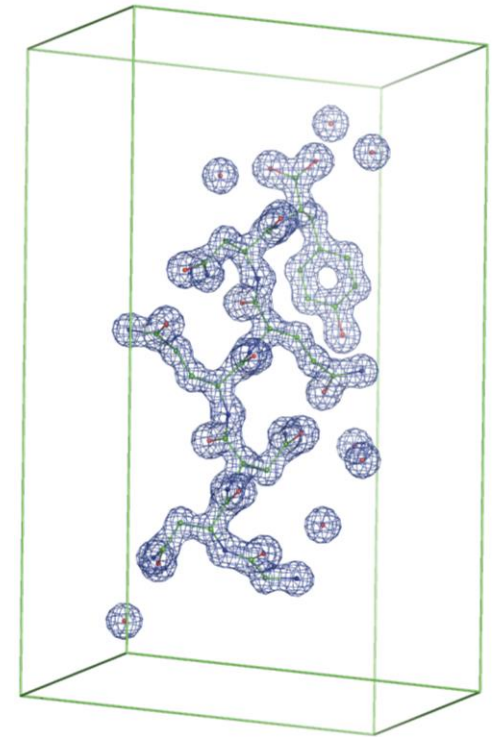
- Crystal is a periodic object, so we can:
  - do diffraction experiment
  - represent electron density as 3D infinite Fourier series

$$\rho(\mathbf{r}) = \frac{1}{V_{cell}} \sum_{h=-\infty}^{+\infty} \sum_{k=-\infty}^{+\infty} \sum_{l=-\infty}^{+\infty} F(\mathbf{s}) \exp(-2\pi i \mathbf{s} \mathbf{r})$$

# Phase problem and finite completeness of the data



Goal: obtain distribution of electron density = Structure



This is what we measure

$$\rho(\mathbf{r}) = \frac{1}{V_{cell}} \sum_{hmin}^{hmax} \sum_{kmin}^{kmax} \sum_{lmin}^{lmax} F(\mathbf{s}) \exp(-2\pi i \mathbf{s} \mathbf{r})$$

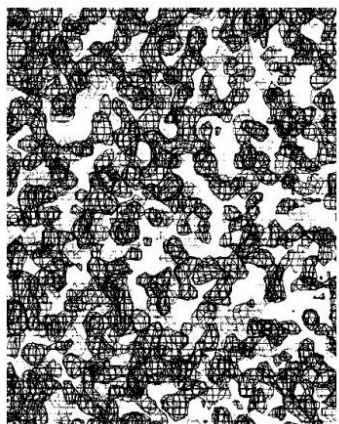
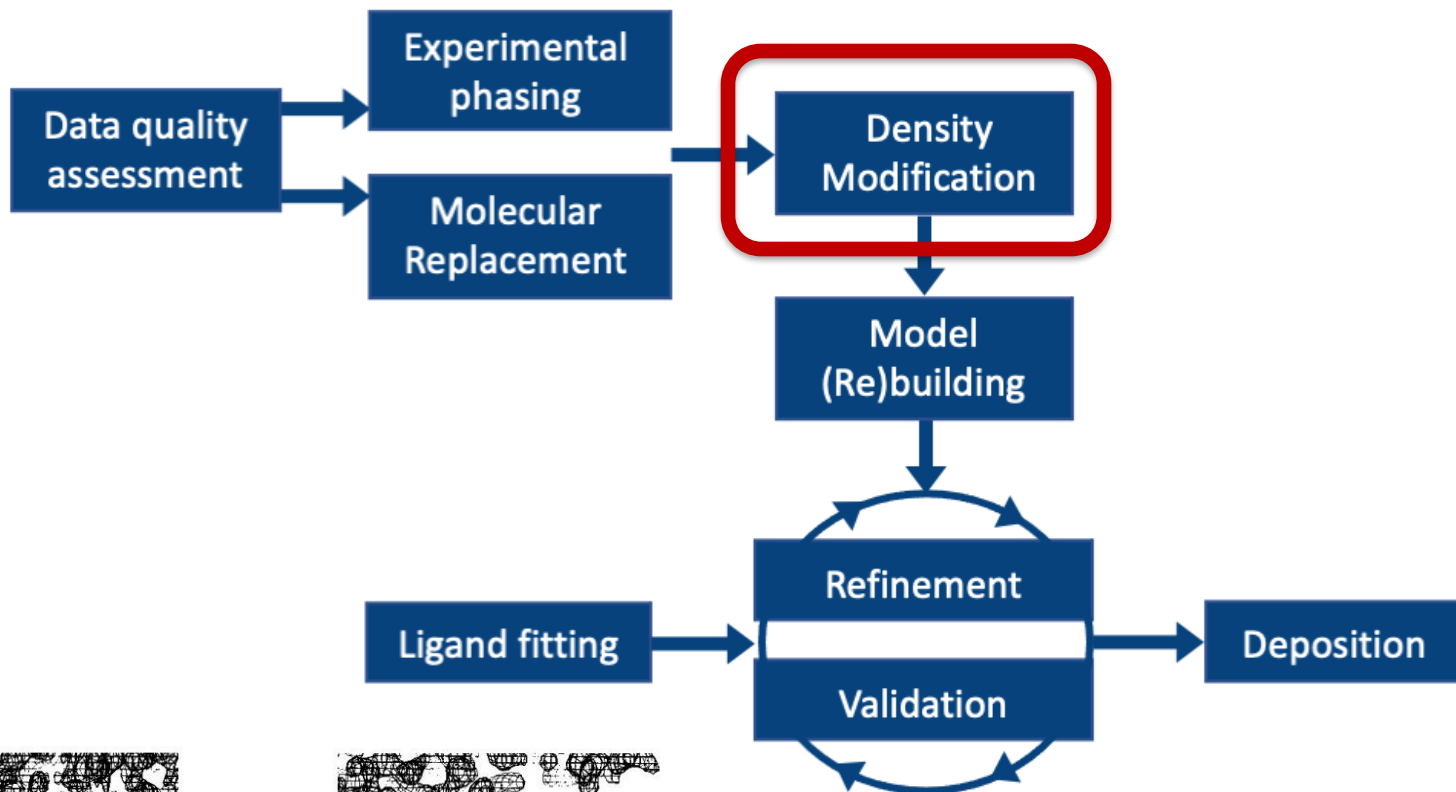
**This is lost: phase problem**

Experimental phasing techniques, direct methods and molecular replacement are the tools to solve the phase problem (recover missing phases)

# Recovering missing phases (solving phase problem)

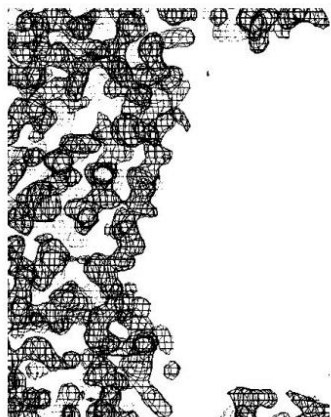
- **Experimental phasing** – exploit properties of special atoms: anomalous scattering, large number of electrons
- **Molecular replacement**: a previously known structure can provide initial phase estimates for the new structure
- **Direct methods**: phase relationships can be formulated by assuming the positivity and atomicity of the electron density

# Density modification (DM)



Poor map

DM  
→

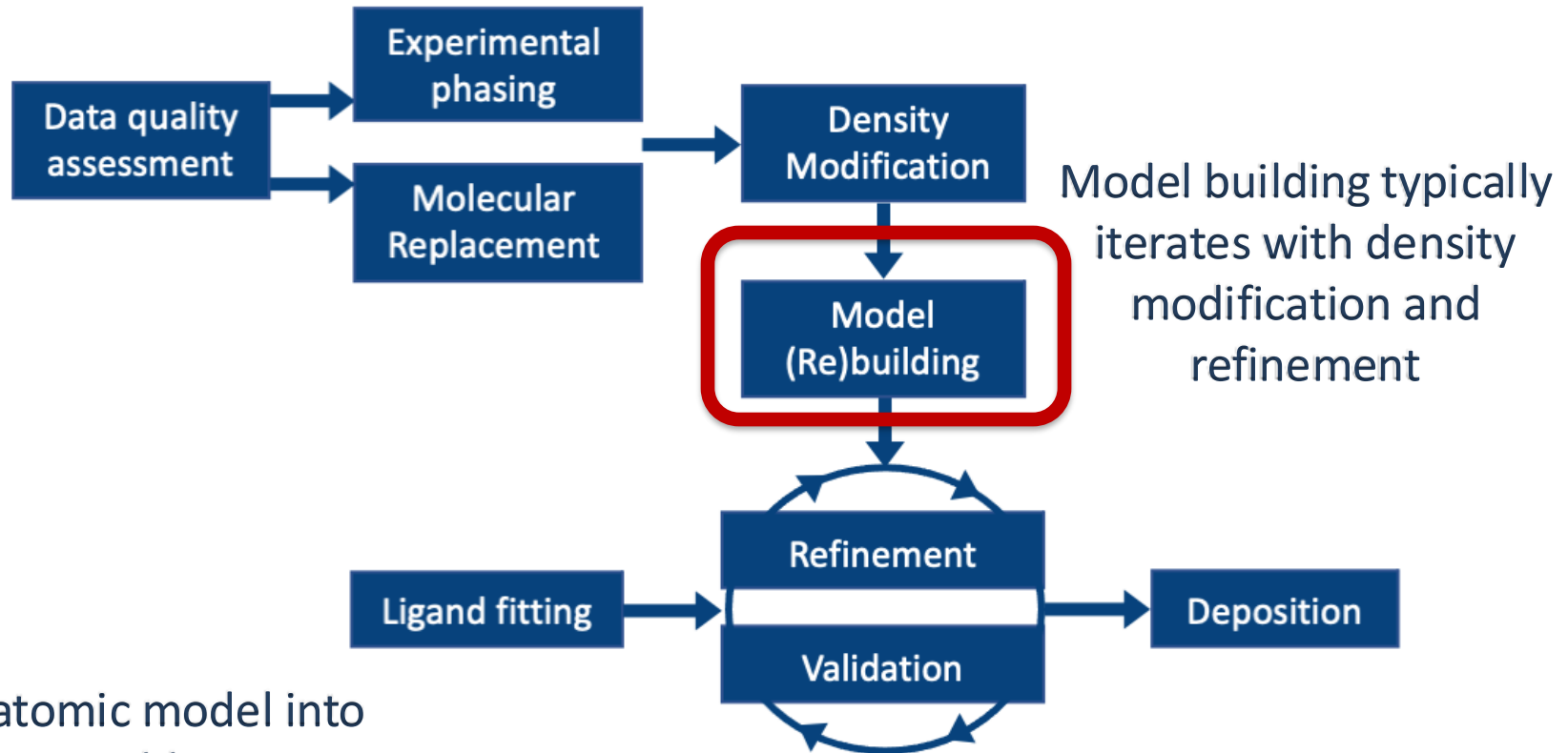


Improved map

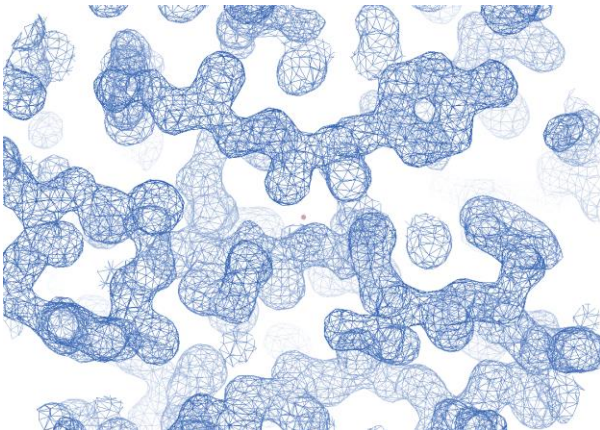
# Density modification

- Class of methods to improve initial density map by applying the information we know about maps, crystals and macromolecular structures:
  - Solvent density distribution (solvent flattening and flipping)
  - Macromolecule density distribution (Histogram Matching)
  - Atomicity and positivity of the map (Maximum Entropy Methods)
  - Similarity between molecules (Symmetry Averaging)
  - Phase and amplitude improvement and extension methods
  - Phase combination
  - Reducing bias (gamma-correction)

# Model building and re-building

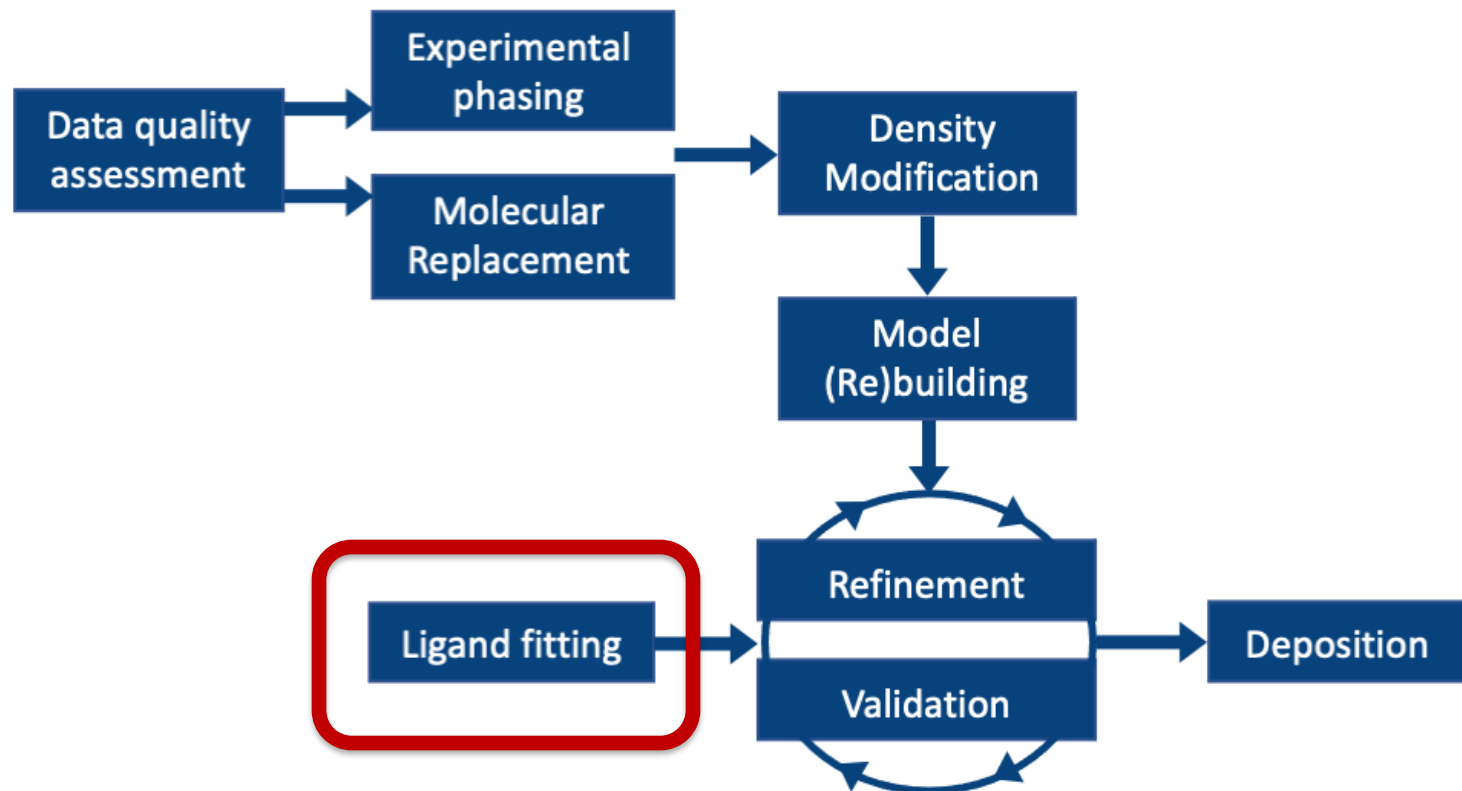


Build atomic model into interpretable map

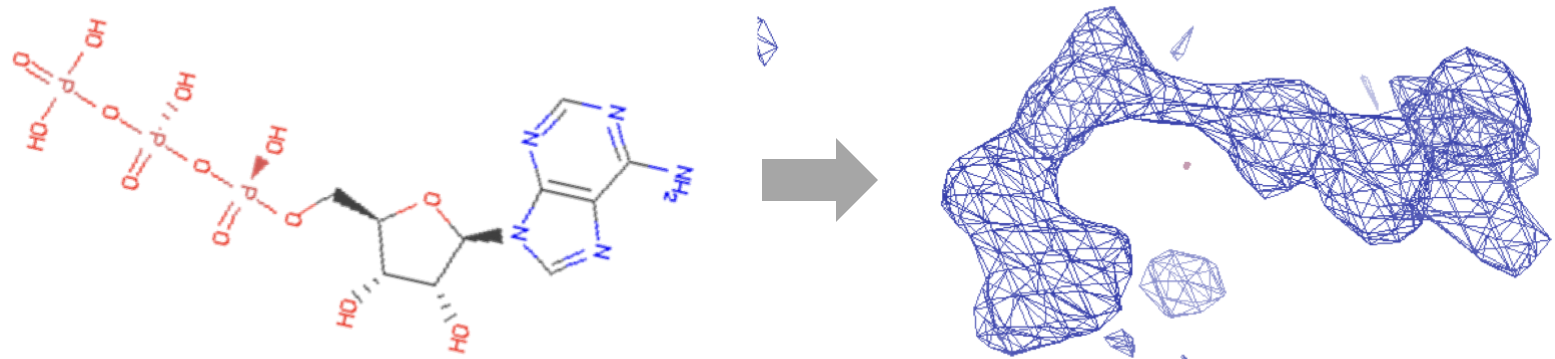
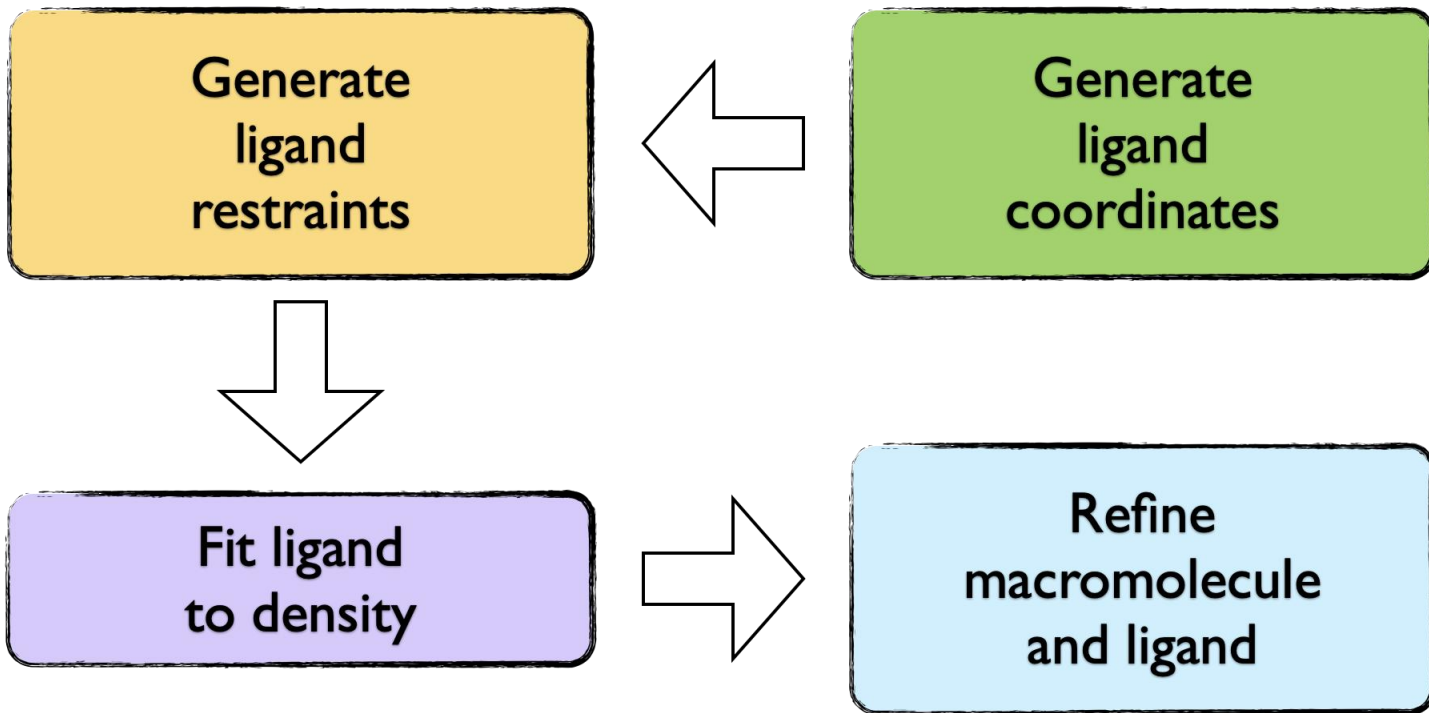




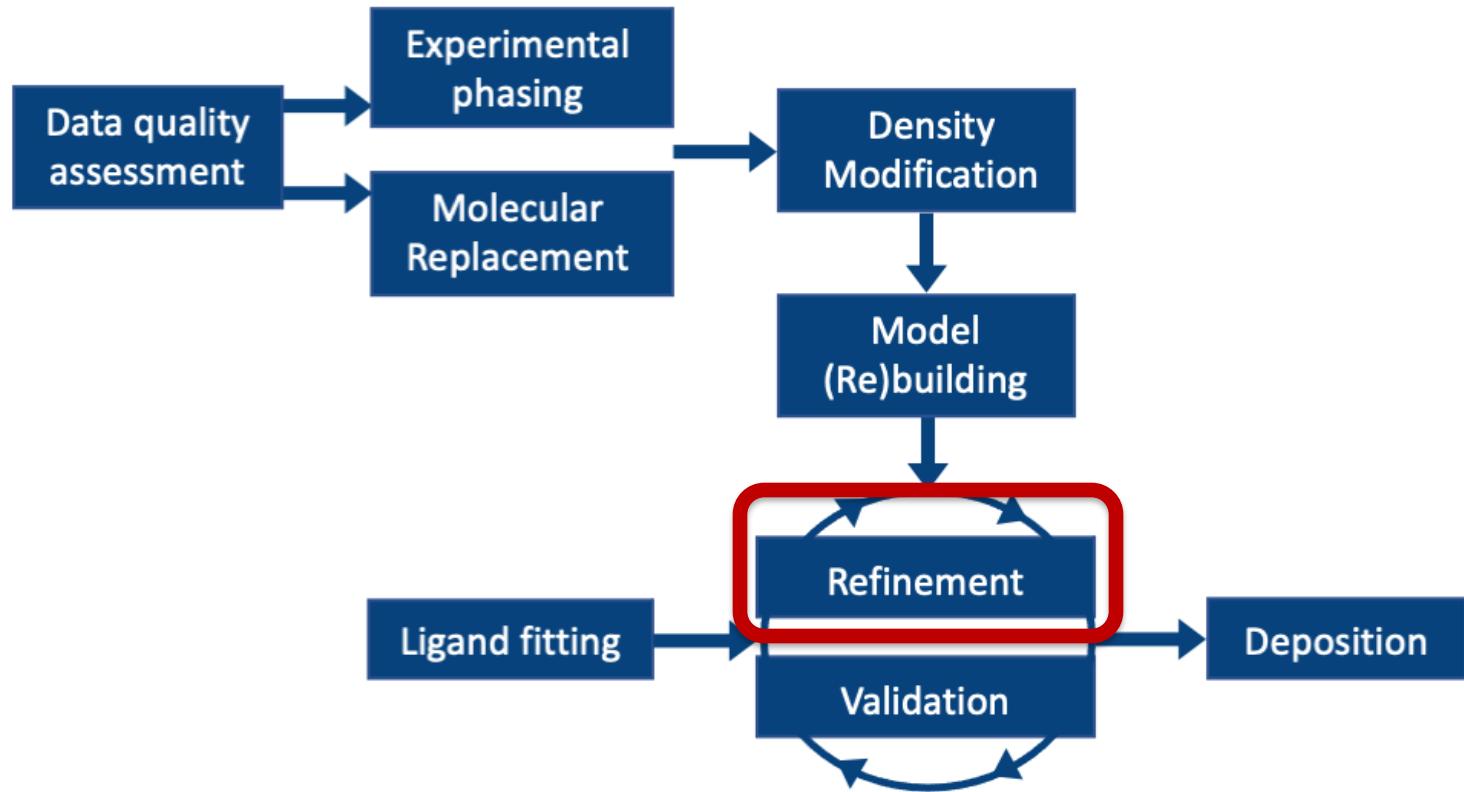
# Ligands



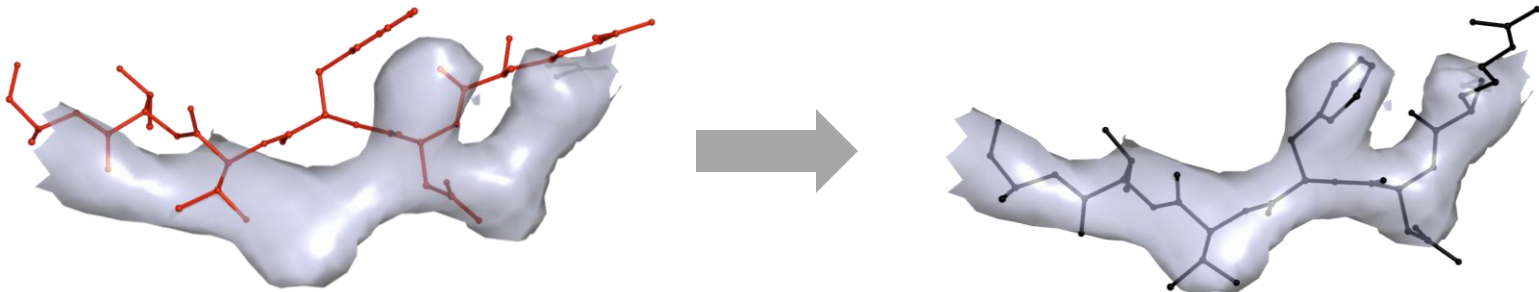
# Ligands: fitting and refinement



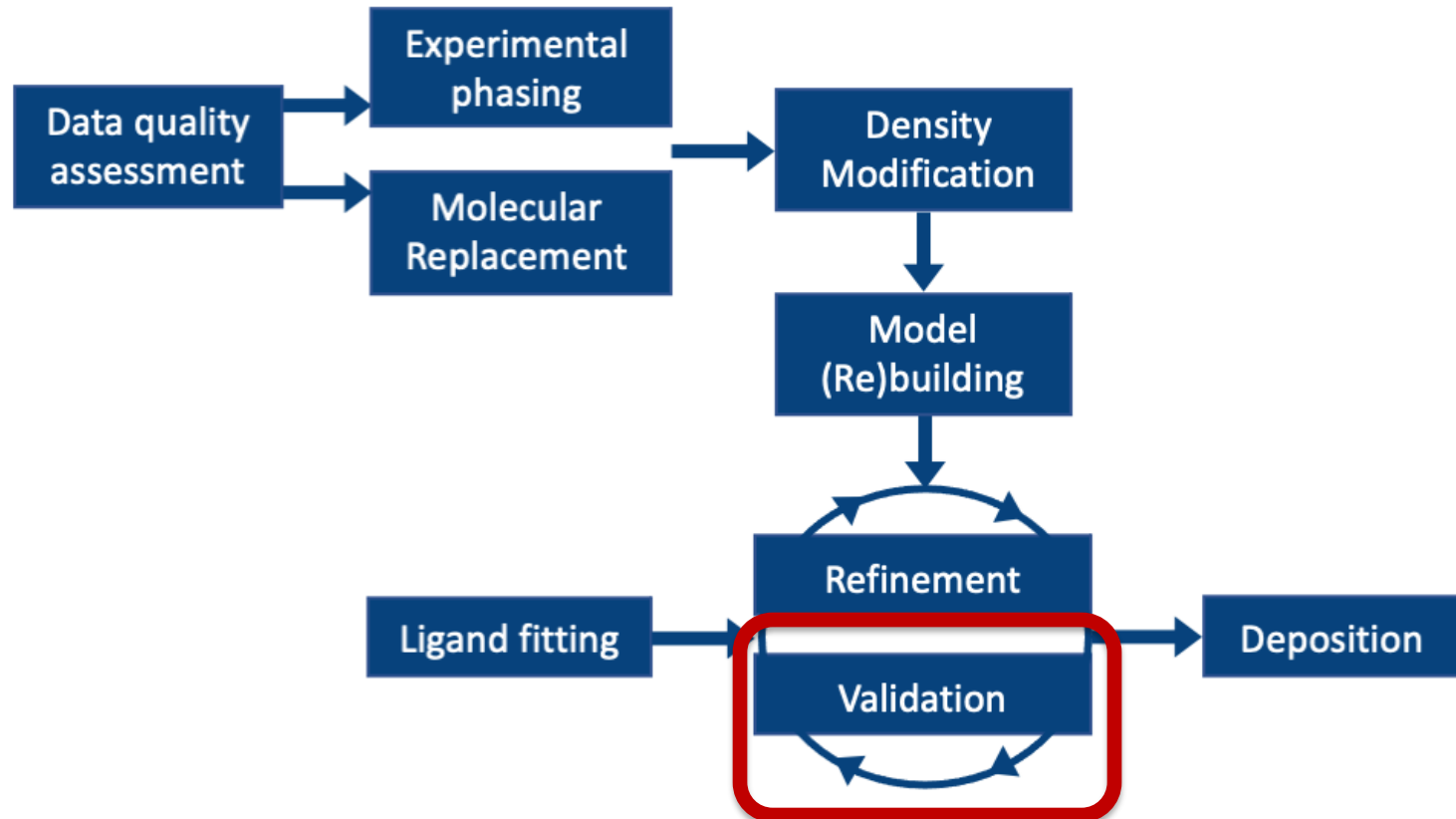
# Model refinement



Model refinement: a cyclic optimization process of making an initial model a better model

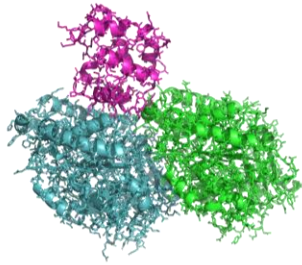


# Validation

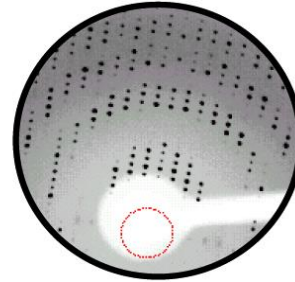


# Validation

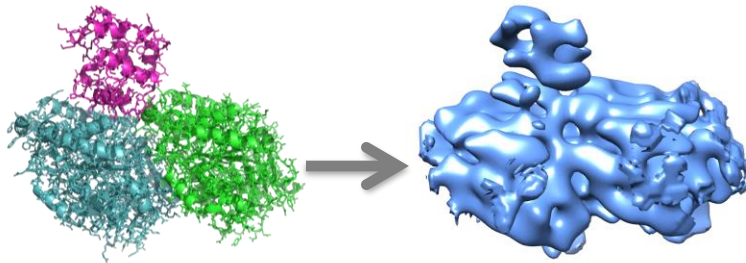
**Model**



**Data**

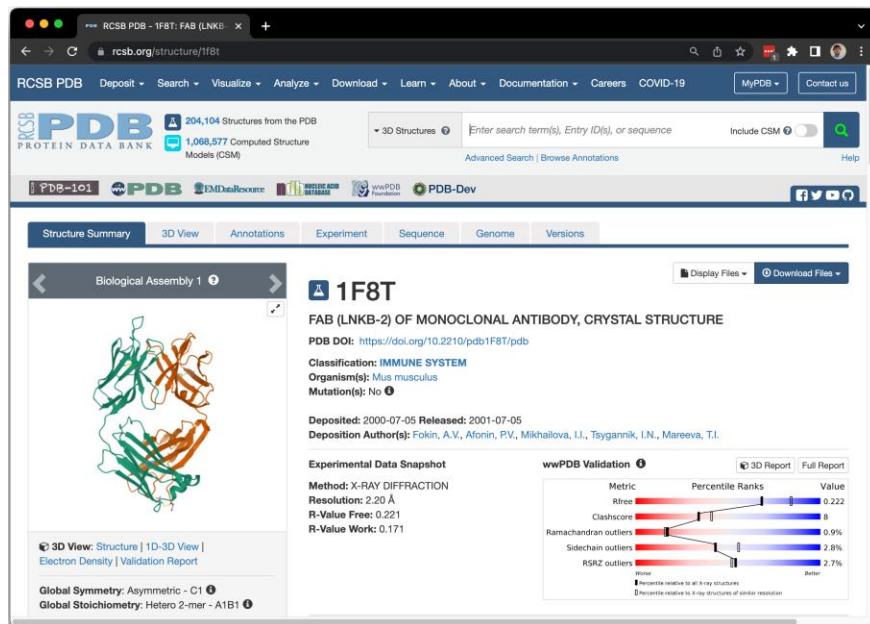
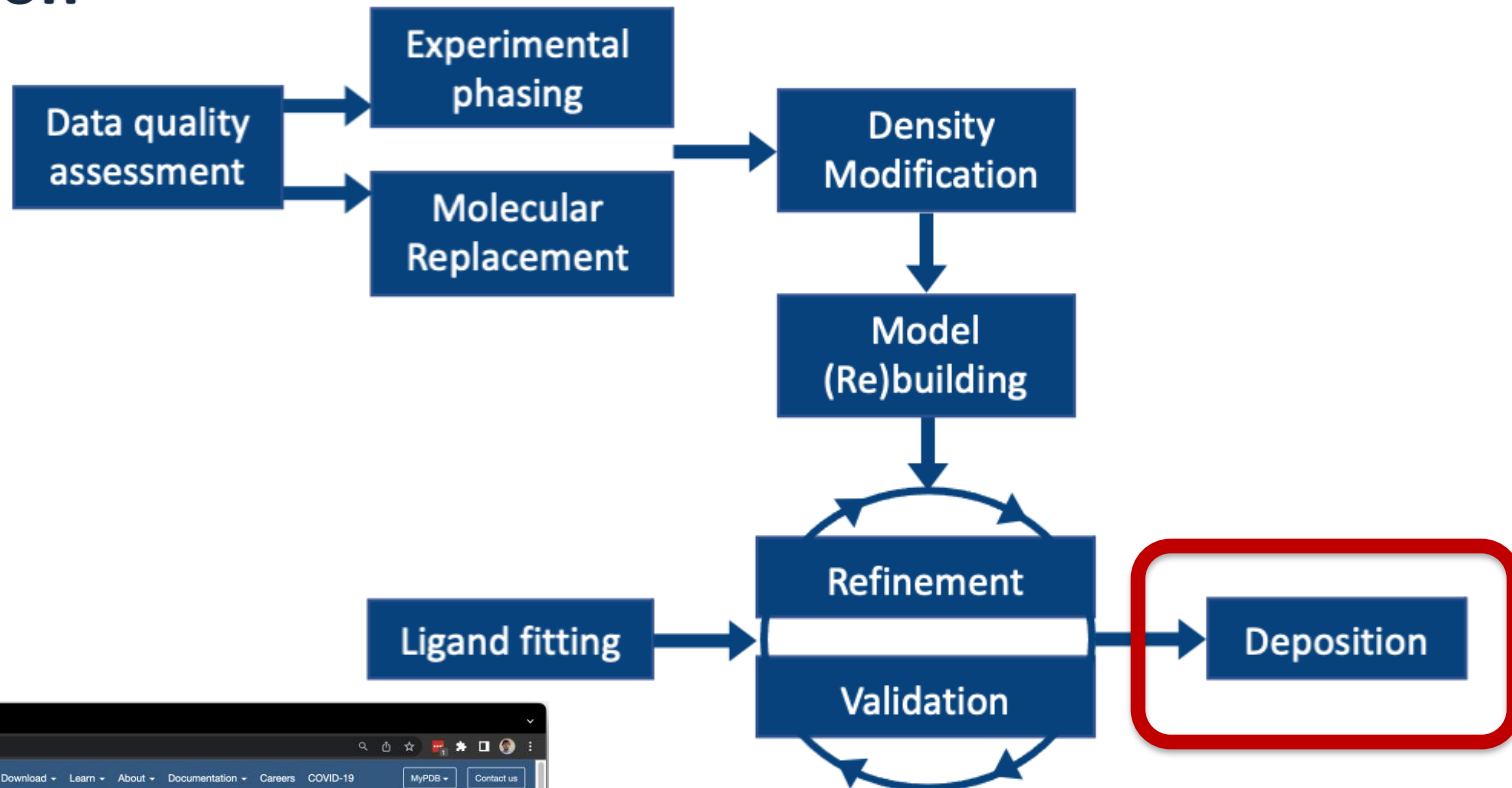


**Model to data fit**



Validation = checking model, data and model-to-data fit are all make sense and obey to prior expectations

# Deposition

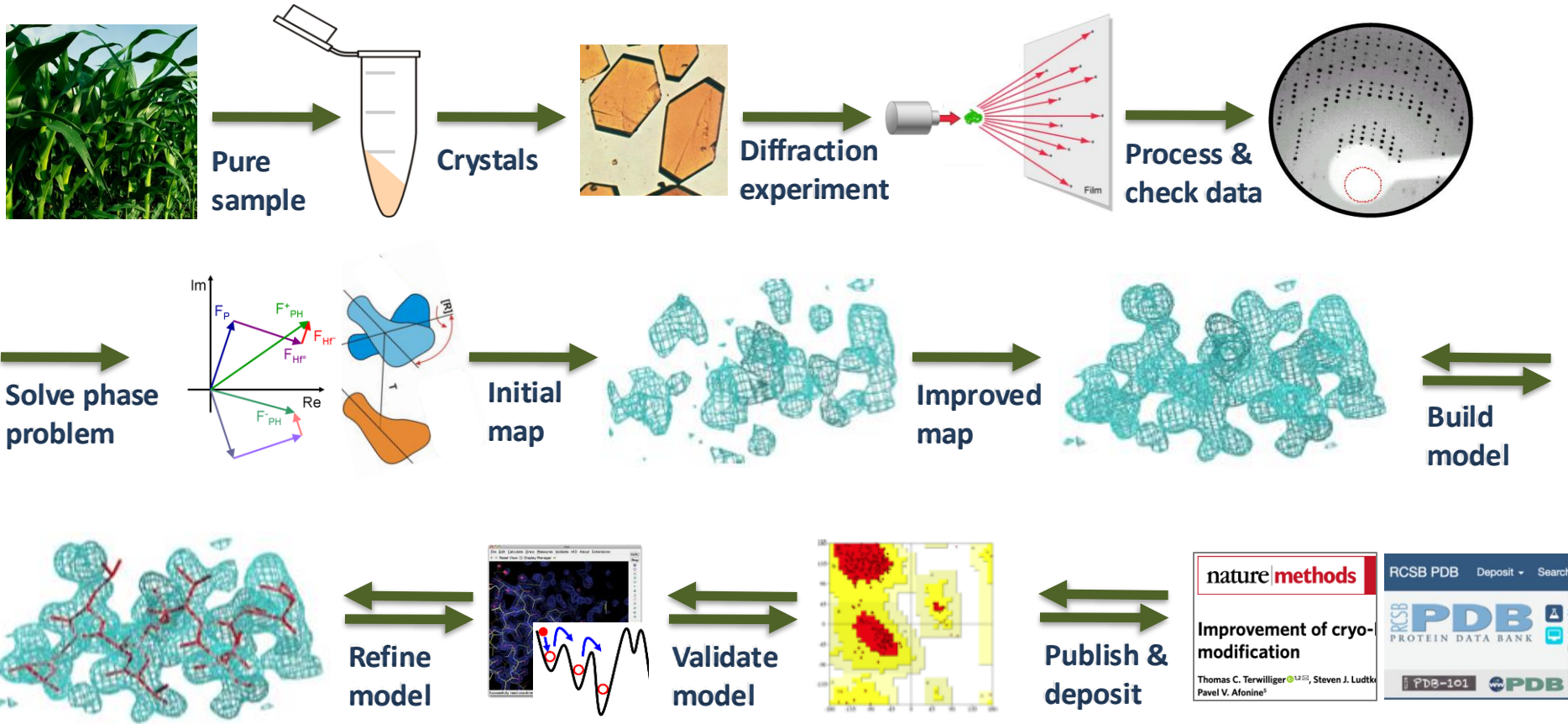


As of 2019, mmCIF is a mandatory format for crystallographic depositions

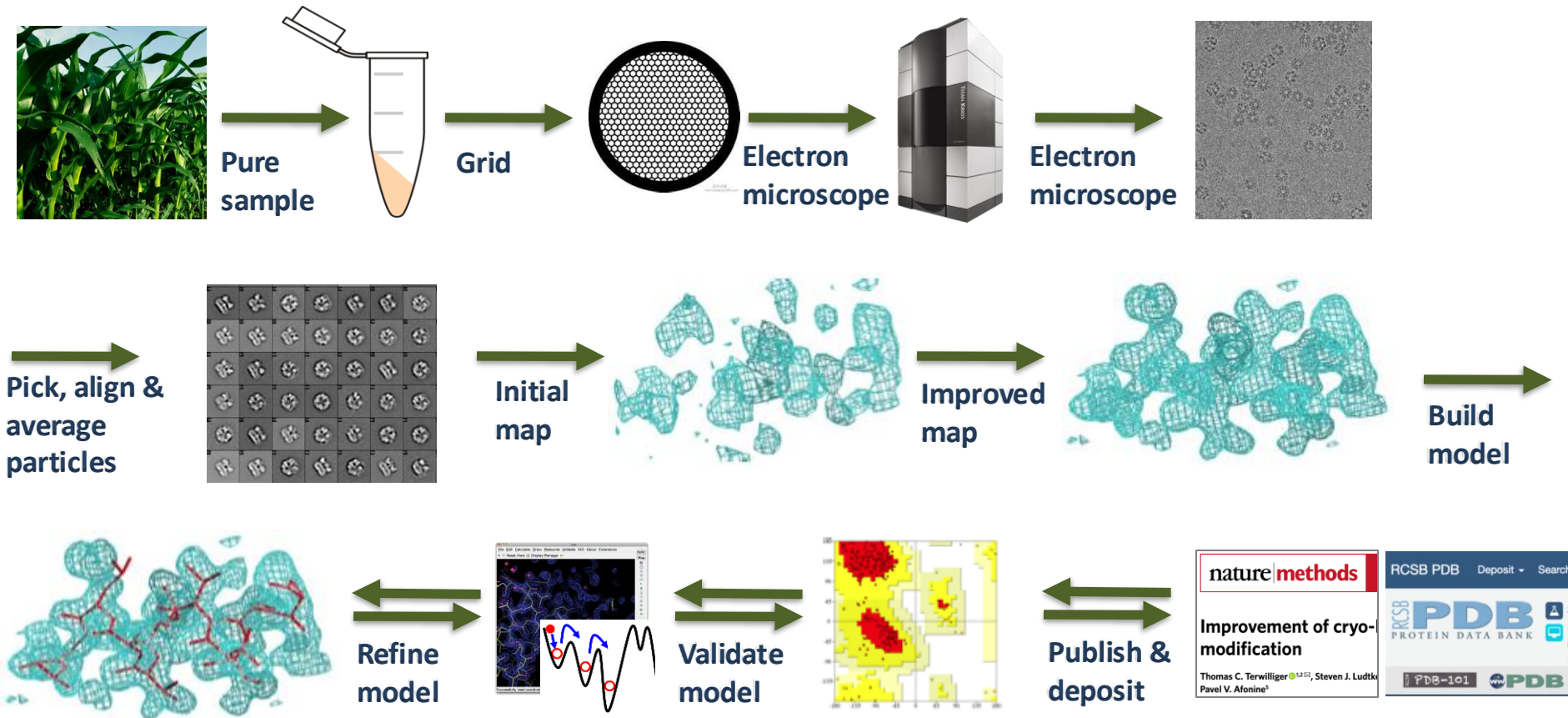


# **Cryo-EM structure solution**

# Solving structure by crystallography



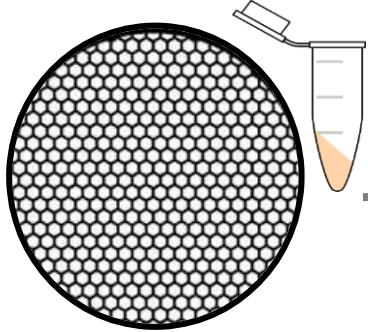
# Solving structure by electron cryo-microscopy



Unlike crystallography, no Phase Problem!

# Phenix tools for cryo-EM

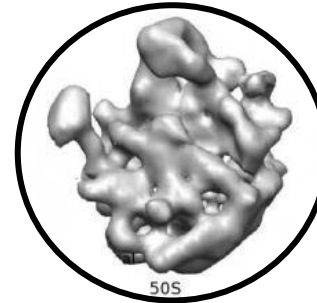
Sample



Data collection

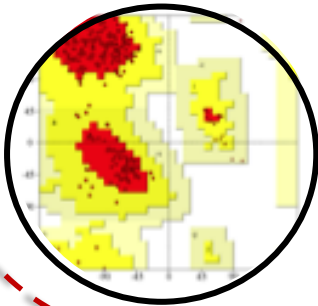


Data processing

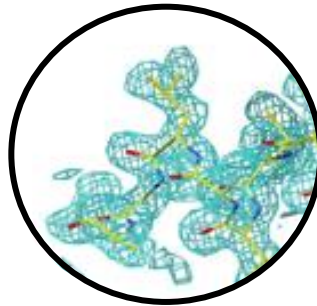


Map manipulations

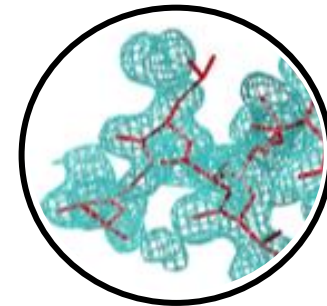
Validation



Model refinement



Model building

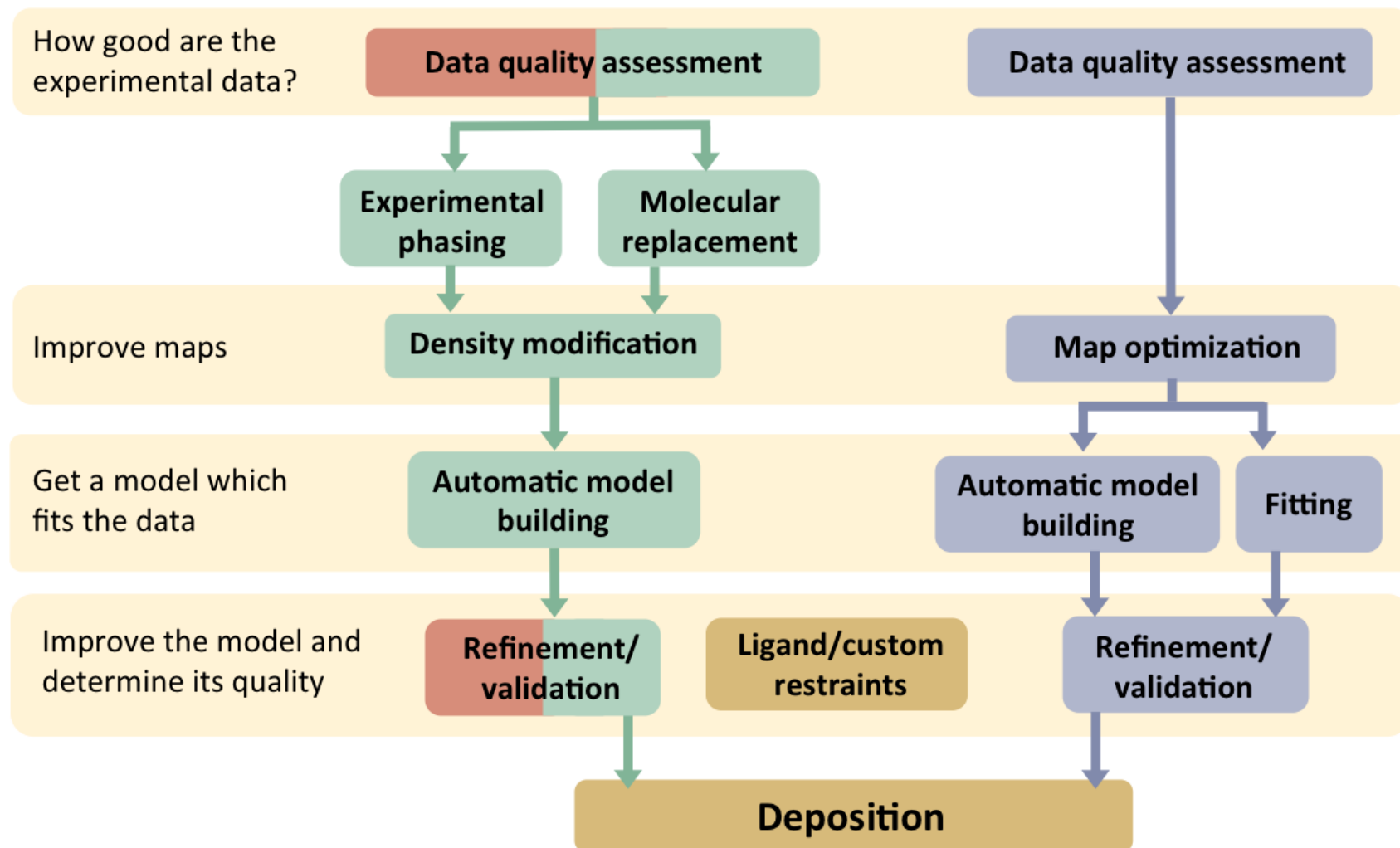


**Phenix**

# Phenix: tools for crystallography and cryo-EM

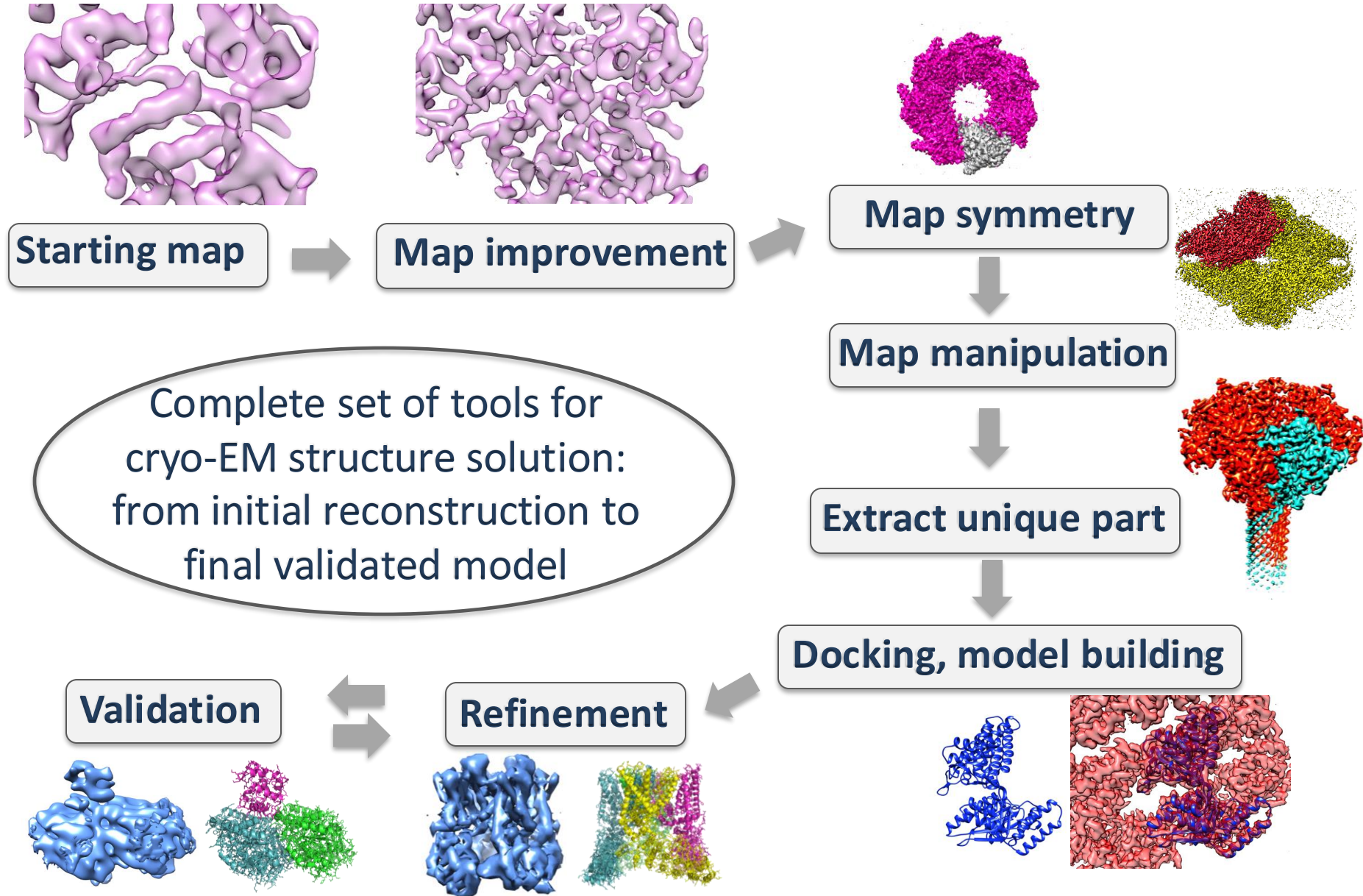
## Xray/neutron crystallography

## Cryo-EM





# Phenix tools for cryo-EM



# Phenix tools for cryo-EM: *GUI and command line*

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**PHENIX home**

Quit Preferences Help Citations ChimeraX Coot PyMOL KiNG Other tools Ask for help

**Actions** Job history

**Projects**

Show group: All groups Manage...

Select Delete New project Settings

ID	Last modified	# of jobs	R-free
63G	Jun 23 2022 05:26...	1	---
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yuya	May 28 2022 12:44...	1	---
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**Crystals:** Data analysis and manipulation  
Validation and map-based comparisons  
Experimental phasing  
Molecular replacement  
Maps (create, manipulate, compare)  
Enhanced maps (Polder, FEM, density-modified...)  
Model building  
Refinement  
Ligands

**Cryo-EM:** Map analysis, symmetry, manipulation  
Validation and map-based comparisons  
Map improvement  
Docking, model building and rebuilding  
Refinement

**Models:** Superpose, search, compare, analyze symmetry  
Modification, minimization and dynamics

**PDB Deposition**

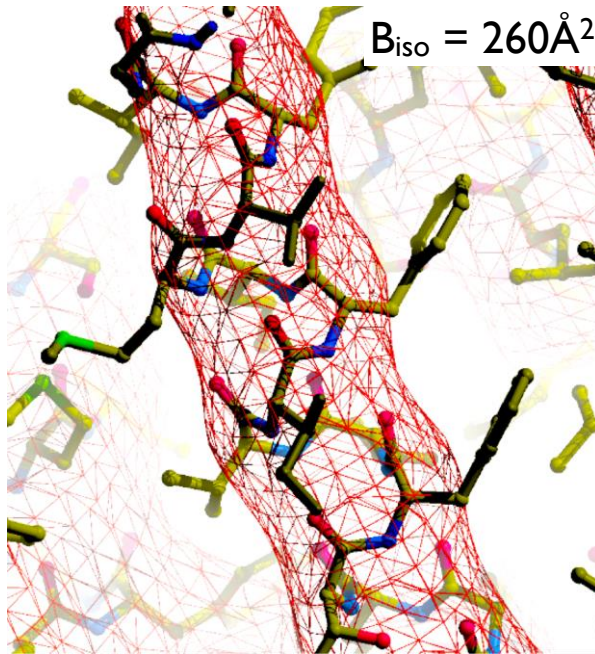
**Program search**

Current directory: /Users/pafonine/63\_goska/polder Browse...

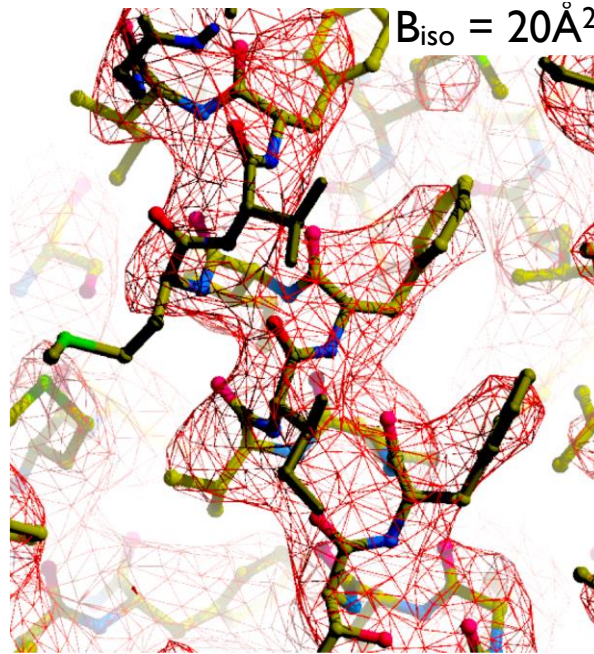
PHENIX version dev-svn-000 Project: 63G



# Automated map sharpening: *phenix.auto\_sharpen*



*Deposited Map*



*Autosharpened Map*

EMDB: 8414, PDB: 5tji

Fully automatic:

No manual trial-and-error | No parameters to adjust | Only inputs: map and resolution



STRUCTURAL  
BIOLOGY

ISSN 2059-7983

Automated map sharpening by maximization of  
detail and connectivity

Thomas C. Terwilliger,<sup>a,b\*</sup> Oleg V. Sobolev,<sup>c</sup> Pavel V. Afonine<sup>c,d</sup> and  
Paul D. Adams<sup>d,e</sup>

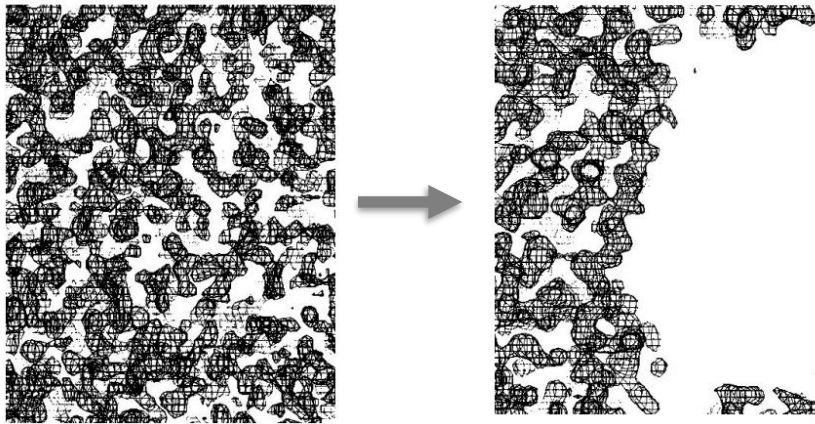
# Density modification: *phenix.density\_modify\_cryo\_em*

Similar principals for crystallography and cryo-EM:

change the map so that it is most consistent with what we know about macromolecules

## Crystallography

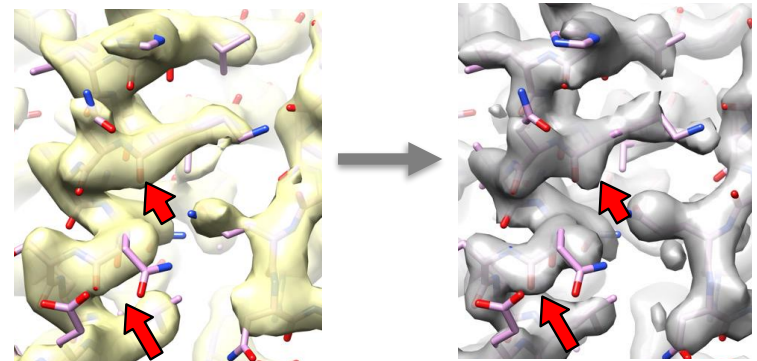
From uninterpretable to interpretable map



## Cryo-EM

Effect is less dramatic as in crystallography

- Can increase map resolution (0.05-0.3 Å)
- Can improve map clarity for interpretation



nature **methods**

ARTICLES

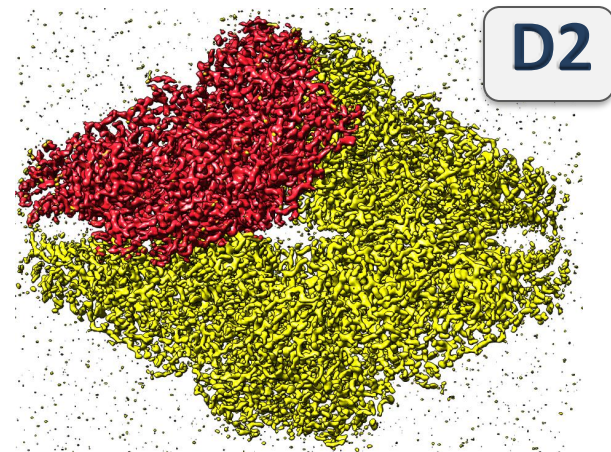
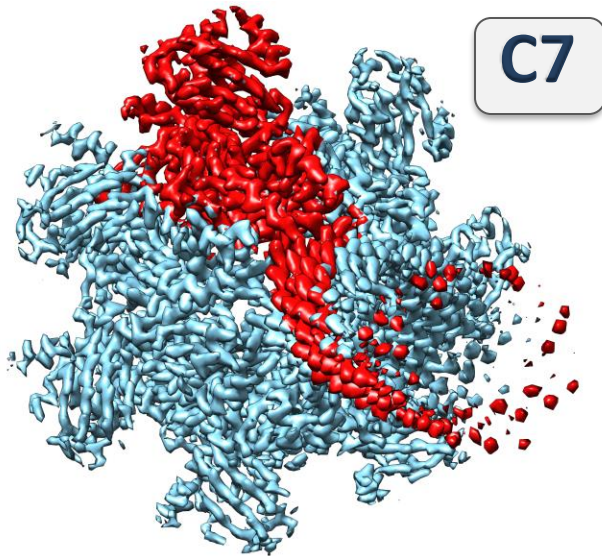
<https://doi.org/10.1038/s41592-020-0914-9>

Check for updates

## Improvement of cryo-EM maps by density modification

Thomas C. Terwilliger<sup>1,2</sup>✉, Steven J. Ludtke<sup>3</sup>, Randy J. Read<sup>4</sup>, Paul D. Adams<sup>5,6</sup> and Pavel V. Afonine<sup>5</sup>

# Finding map symmetry: *phenix.symmetry\_from\_map*

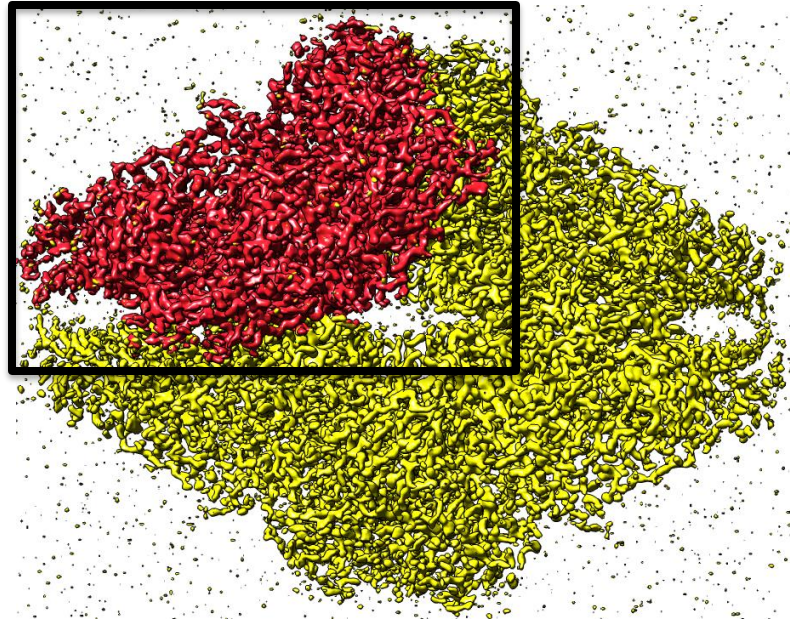


<http://phenix-online.org/newsletter/>

Tools for interpreting cryo-EM maps using models from the PDB



# Extracting unique part of map using *phenix.map\_box*

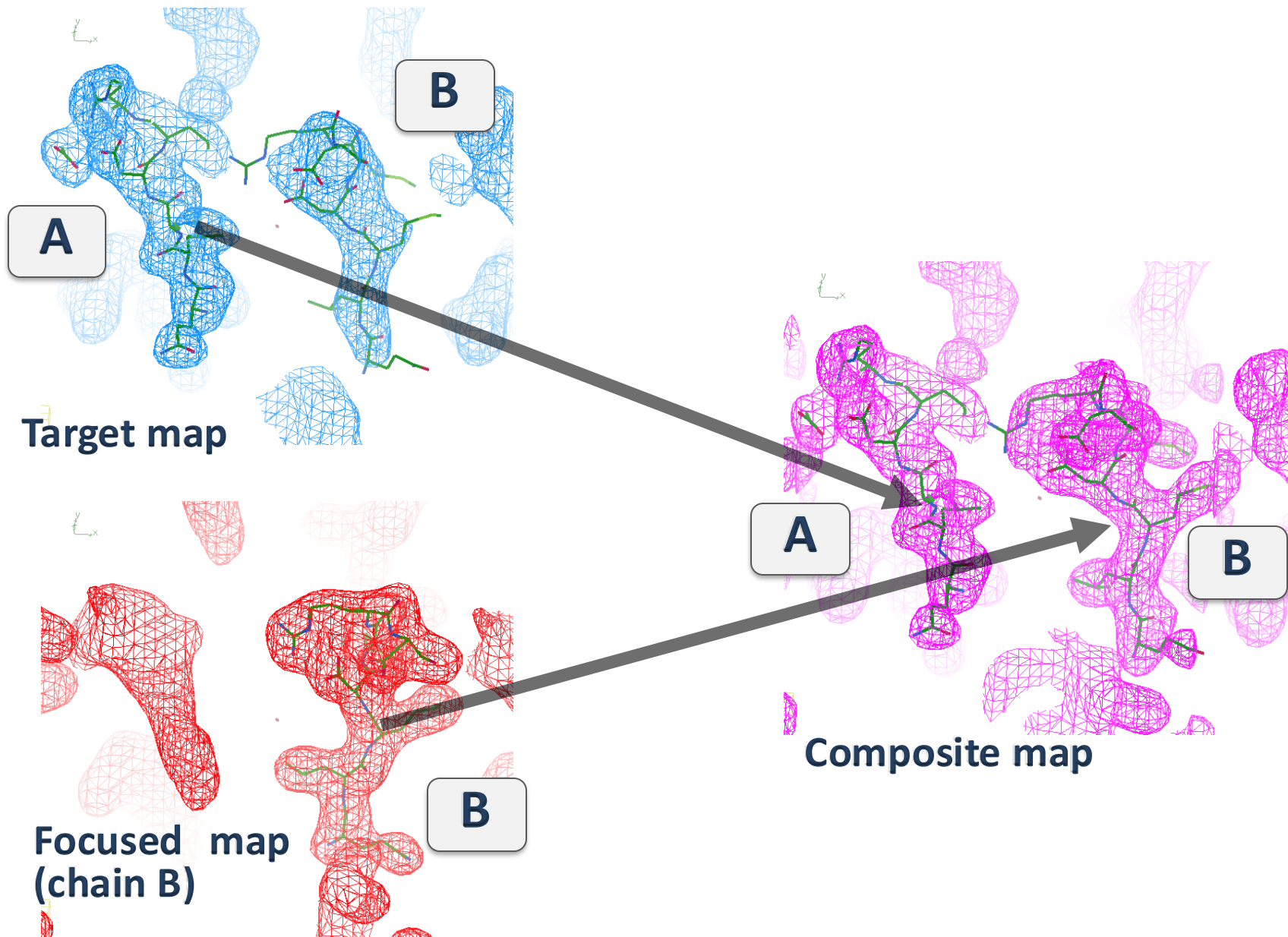


***Lots of options:*** use map only, use model, use symmetry, mask boxed map, and many more!

<http://phenix-online.org/newsletter/>

Tools for interpreting cryo-EM maps using models from the PDB

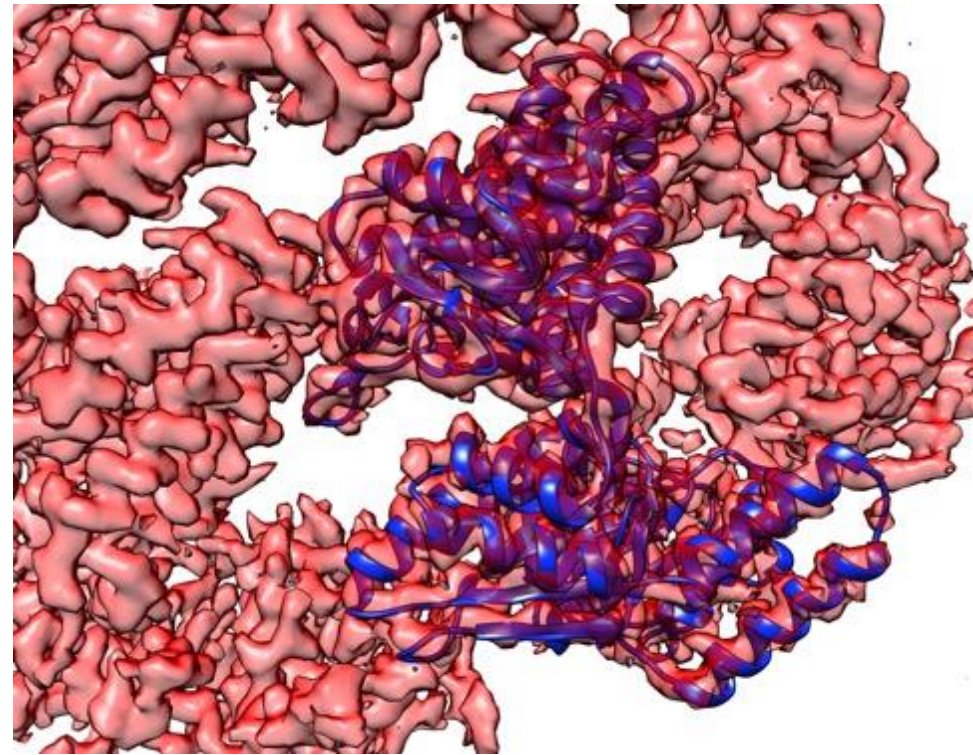
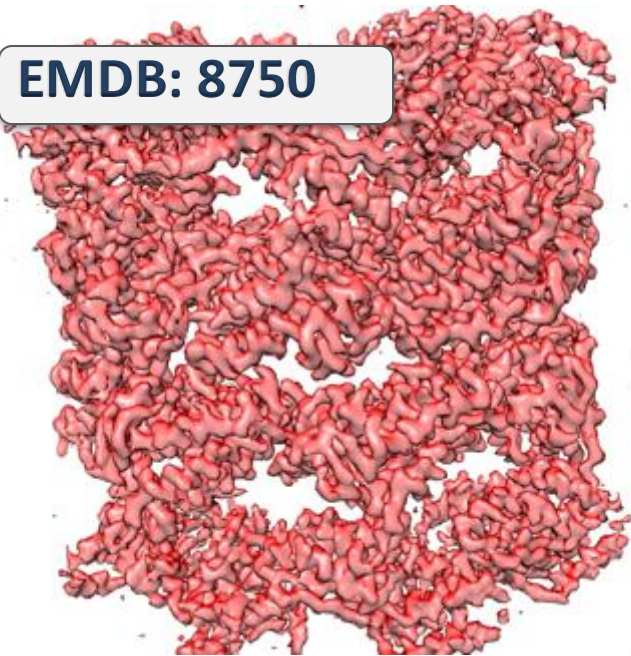
# Combining maps with *phenix.combine\_focused\_maps*





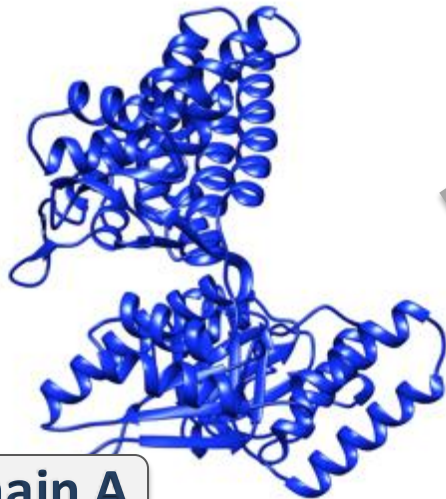
# Docking models with *phenix.dock\_in\_map*

EMDB: 8750



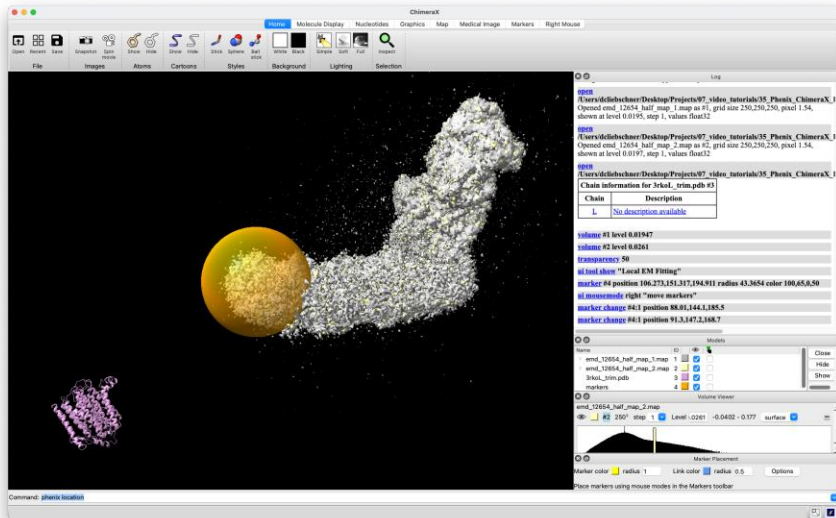
Chain A docked in map

1ss8 chain A

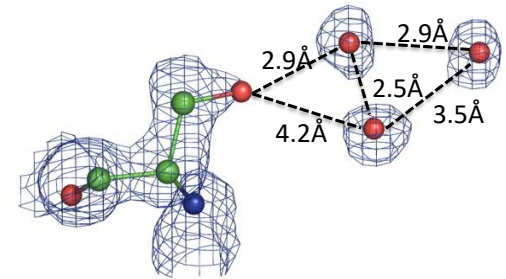
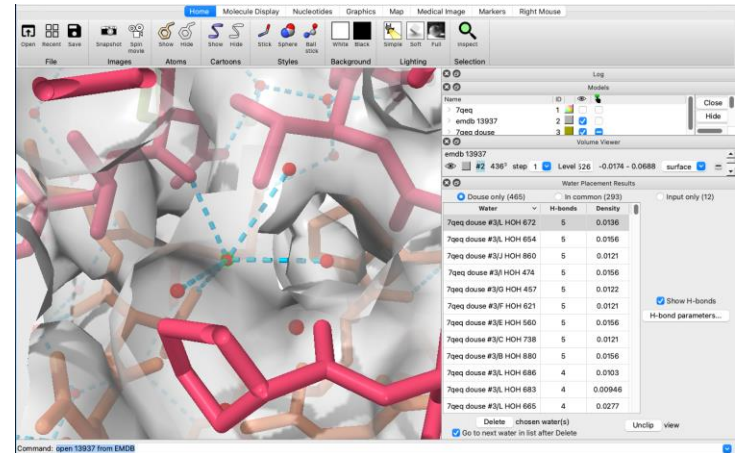


# Integration with ChimeraX

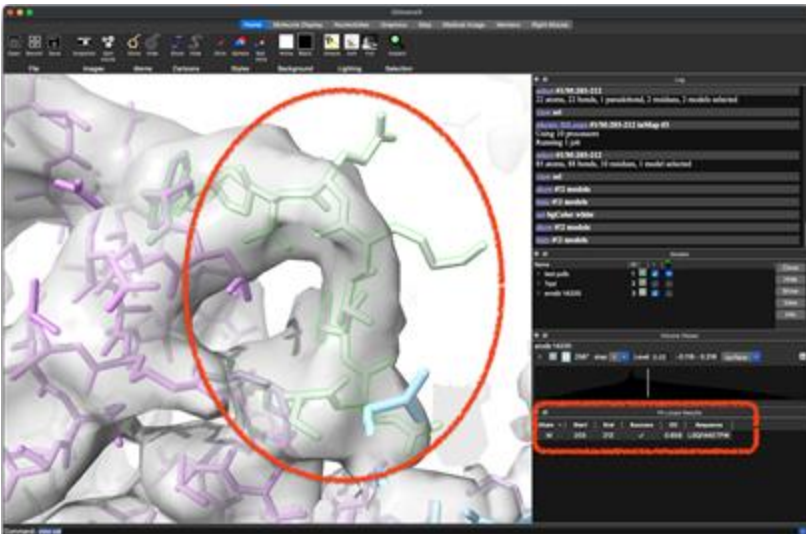
## Local EM map fitting



## Water building into EM maps



## Loop Fitting







# Automated model building: *phenix.map\_to\_model*

nature|**methods**

BRIEF COMMUNICATION

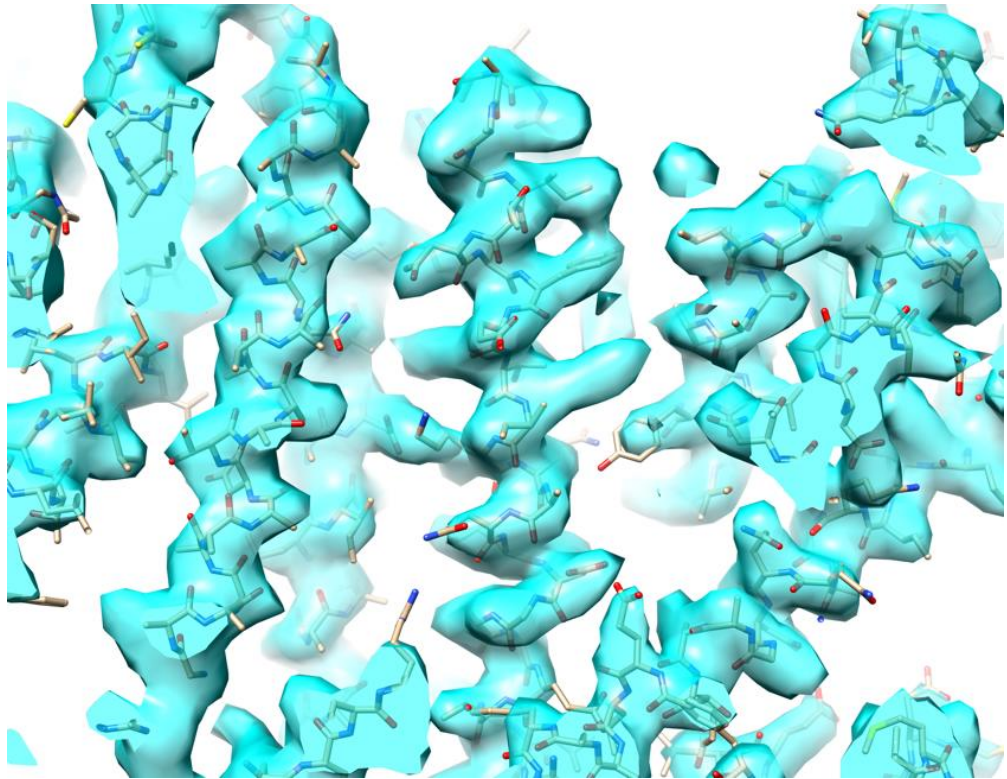
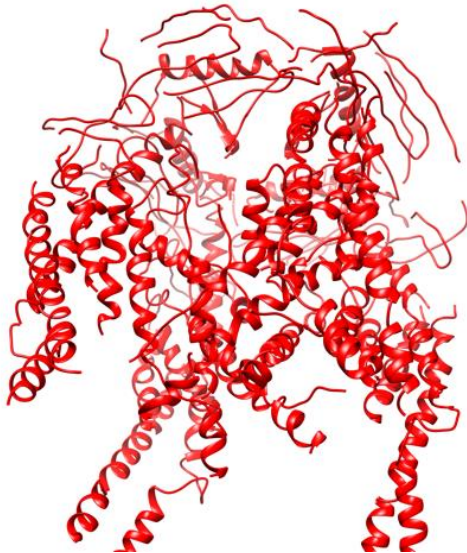
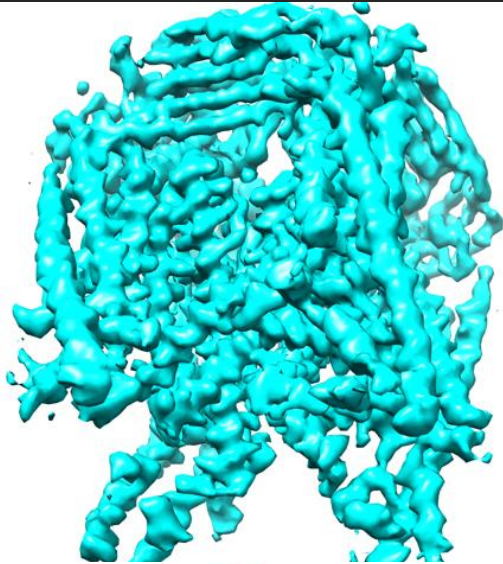
<https://doi.org/10.1038/s41592-018-0173-1>

## A fully automatic method yielding initial models from high-resolution cryo-electron microscopy maps

Thomas C. Terwilliger <sup>1,2\*</sup>, Paul D. Adams<sup>3,4</sup>, Pavel V. Afonine<sup>3,5</sup> and Oleg V. Sobolev <sup>3</sup>

# Automated model building: *phenix.map\_to\_model*

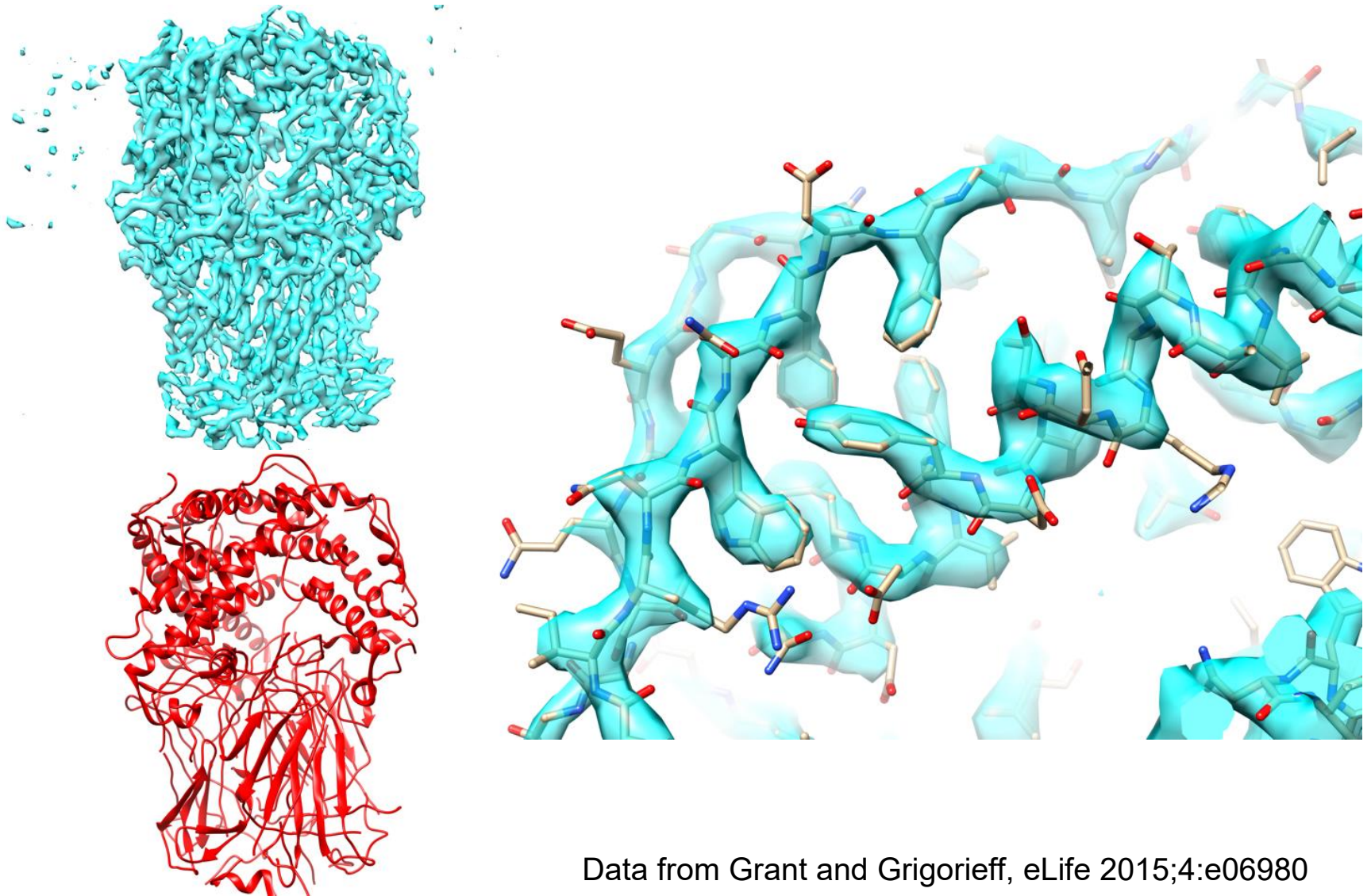
TRPML3 channel (4.1 Å, 78% built, 1.3 Å rmsd)



Data from Zhou, X. et al. (2017) Nat. Struct. Mol. Biol. 24: 1146

# Automated model building: *phenix.map\_to\_model*

Rotavirus VP6 (2.6 Å, 100% built, 0.9 Å rmsd)



Data from Grant and Grigorieff, eLife 2015;4:e06980

# Automated model building: *phenix.map\_to\_model*

## Automated model building, facts:

- No automated model building produces 100% complete and accurate model
- Produces initial model for further manual building
- The lower the resolution, the less complete and accurate the auto built model

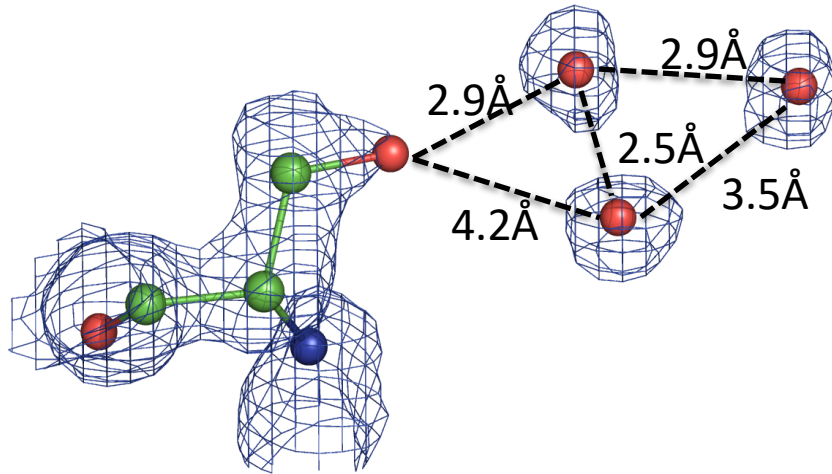


# Manual model building steps

If *phenix.map\_to\_model* fails or model is too big or else:

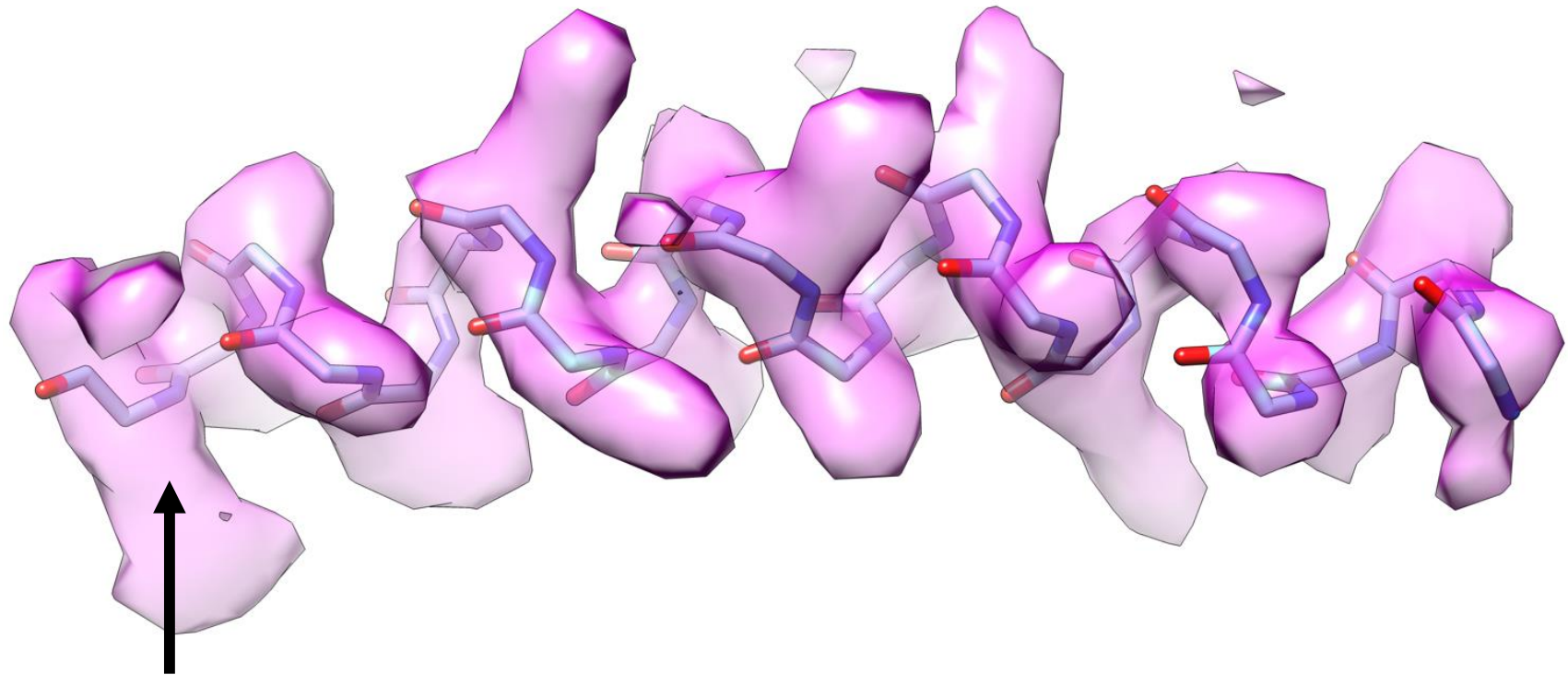
- Auto-sharpen the map
- Run Map Symmetry to obtain symmetry
- Run Map Box to obtain asymmetric unit (using symmetry)
- Run Map to Model on asymmetric unit
- Run Apply NCS Operators on model, with the trim overlap option (supplying the full map)

# Automated water building: *phenix.douse*



**Available in ChimeraX!**

# Sequence from map: *phenix.sequence\_from\_map*



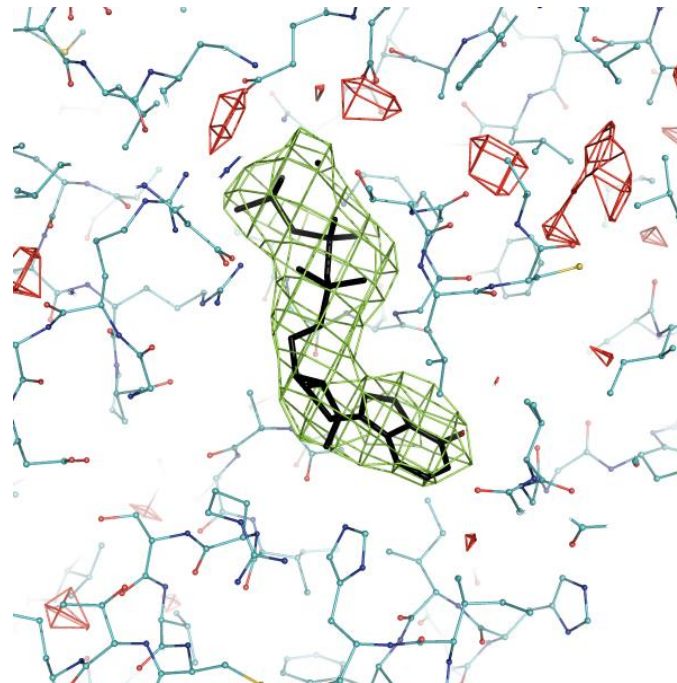
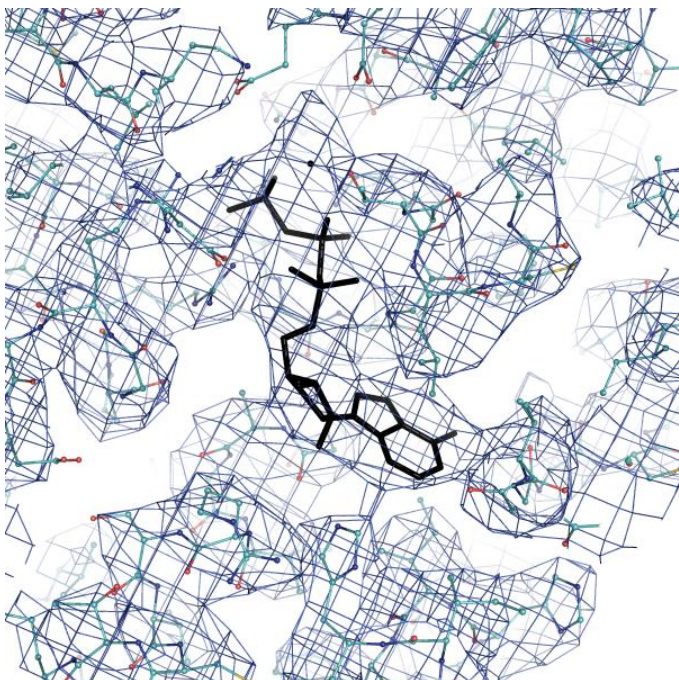
Residue	G	A	S	V	I	L	M	C	F	Y	K	R	W	H	E	D	Q	N	P	T
CC	0.30	0.50	0.53	0.47	0.58	0.62	0.68	0.59	0.83	0.77	0.71	0.69	0.70	0.82	0.65	0.64	0.60	0.60	0.35	0.47
Prob	3	0	0	0	0	0	1	0	40	23	5	5	4	9	2	2	1	0	2	0

- Determine probability of side chain at each C!
- Align sequence to maximize total probability for the chain



# Difference maps: *phenix.real\_space\_diff\_map*

5L4g, EMDDB 4002

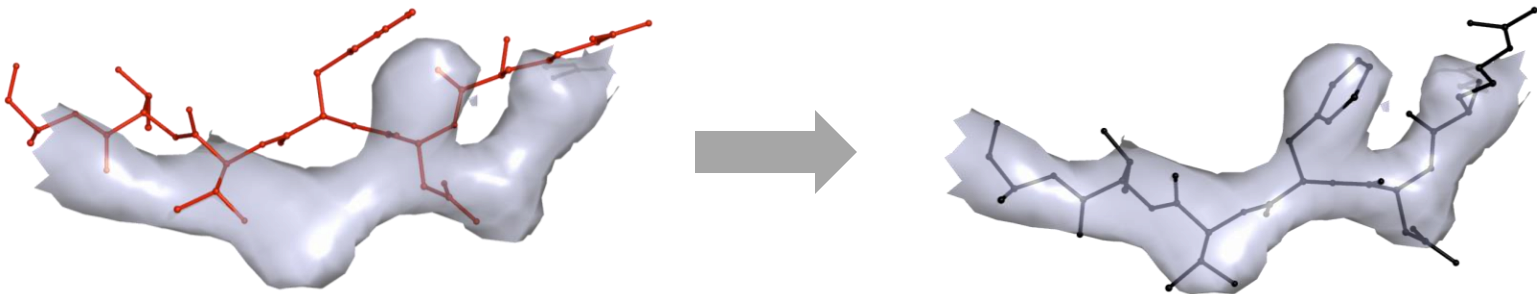


- Analogue of crystallographic Fo-Fc map
- Requires well-refined model (including B factors)

# Atomic model refinement: *phenix.real\_space\_refine*

## Direct refinement of atomic models against the map

 <p>Acta Cryst D</p>	<p>STRUCTURAL BIOLOGY</p>	<h3>Real-space refinement in <i>PHENIX</i> for cryo-EM and crystallography</h3>
<p>ISSN 2059-7983</p>		<p>Pavel V. Afonine,<sup>a,b*</sup> Billy K. Poon,<sup>a</sup> Randy J. Read,<sup>c</sup> Oleg V. Sobolev,<sup>a</sup> Thomas C. Terwilliger,<sup>d,e</sup> Alexandre Urzhumtsev<sup>f,g</sup> and Paul D. Adams<sup>a,h</sup></p>



# Structure prediction: new era in structural biology

AlphaFold changes everything and... nothing

*Tom Terwilliger, LANL*

# Structure determination in the AlphaFold era

## 1. Predict your structure



*Design your experiment based on predicted models  
(choose experimental approach, consider trimming at domain boundaries)*

## 2. Solve your structure



*Cryo-EM or X-ray MR with your prediction, SAD*

## 3. Update your prediction



*Run AlphaFold with your best model as a template*

## 4. Improve your structure



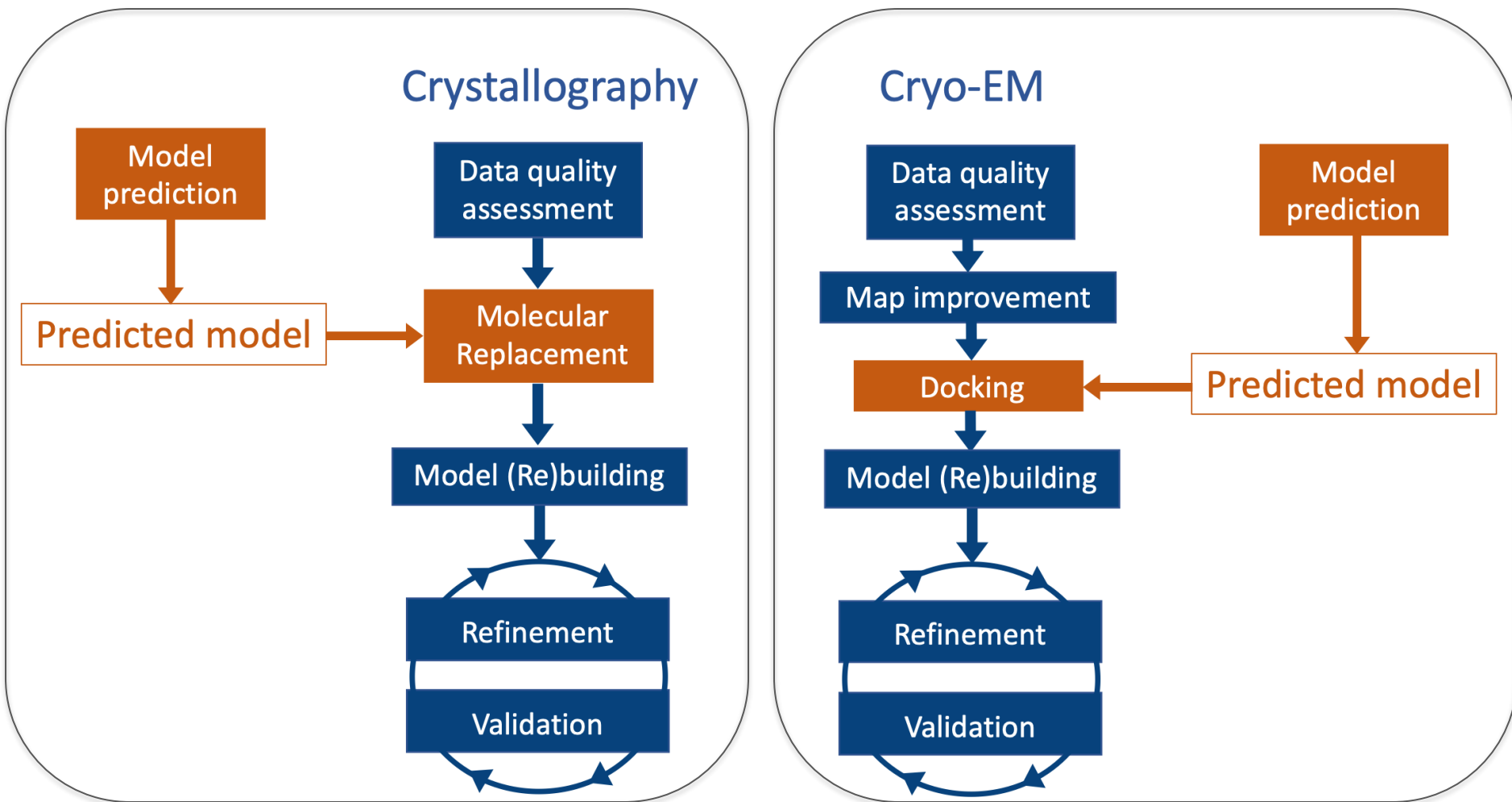
*Use your new predictions as hypotheses*

A large, thick, blue curved arrow that starts from the bottom right and points upwards and to the left, looping back to the 'Update your prediction' step.

*Iterate*

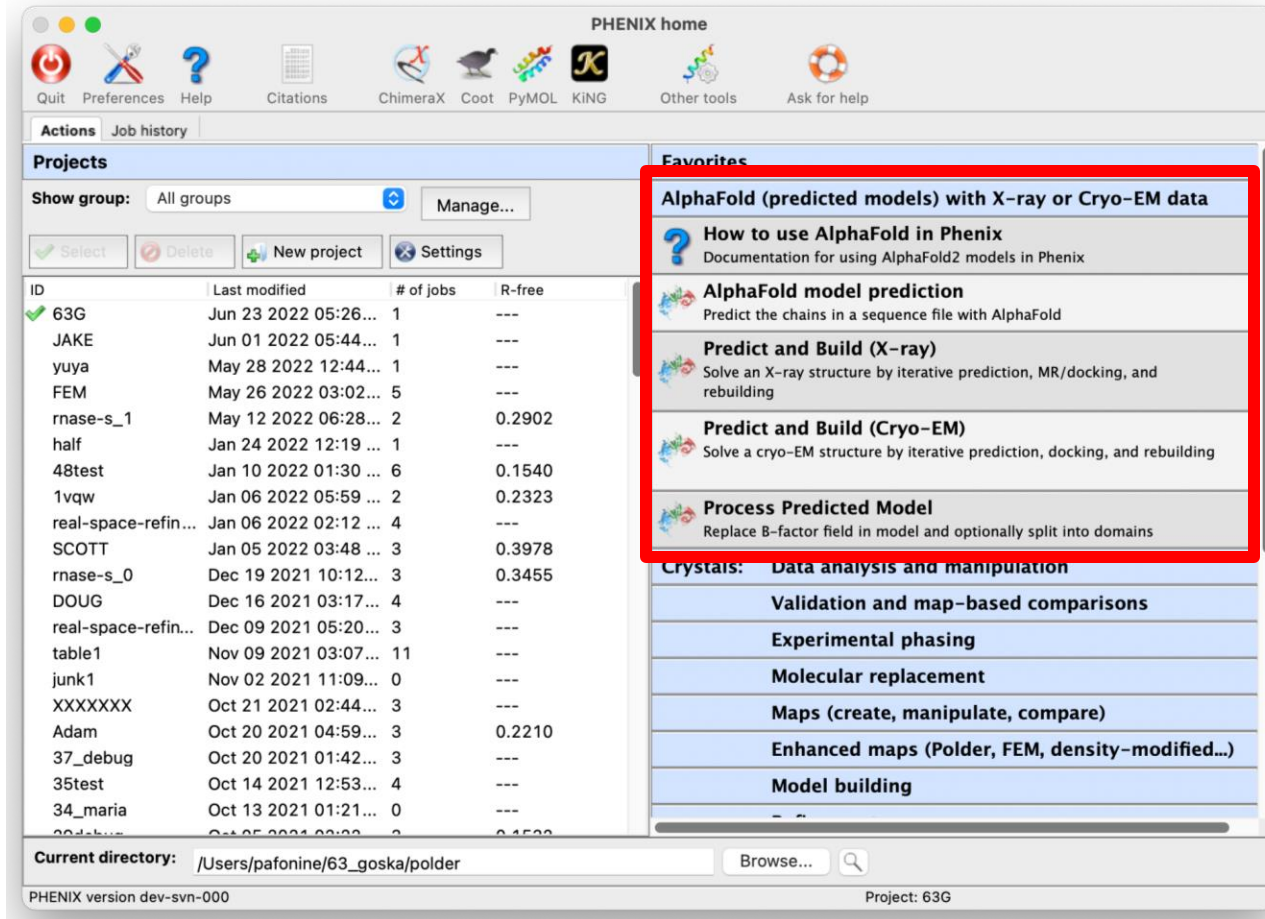
# How to use predictions?

Incorporate predictions into your structure determination workflow



Millán C. et al.. Assessing the utility of CASP14 models for molecular replacement. *Proteins*. 2021 Dec;89(12):1752–69.

# AphaFold and Phenix



The screenshot shows the PHENIX home window. The top menu bar includes 'Quit', 'Preferences', 'Help', 'Citations', 'ChimeraX', 'Coot', 'PyMOL', 'KING', 'Other tools', and 'Ask for help'. The main window is divided into two panes. The left pane, titled 'Projects', shows a table of projects with columns for ID, Last modified, # of jobs, and R-free. The right pane, titled 'Favorites', shows a list of actions. A red box highlights the 'AlphaFold (predicted models) with X-ray or Cryo-EM data' section, which includes the following actions:

- How to use AlphaFold in Phenix**: Documentation for using AlphaFold2 models in Phenix
- AlphaFold model prediction**: Predict the chains in a sequence file with AlphaFold
- Predict and Build (X-ray)**: Solve an X-ray structure by iterative prediction, MR/docking, and rebuilding
- Predict and Build (Cryo-EM)**: Solve a cryo-EM structure by iterative prediction, docking, and rebuilding
- Process Predicted Model**: Replace B-factor field in model and optionally split into domains

The bottom of the window shows the 'Current directory' as '/Users/pafonine/63\_goska/polder' and the 'Project' as '63G'.

ID	Last modified	# of jobs	R-free
63G	Jun 23 2022 05:26...	1	---
JAKE	Jun 01 2022 05:44...	1	---
yuya	May 28 2022 12:44...	1	---
FEM	May 26 2022 03:02...	5	---
rnase-s_1	May 12 2022 06:28...	2	0.2902
half	Jan 24 2022 12:19 ...	1	---
48test	Jan 10 2022 01:30 ...	6	0.1540
1vqw	Jan 06 2022 05:59 ...	2	0.2323
real-space-refin...	Jan 06 2022 02:12 ...	4	---
SCOTT	Jan 05 2022 03:48 ...	3	0.3978
rnase-s_0	Dec 19 2021 10:12...	3	0.3455
DOUG	Dec 16 2021 03:17...	4	---
real-space-refin...	Dec 09 2021 05:20...	3	---
table1	Nov 09 2021 03:07...	11	---
junk1	Nov 02 2021 11:09...	0	---
XXXXXXX	Oct 21 2021 02:44...	3	---
Adam	Oct 20 2021 04:59...	3	0.2210
37_debug	Oct 20 2021 01:42...	3	---
35test	Oct 14 2021 12:53...	4	---
34_maria	Oct 13 2021 01:21...	0	---
34_test	Oct 05 2021 02:22...	2	0.1522

`phenix.predict_and_build:`

Sequence + Data (X-ray or cryo-EM) **in** -> Refined model **out**

# Feedback & need driven



# Feedback & need driven – Example

## Vincent's post on phenix mailing list (phenixbb)

From

vincent Chaptal <vincent.chaptal@ibcp.fr>

↩ Reply

📧 Reply List

⌵

➡ Forward

📁 Archive

🗑 Junk

🗑 Delete

⋮ More

★

To

PHENIX user mailing list <phenixbb@phenix-online.org>

1/11/22, 04:48

Subject

**[phenixbb] refinement of an ensemble of structures -> cryoEM variability**

Hi Phenix-ers,

I thought to ask for something that I believe you have already implemented, but I'm not sure of the best tool to use.

I have a cryoEM map where I refine my "high resolution" structure. I also have the 3D variability of this map that shows several maps varying around the consensus high-res map. I want to refine an ensemble (20) of structures, one for every 20 maps around the consensus map. Is there a tool in phenix to do this?

I could refine individually the high-res structure into each map incrementally; since every map differs a little from the original one, Real-space-refinement could move the structure a little at a time. Then I could combine all the PDBs in an ensemble?

A tool to refine variability would be very useful. Input could be a PDB and an ensemble of maps, and output would be all the PDBs combined?

Thank you.

All the best  
Vincent

--  
Vincent Chaptal, PhD  
Director of GdR APPICOM  
Drug Resistance and Membrane Proteins Lab

MMSB -UMR5086  
7 passage du Vercors  
69007 LYON  
FRANCE  
+33 4 37 65 29 01  
<http://www.appicom.cnrs.fr>  
<http://mmsb.cnrs.fr/en/>

# Feedback & need driven – Example

The screenshot shows an email client interface with a list of conversations at the top and detailed views of four specific conversations below. The interface includes a search bar, filter buttons (Unread, Starred, Contact, Tags, Attachment), and a list of 11 conversations. The detailed view for each conversation shows the subject, correspondent, date, and the first message.

Subject	Correspondents	Date
Re: [phenixbb] refinement of an ensemble of structures -> cr...	vincent Chaptal	1/11/22, 16:36
Re: [phenixbb] refinement of an ensemble of structures -> cr...	Guillaume Gaullier	1/12/22, 04:09
Re: [phenixbb] refinement of an ensemble of structures -> cr...	vincent Chaptal	1/12/22, 04:36
Re: [phenixbb] refinement of an ensemble of structures -> cr...	vincent Chaptal	1/12/22, 11:18
Re: [phenixbb] refinement of an ensemble of structures -> cr...	vincent Chaptal	1/12/22, 16:03
Re: [phenixbb] refinement of an ensemble of structures -> cr...	Oliver Clarke	1/13/22, 11:17
Re: [phenixbb] refinement of an ensemble of structures -> cr...	phenixbb@phenix-online.org	1/13/22, 11:40

11 conversations Archive Delete

**[phenixbb] refinement of an ensemble of structures -> cryoEM** **vincent Chaptal** <vincent.chaptal@ibcp.fr> **variability** (9 messages)

Hi Phenix-ers, I thought to ask for something that I believe you have already implemented, but I'm not sure of the best tool to use. I have a cryoEM map where I refine my "high resolution" structure. I also have the 3D variability of this map that shows several maps varying around the consensu...

**[phenixbb] refinement of an ensemble of structures -> cryoEM** **Oliver Clarke** <olibclarke@gmail.com> **variability** (16 messages)

Hi, Just to add my two cents, I agree this would be really useful for a lot of folks. Analysis of continuously distributed variability is very common these days in cryoEM, and having a way to jointly refine an ensemble of models against a series of maps would be very handy. Cryodrgn, 3D-VA in cryo...

**[phenixbb] refinement of an ensemble of structures -> cryoEM** **Oliver Clarke** <olibclarke@gmail.com> **variability** (2 messages)

I guess it isn't all that different. If you run all jobs naively starting from a single model corresponding to the overall reconstruction, depending on the magnitude of the conformational changes the maps at either end of the series may be outside the radius of convergence of phenix.real\_space\_ref...

**link to data 3D variability cryoEM** (4 messages) **vincent Chaptal** <vincent.chaptal@ibcp.fr>

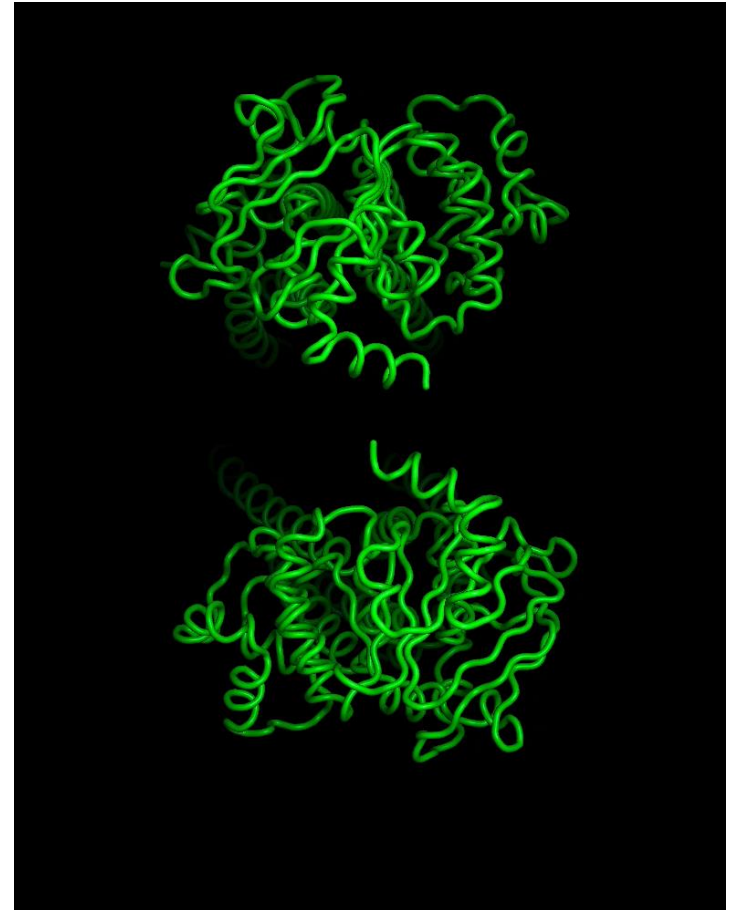
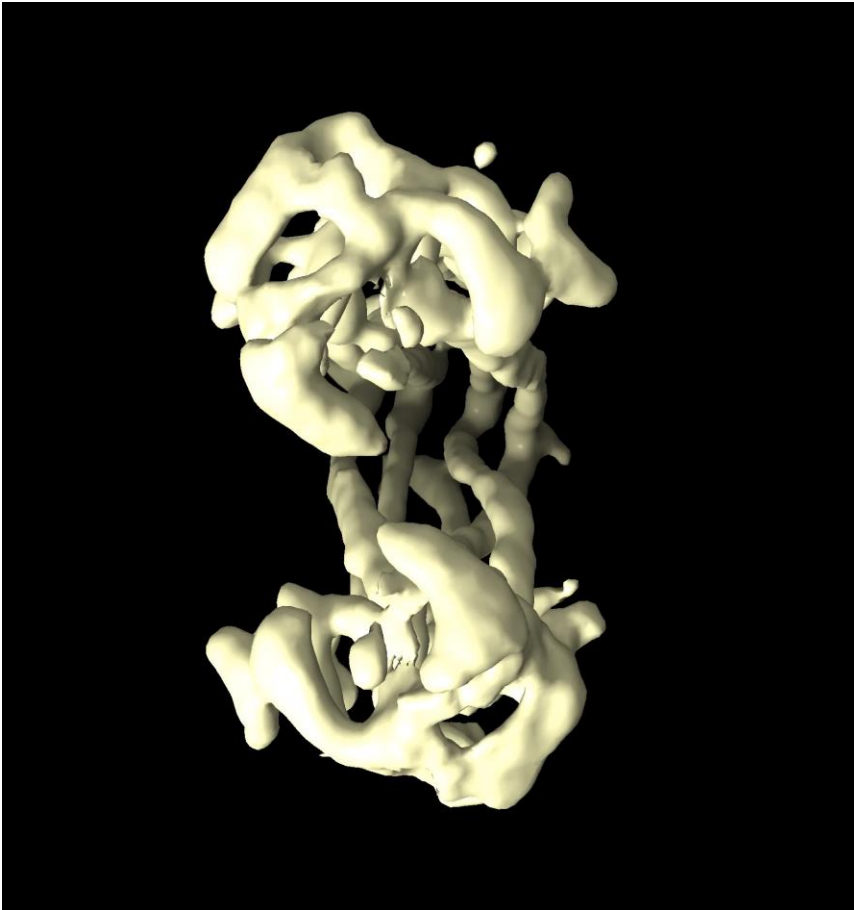
Hi Pavel, here are the data: <https://filesender.renater.fr/?s=download&token=da52b4ab-ecf1-4329-88dd-bec9433f155c> Thanks for looking into this, I'm sure it's going to be a very used tool. Best Vincent -- Vincent Chaptal, PhD Director of GdR APPICOM Drug Resistance and Membrane Proteins Lab...

Selected: 63 Total: 63

63 emails later....


## Feedback & need driven – Example

... **63 emails later**, we came up with a tool to effectively model ensemble of maps with ensemble of atomic models in a fully automated manner...



# Feedback & need driven – Example


... and wrapped that into a publication and a user-accessible tool



Contents lists available at [ScienceDirect](#)

## BBA - Biomembranes

journal homepage: [www.elsevier.com/locate/bbamem](http://www.elsevier.com/locate/bbamem)




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Review

### Conformational space exploration of cryo-EM structures by variability refinement

Pavel V. Afonine<sup>a,\*</sup>, Alexia Gobet<sup>b</sup>, Loïck Moissonnier<sup>b</sup>, Juliette Martin<sup>b</sup>, Billy K. Poon<sup>a</sup>, Vincent Chaptal<sup>b,\*</sup>

<sup>a</sup> Molecular Biosciences and Integrated Bioimaging, Lawrence Berkeley National Laboratory, 1 Cyclotron Road, Berkeley, CA 94720, USA  
<sup>b</sup> Molecular Microbiology and Structural Biochemistry, UMR5086 CNRS University Lyon1, 7 passage du Vercors, 69007 Lyon, France



Phenix home

Quit Preferences Help Citations ChimeraX Coot PyMOL KING Tools Help Server

**Actions** Job history

### Projects

Show group: All groups Manage...

Select Delete New project Import project Settings

ID	Last modified	# of jobs	R-free
✓ zz19	Apr 25 2025 04:09 ...	5	0.1320
aleks	Apr 14 2025 09:41 ...	1	---
zkdebug	Apr 13 2025 03:02 ...	5	---
LPtests	Apr 10 2025 12:59 ...	0	---
9ewk	Apr 09 2025 01:05 ...	3	0.0713
JoseB	Apr 01 2025 04:50 ...	5	0.1996
moid	Mar 31 2025 03:14 ...	1	---
7tur	Mar 14 2025 03:10 ...	1	---
8tk6	Mar 10 2025 12:46 ...	1	---
hnn46	Feh 27 2025 03:39 ...	1	---

**Current directory:** /Users/pafonine/Desktop/all/phenix/zz19 Browse...

Phenix version dev-5661 Project: zz19

- map analysis and manipulation
  - Map improvement
  - Docking and model building
  - Real-space refinement
    - Real-space refinement**  
Automated refinement using real-space maps (Cryo-EM, X-)
    - Variability refinement**  
Variability refinement using real-space maps
  - Validation
  - Ligands
  - Models
  - Superpose, search, compare, analyze symmet

# Automated re-refinement of deposited cryo-EM models

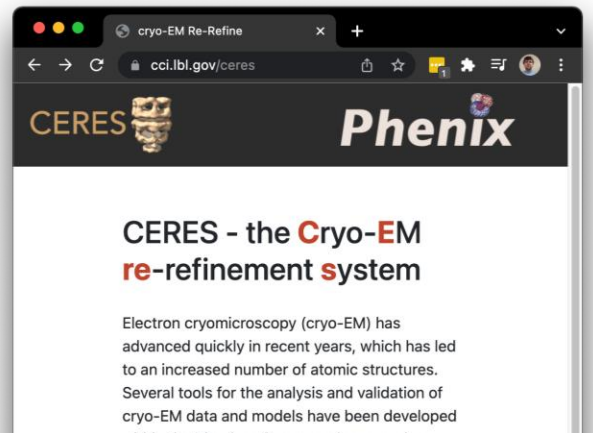


Table showing results for November 2021

Rerefinement month: November 2021

The table shows a selection of parameters. Activate more columns using the drop-down menus below.  
peach highlight: initial model; blue highlight: re-refined model

Resolution Map vs model Geometry Ramachandran Composition Other

Status	PDB code	EMDB code	Release Date	Resolution	Bond rmsd	Angle rmsd	Clashscore	Rama outliers	CC <sub>box</sub>	CC <sub>mask</sub>	Bond rmsd	Angle rmsd	Clashscore	Rama outliers	RMSD	CC <sub>box</sub>	CC <sub>mask</sub>	# residues	Logfile
✓	7SDE	23517	2021-09-29	3.2	0.004	0.92	25.84	0.3	0.74	0.77	0.005	0.79	10.50	0.0	0.61	0.75	0.79	726	<a href="#">logfile</a>
✓	7VH1	31983	2021-09-20	4.2	0.009	1.32	5.93	0.2	0.79	0.75	0.003	0.74	11.51	0.0	3.34	0.79	0.76	1661	<a href="#">logfile</a>
✓	7VH3	31985	2021-09-20	3.6	0.005	1.09	7.95	0.4	0.68	0.69	0.011	1.13	13.24	0.1	1.67	0.70	0.73	1612	<a href="#">logfile</a>
✓	7VGQ	31975	2021-09-18	4.0	0.009	1.27	7.11	0.2	0.71	0.75	0.004	0.75	9.16	0.1	0.72	0.70	0.76	1661	<a href="#">logfile</a>
X	7PQE	13591	2021-09-17	3.7	0.006	0.68	12.28	0.0	0.81	0.82	—	—	—	—	—	—	—	1365	<a href="#">logfile</a>
✓	7VG2	31963	2021-09-14	3.1	0.016	1.43	17.56	0.2	0.72	0.79	0.004	0.67	14.49	0.0	1.24	0.72	0.79	845	<a href="#">logfile</a>
X	7PPo	13583	2021-09-14	2.91	—	—	—	—	—	—	—	—	—	—	—	—	—	1382	<a href="#">logfile</a>
✓	7VG3	31964	2021-09-14	3.73	0.006	1.15	7.51	0.6	0.80	0.84	0.004	0.74	9.83	0.0	1.09	0.79	0.84	860	<a href="#">logfile</a>

- Developers: track the impact of new
- Users: see how your models benefit from improved methods and tools

## research papers



STRUCTURAL  
BIOLOGY

ISSN 2059-7983

## CERES: a cryo-EM re-refinement system for continuous improvement of deposited models

Dorothee Liebschner,<sup>a,\*</sup> Pavel V. Afonine,<sup>a</sup> Nigel W. Moriarty,<sup>a</sup> Billy K. Poon,<sup>a</sup>  
Vincent B. Chen<sup>b</sup> and Paul D. Adams<sup>a,c</sup>

# Phenix: tools for crystallography and cryo-EM



STRUCTURAL  
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Germany

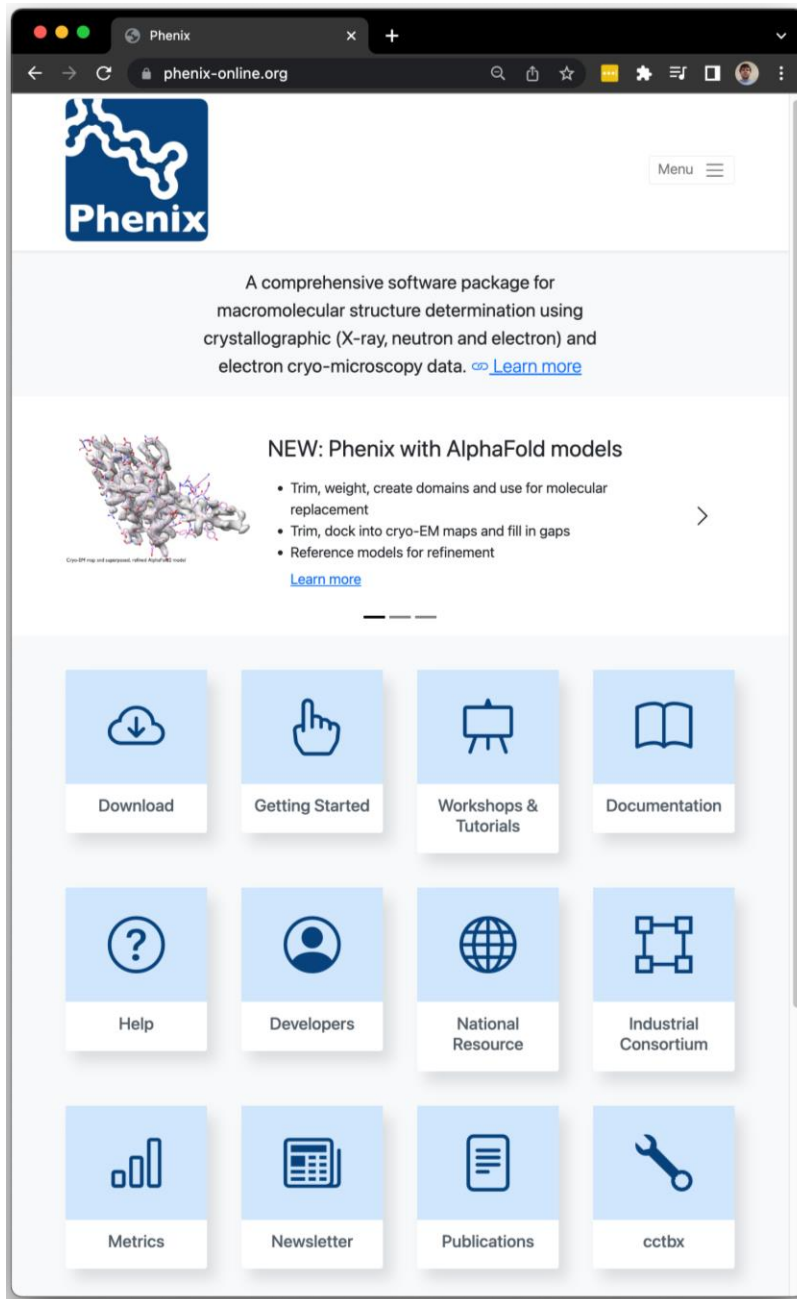
feature articles

## Macromolecular structure determination using X-rays, neutrons and electrons: recent developments in *Phenix*

Dorothee Liebschner,<sup>a</sup> Pavel V. Afonine,<sup>a</sup> Matthew L. Baker,<sup>b</sup> Gábor Bunkóczi,<sup>c,‡</sup> Vincent B. Chen,<sup>d</sup> Tristan I. Croll,<sup>c</sup> Bradley Hintze,<sup>d,§</sup> Li-Wei Hung,<sup>e</sup> Swati Jain,<sup>d,¶</sup> Airlie J. McCoy,<sup>c</sup> Nigel W. Moriarty,<sup>a</sup> Robert D. Oeffner,<sup>c</sup> Billy K. Poon,<sup>a</sup> Michael G. Prisant,<sup>d</sup> Randy J. Read,<sup>c</sup> Jane S. Richardson,<sup>d</sup> David C. Richardson,<sup>d</sup> Massimo D. Sammito,<sup>c</sup> Oleg V. Sobolev,<sup>a</sup> Duncan H. Stockwell,<sup>c</sup> Thomas C. Terwilliger,<sup>e,f</sup> Alexandre G. Urzhumtsev,<sup>g,h</sup> Lizbeth L. Videau,<sup>d</sup> Christopher J. Williams<sup>d</sup> and Paul D. Adams<sup>a,i,\*</sup>



# Phenix resources



Phenix paper

Video tutorials

Documentation

Relevant papers

Bi-annual newsletters

PDFs with slides from workshops

# User support

- **Feedback, questions, help**

Mailing list (anyone signed up): [phenixbb@phenix-online.org](mailto:phenixbb@phenix-online.org)

Bug reports (developers only): [bugs@phenix-online.org](mailto:bugs@phenix-online.org)

Ask for help (developers only): [help@phenix-online.org](mailto:help@phenix-online.org)

- **Reporting a bug or asking for help:**

- We can't help you if you don't help us to understand your problem
- Make sure the problem still exist using the latest *Phenix* version
- Send us all inputs (files, non-default parameters) and tell us steps that lead to the problem
- All data sent to us is kept confidentially