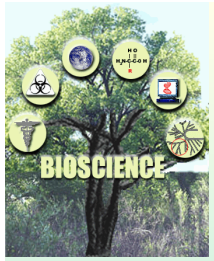
Phenix

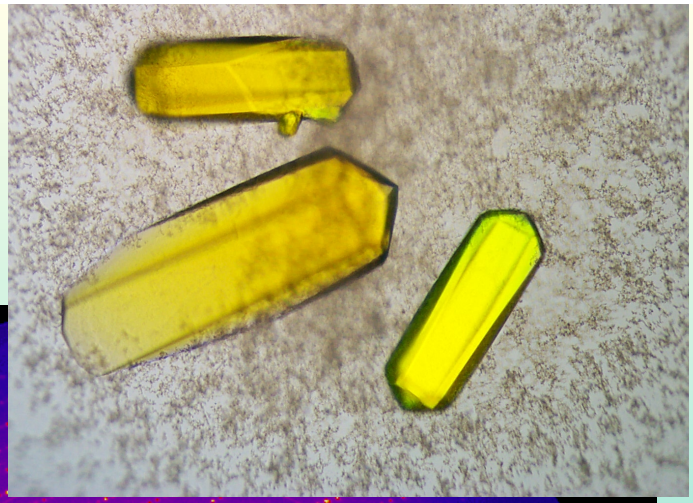
Tools for Easy and Difficult Problems: Automation of Structure Determination by Macromolecular Crystallography

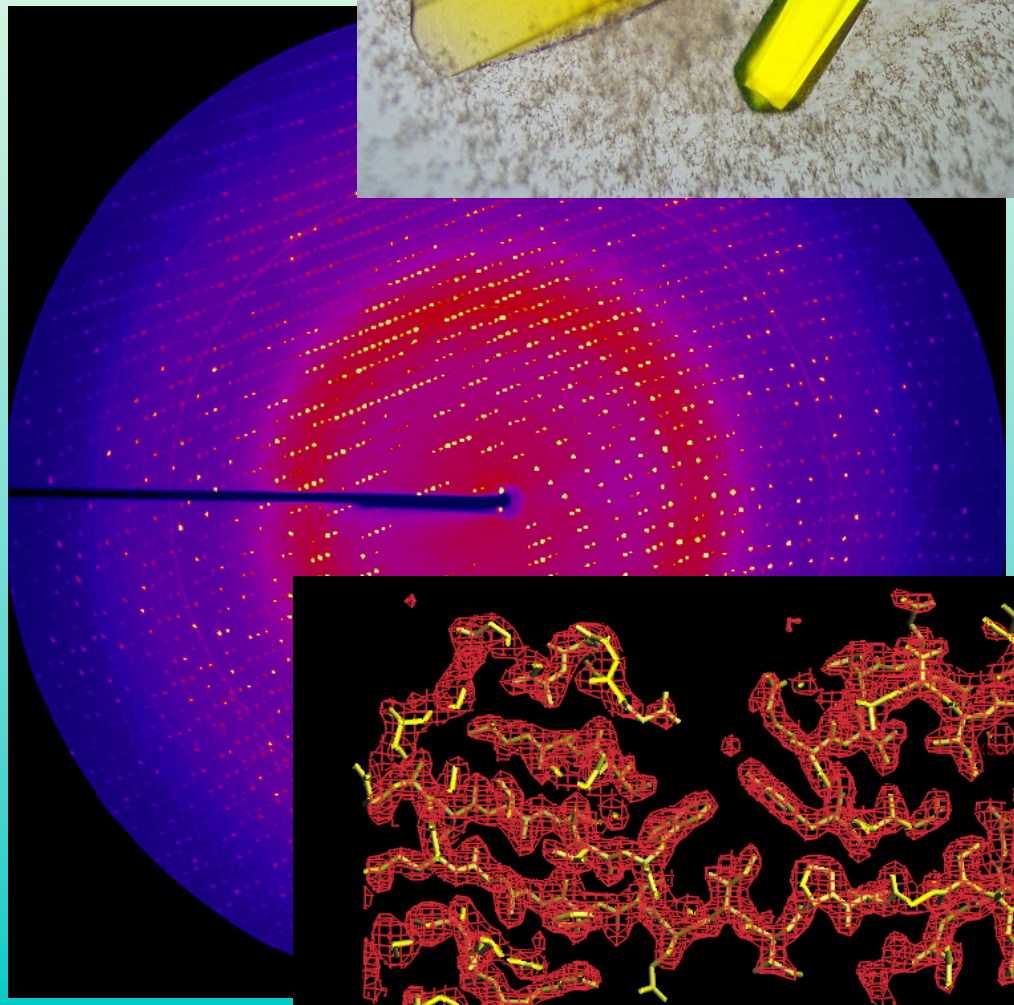
Indo-US workshop on
Macromolecular Structure Determination
Feb. 21-24, 2011

Tom Terwilliger

Los Alamos National Laboratory





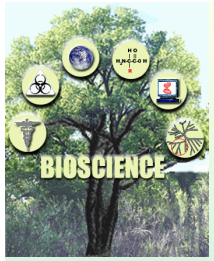


A brief introduction to X-ray crystallography

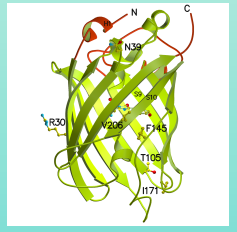
Growing protein crystals

Looking at crystals with X-rays

Getting pictures of proteins from diffraction spots

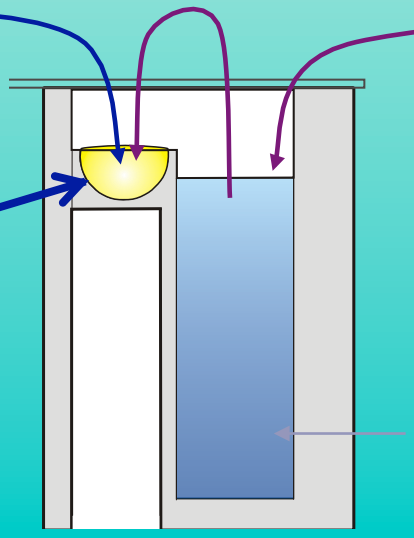


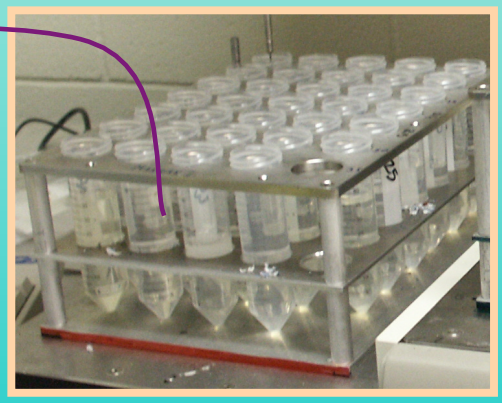
Growing protein crystals



Protein

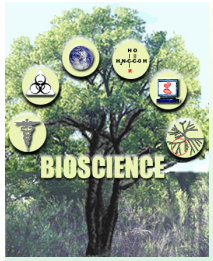
Protein +alcohol=Crystals



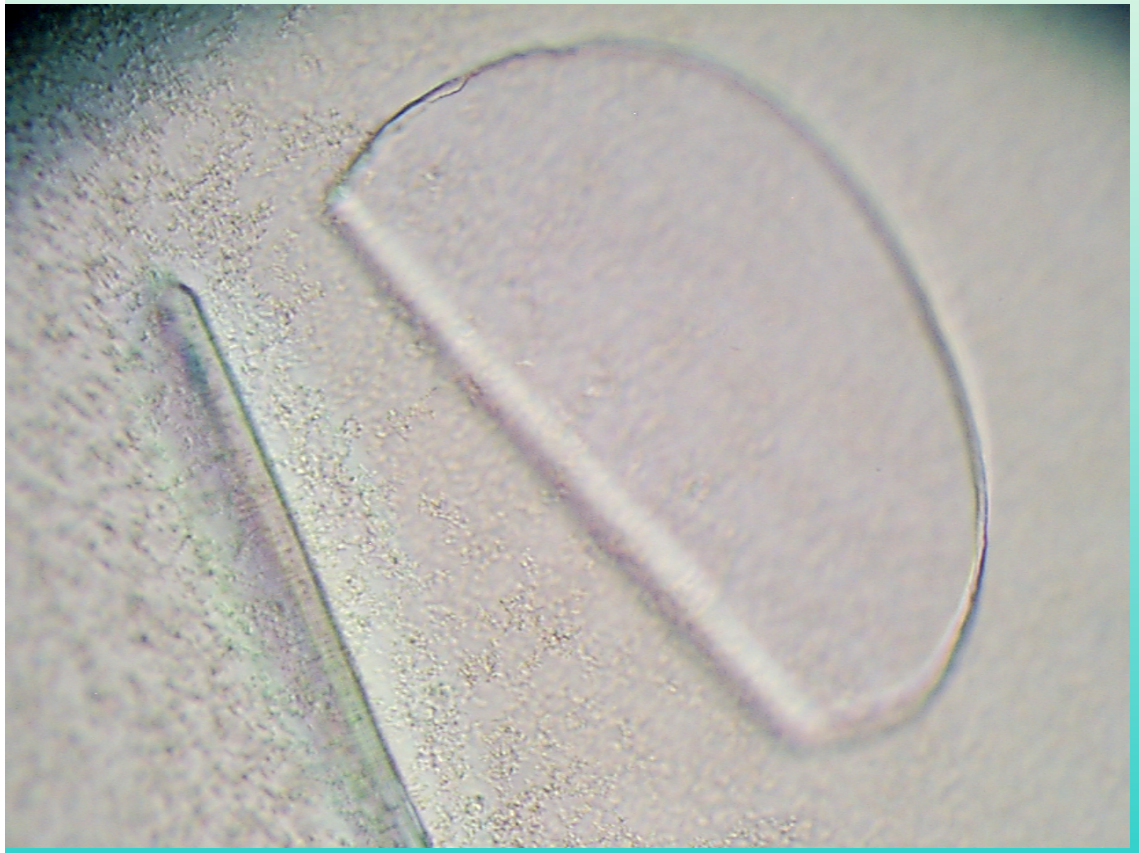


Salt, alcohol,
poly ethylene glycol…

Reservoir



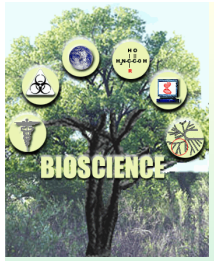
A brief introduction to X-ray crystallography

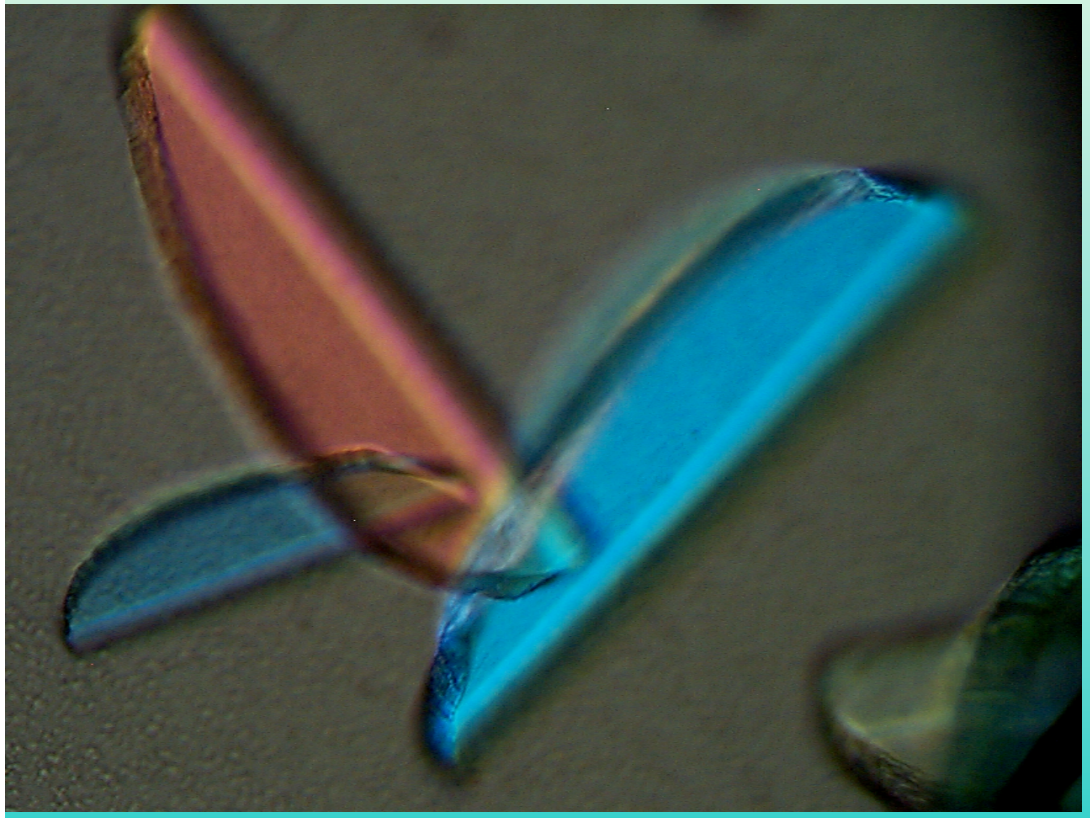


Growing protein crystals

Looking at crystals with X-rays

Getting pictures of proteins from diffraction spots



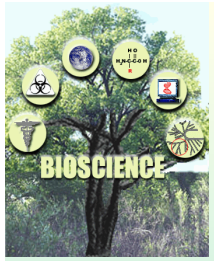


A brief introduction to X-ray crystallography

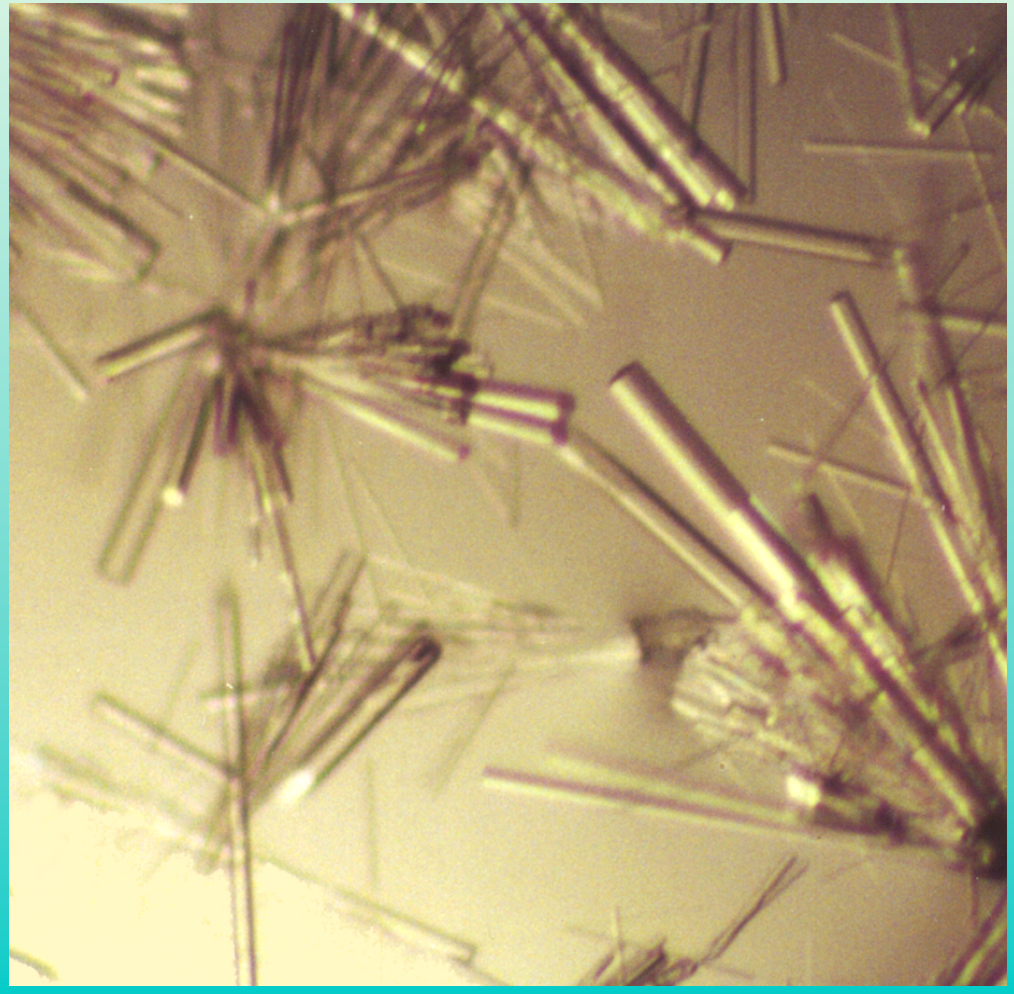
Growing protein crystals

Looking at crystals with X-rays

Getting pictures of proteins from diffraction spots



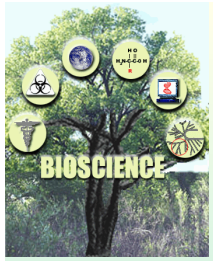
A brief introduction to X-ray crystallography



Growing protein crystals

Looking at crystals with X-rays

Getting pictures of proteins from diffraction spots



A brief introduction to X-ray crystallography

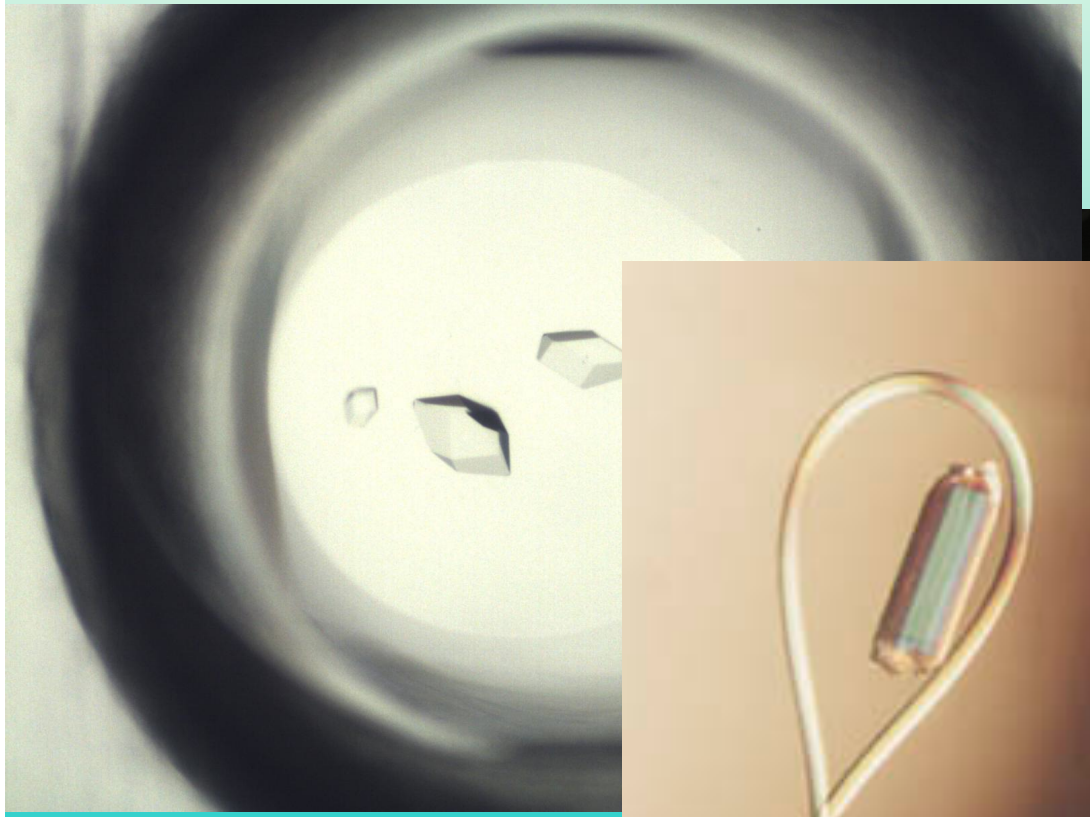
1.5 mm

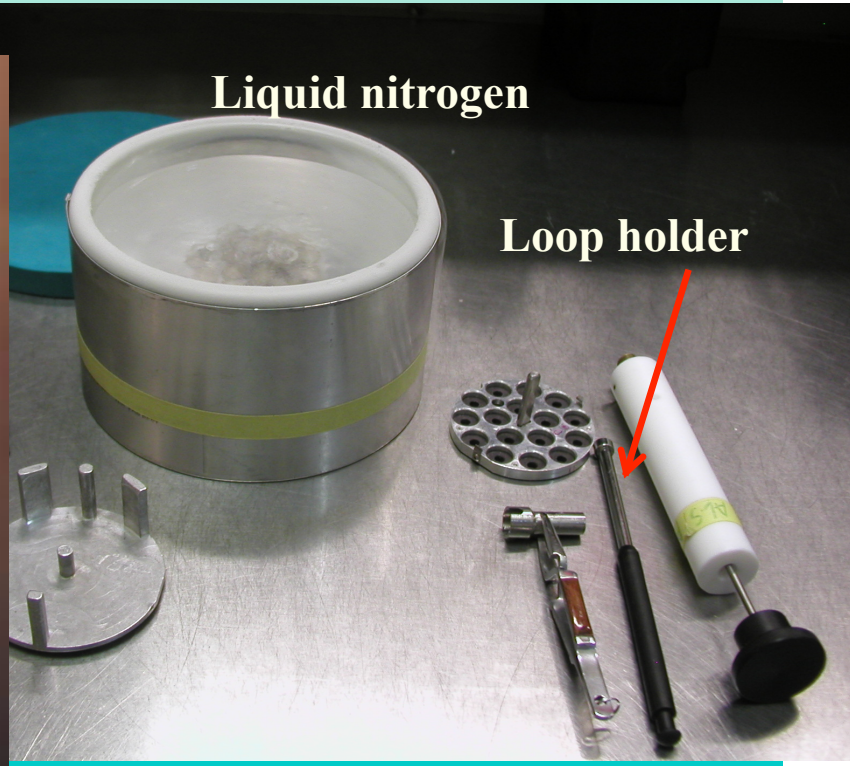
Growing protein crystals

Looking at crystals with X-rays

Getting pictures of proteins from diffraction spots

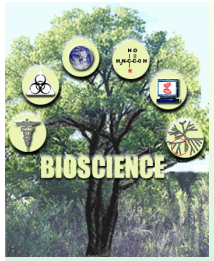
Mounting crystals in nylon loops and cryo-cooling them



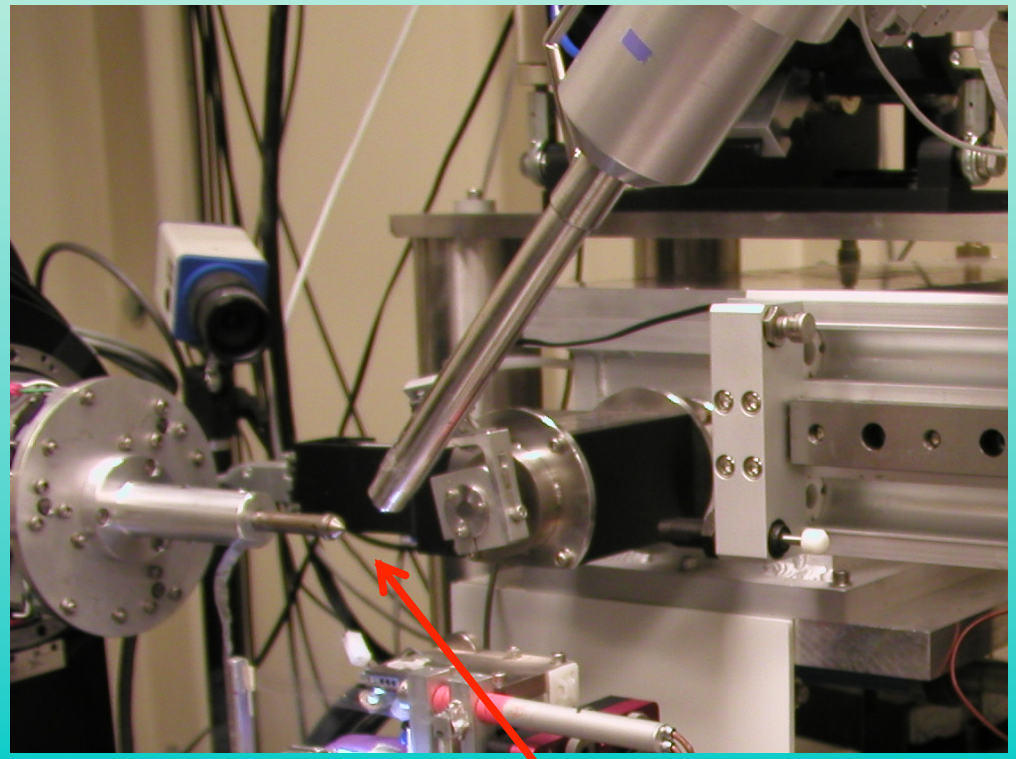


Los Alamos
Bioscience Division

Innovation for Health and Security



Advanced Light Source, Berkeley, CA

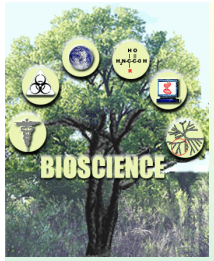


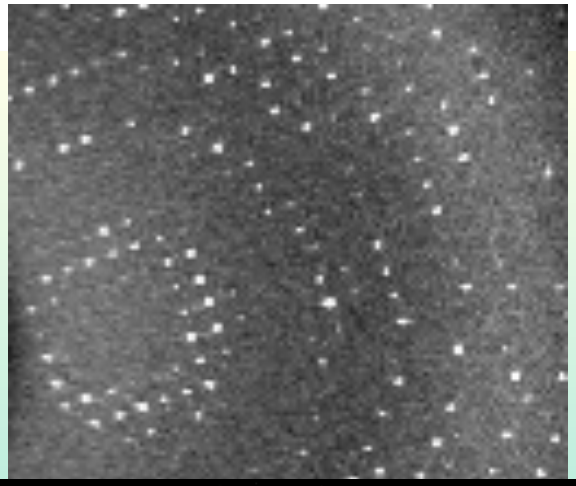
A brief introduction to X-ray crystallography

Growing protein crystals

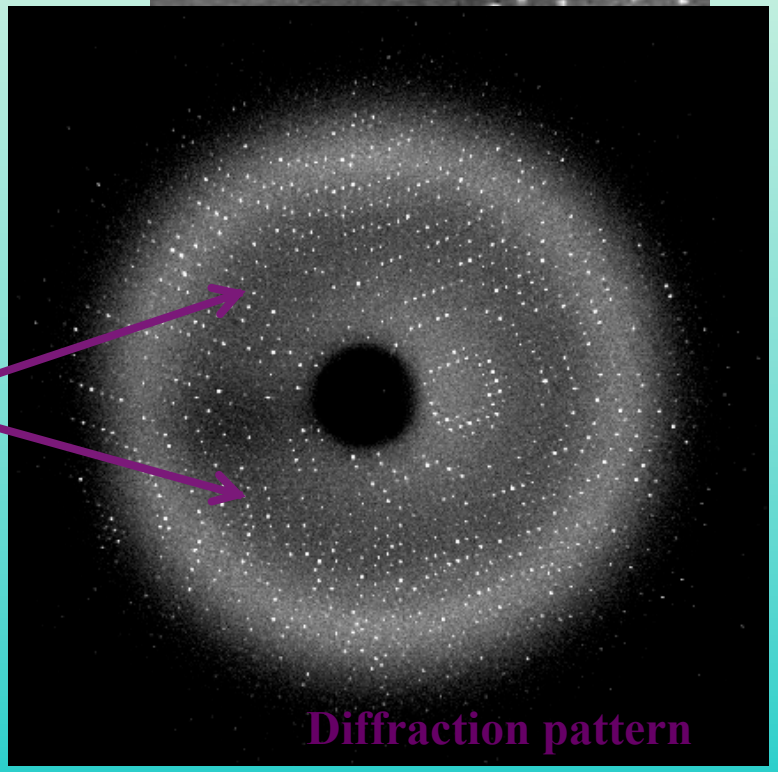
Looking at crystals with X-rays

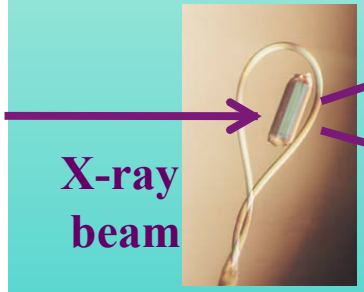
Getting pictures of proteins from diffraction spots





A brief introduction to X-ray crystallography



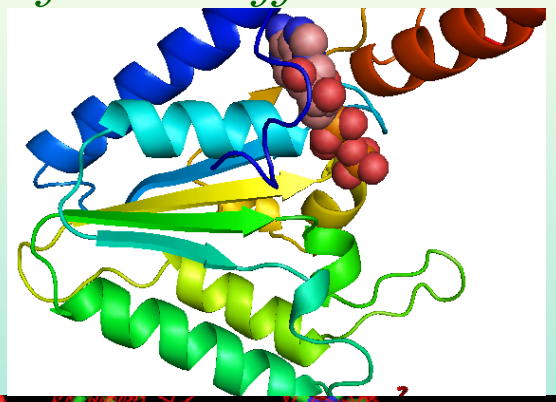


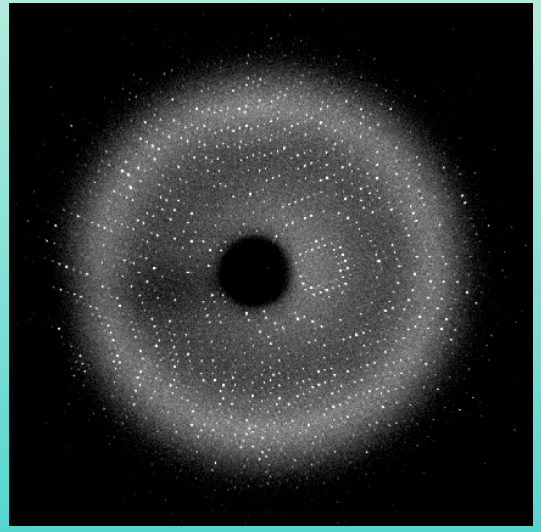
Growing protein crystals

Looking at crystals with X-rays

Getting pictures of proteins from diffraction spots

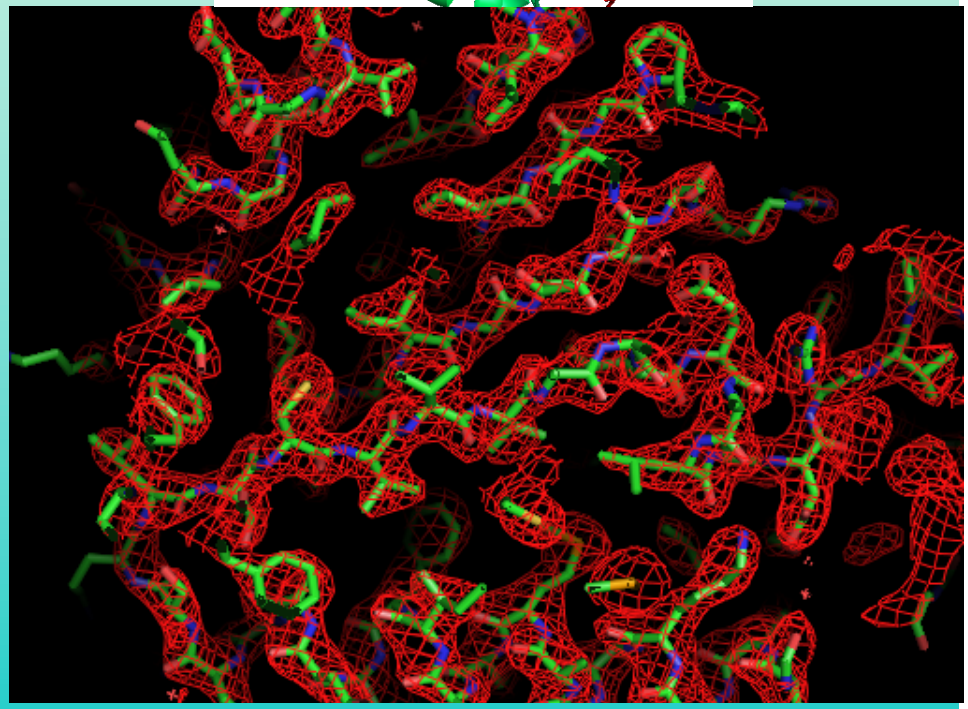
Getting pictures of proteins from diffraction spots





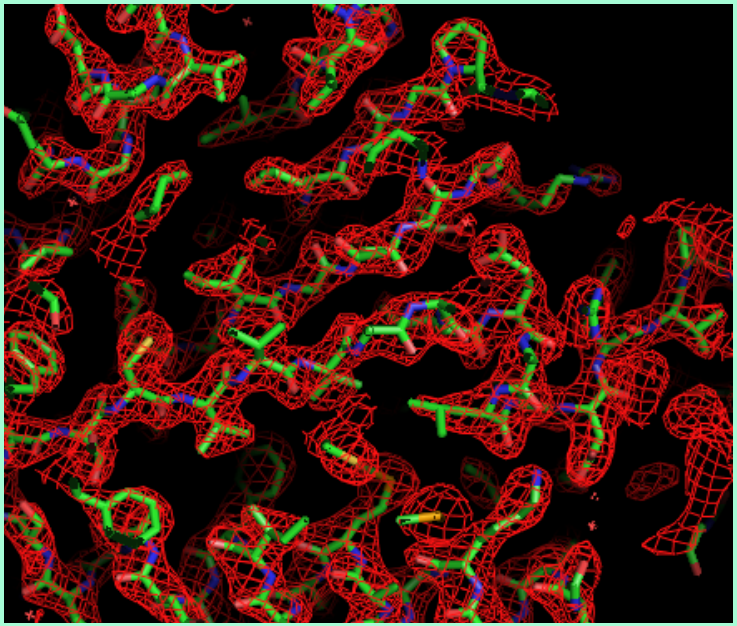
Diffraction pattern

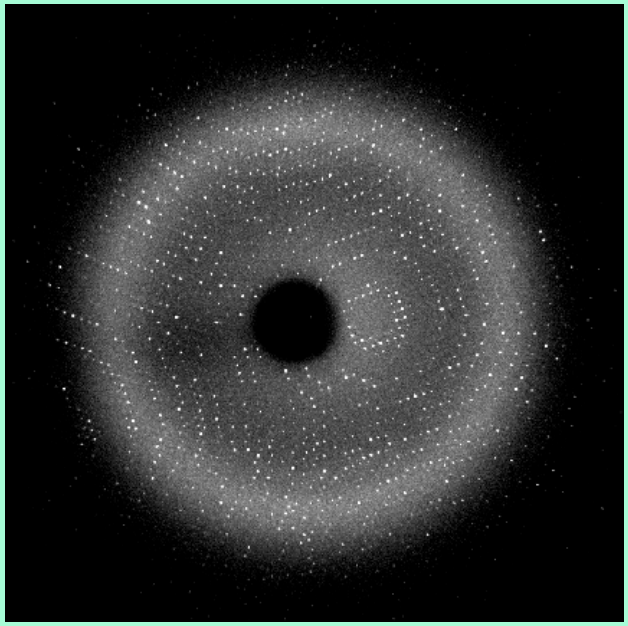
Analysis of diffraction spots

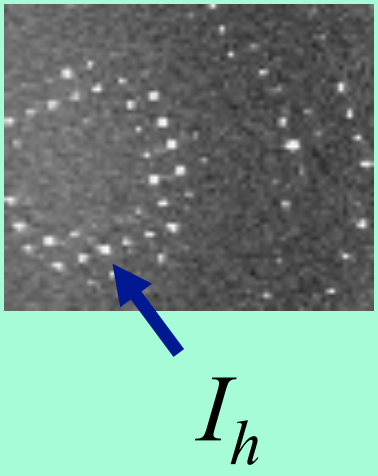


Picture and model of macromolecule

The intensities of X-ray diffraction spots depend on what is in the crystal





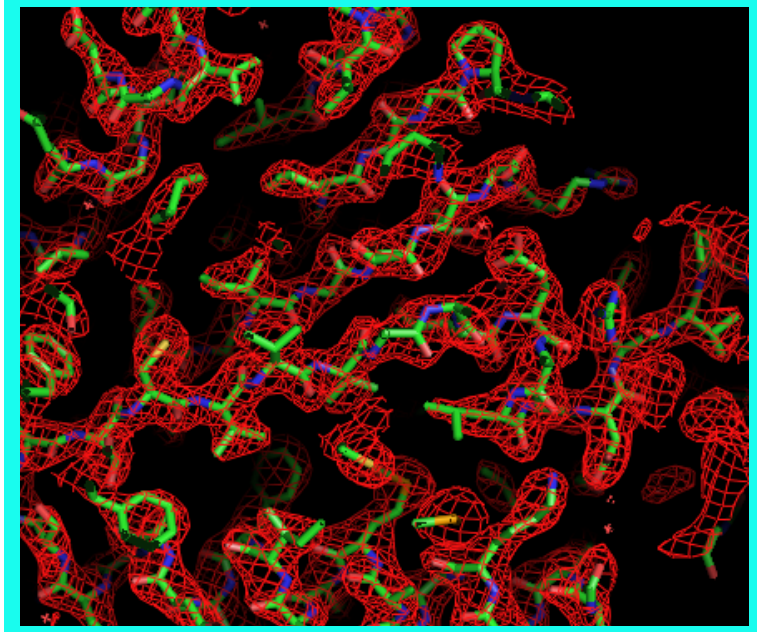


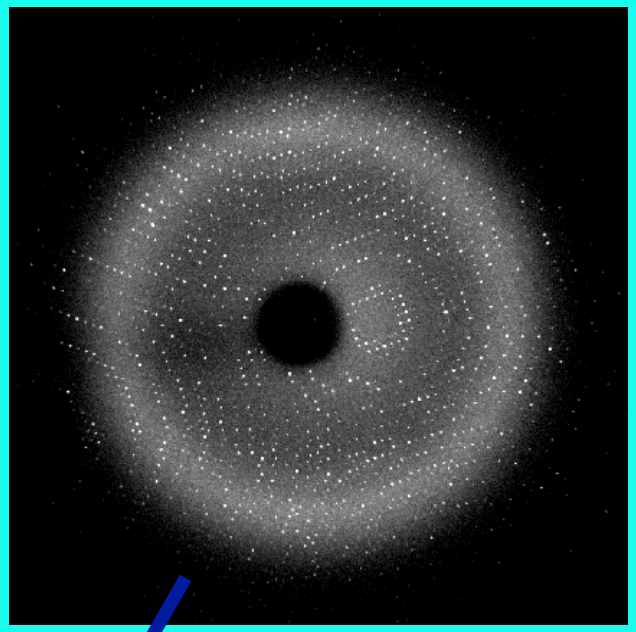
Electrons in a protein crystal (ρ is high where there are many electrons)

Diffraction pattern (I_h is intensity of spot “h”)

$$\rho(x) = \sum_h F_h e^{i\phi_h} e^{-2\pi i h x} \qquad F_h e^{i\phi_h} = \int \rho(x) e^{2\pi i h x} dx \qquad I_h = F_h^2$$

We can almost calculate a picture of where the atoms are from the diffraction pattern (but are missing the phases of diffraction spots)





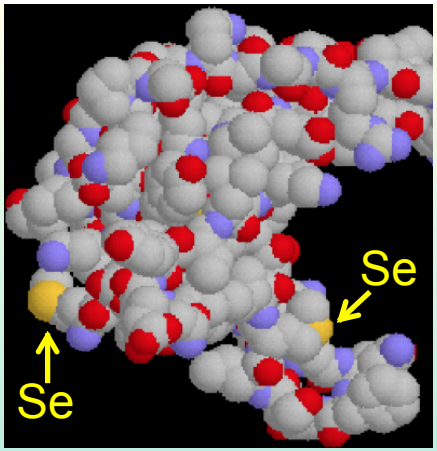
$\rho(x)$

(Where the atoms are)

F_h is square root of measured intensity of spot *h*

$$\rho(x) = \sum_h F_h e^{i\phi_h} e^{-2\pi i h x}$$

We do not know the phase (ϕ_h)



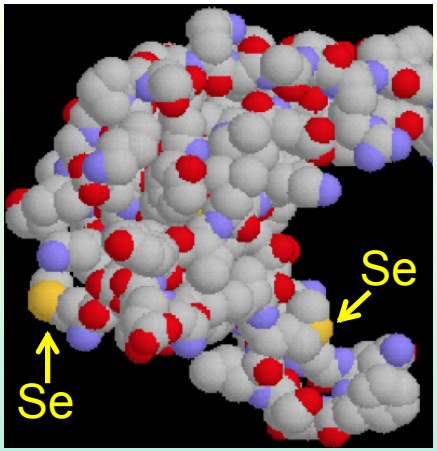
Estimating crystallographic phases: example with multiwavelength X-ray data

- Measure diffraction (I_h , I'_h) at two X-ray wavelengths near absorption edge of selenium

- Differences in diffraction are due to changes in scattering from the Se atoms (ΔF_h)

First figure out where the Se atoms are located

Then use the Se atoms and the diffraction intensities to draw a picture of all the atoms

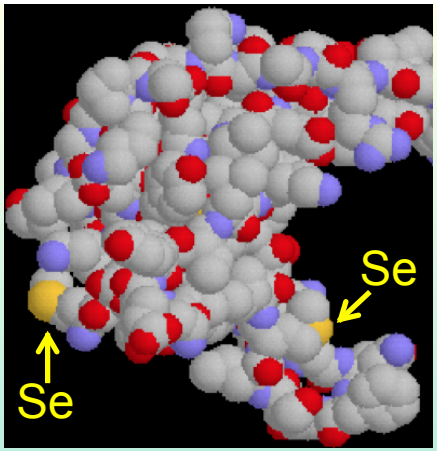


Estimating crystallographic phases with multiwavelength X-ray data

- Measure diffraction (I_h , I'_h) at two X-ray wavelengths near absorption edge of selenium

- Differences in diffraction are due to changes in scattering from the Se atoms ($\Delta\mathbf{F_h}$)

Wavelength	*Scattering density*	*Structure Factor*	*Intensity of diffraction spot*
λ_1	$\rho(x)$	$\mathbf{F_h} = F_h e^{i\phi_h} = \int \rho(x) e^{2\pi i h x} dx$	$I_h = \lvert\mathbf{F_h}\rvert^2$

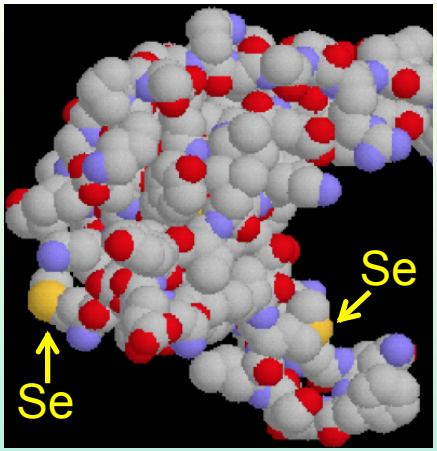


Estimating crystallographic phases with multiwavelength X-ray data

- Measure diffraction (I_h , I'_h) at two X-ray wavelengths near absorption edge of selenium

- Differences in diffraction are due to changes in scattering from the Se atoms ($\Delta\mathbf{F_h}$)

Wavelength	*Scattering density*	*Structure Factor*	*Intensity of diffraction spot*
λ_1	$\rho(x)$	$\mathbf{F_h} = F_h e^{i\phi_h} = \int \rho(x) e^{2\pi i h x} dx$	$I_h = \lvert\mathbf{F_h}\rvert^2$
λ_2	$\rho'(x) = \rho(x) + \Delta\rho(x)$	$\mathbf{F'_h} = \mathbf{F_h} + \Delta\mathbf{F_h}$	$I'_h = \lvert\mathbf{F_h} + \Delta\mathbf{F_h}\rvert^2$



Estimating crystallographic phases with multiwavelength X-ray data

- Measure diffraction (I_h , I'_h) at two X-ray wavelengths near absorption edge of selenium

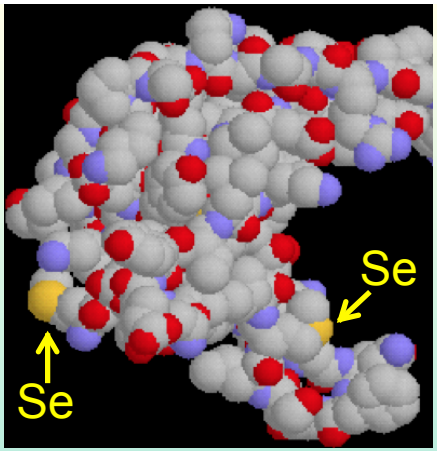
- Differences in diffraction are due to changes in scattering from the Se atoms ($\Delta\mathbf{F_h}$)

How to figure out where the Se atoms are:

Assume that structure factors for Se are similar to changes between wavelengths:

$$|\Delta\mathbf{F_h}| \approx |F'_h - F_h|$$

Then use techniques from small-molecule crystallography to find the Se atoms (guess locations, compare calculated and observed $\Delta\mathbf{F_h}$, update guess)



Estimating crystallographic phases with multiwavelength X-ray data

- Measure diffraction (I_h , I'_h) at two X-ray wavelengths near absorption edge of selenium

- Differences in diffraction are due to changes in scattering from the Se atoms ($\Delta\mathbf{F_h}$)

Wavelength	*Scattering density*	*Structure Factor*	*Intensity of diffraction spot*
λ_1	$\rho(x)$	$\mathbf{F_h} = F_h e^{i\phi_h} = \int \rho(x) e^{2\pi i h x} dx$	$I_h = \lvert\mathbf{F_h}\rvert^2$
λ_2	$\rho'(x) = \rho(x) + \Delta\rho(x)$	$\mathbf{F'_h} = \mathbf{F_h} + \Delta\mathbf{F_h}$	$I'_h = \lvert\mathbf{F_h} + \Delta\mathbf{F_h}\rvert^2$

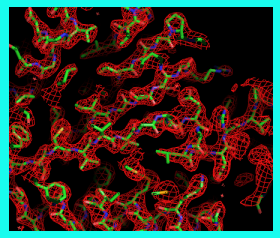
If we know where the Se atoms are ...

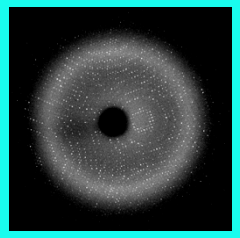
we know: $\Delta\rho(x)$

...so we can calculate: $\Delta\mathbf{F_h}$

and the phase (ϕ_h) must satisfy: $I'_h = \lvert I_h^{1/2} e^{i\phi_h} + \Delta\mathbf{F_h}\rvert^2$

Many ways to find the phases





Method	Source of phasing information
SIR – single isomorphous replacement	A few heavy atoms (e.g., Hg, Au) in "derivative" contribute to differences from "native"
SAD – single-wavelength anomalous diffraction	A few atoms (e.g., Se, I, Hg atoms) contribute to "anomalous" differences in diffraction between spot *h* and spot *-h*
MAD – multiple-wavelength anomalous diffraction	A few atoms contribute to anomalous and wavelength-dependent "dispersive" differences
SIRAS, MIR	Combinations of SIR and SAD
Molecular replacement	Molecular location and phases are found using a related molecule as a template
Direct methods	Guess where atoms are, good guesses match the measured structure factors

Automation of structure determination

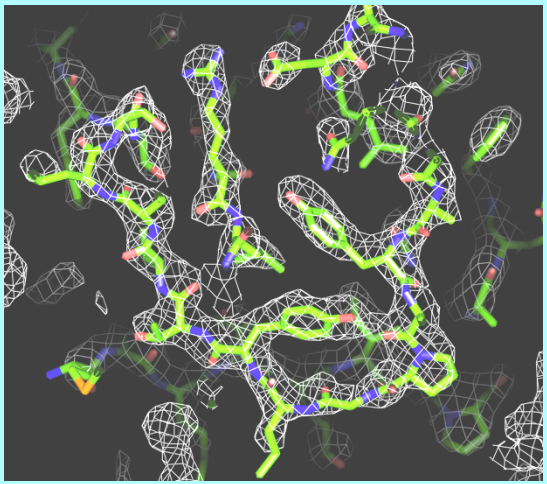
Automation…

makes straightforward cases accessible to a wider group of structural biologists

makes difficult cases more feasible for experts

can speed up the process

can help reduce errors



Automation also allows you to…

try more possibilities

estimate uncertainties

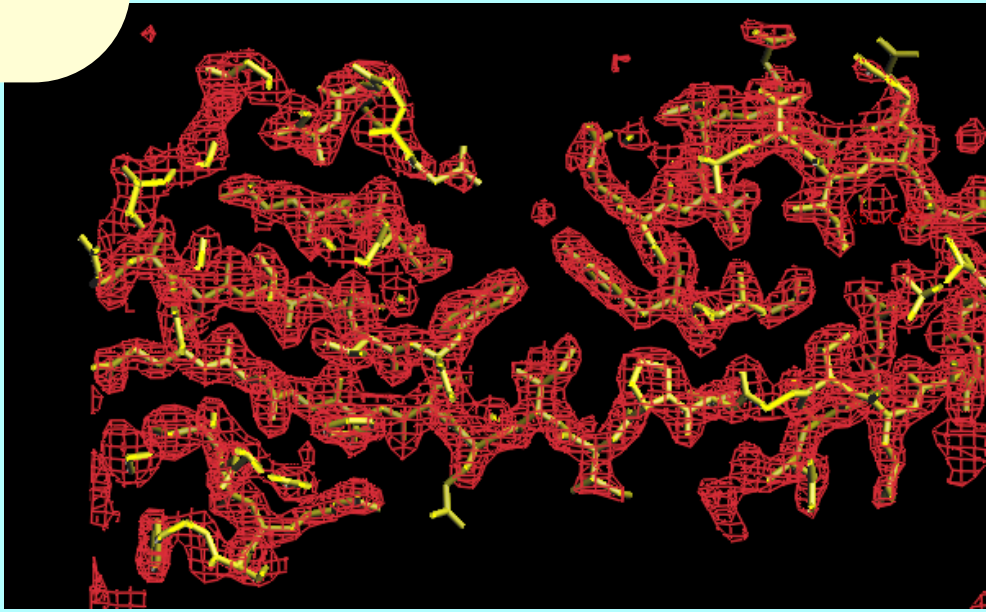
Requirements for automation of structure determination of macromolecules by X-ray crystallography

(1) Software carrying out individual steps

(2) Seamless connections between steps

(3) A way to decide what is good

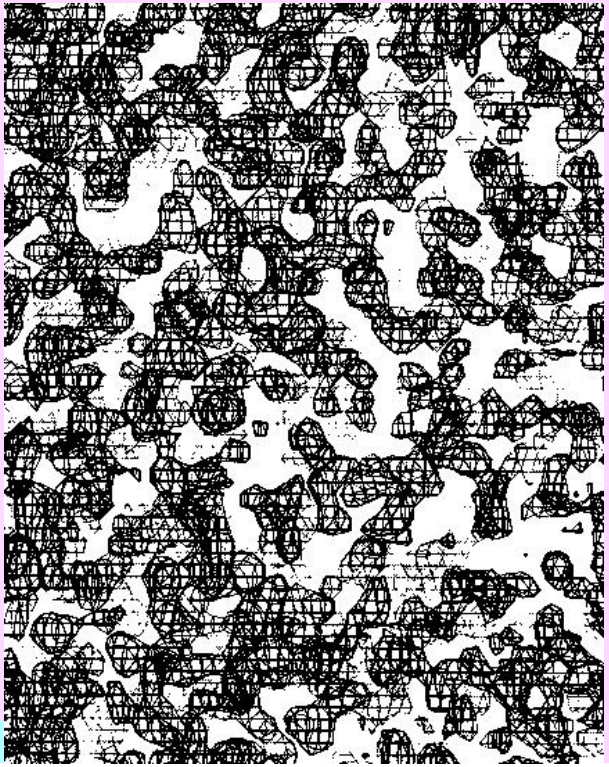
(4) Strategies for structure determination and decision-making



Why we need good measures of the quality of an electron-density map:

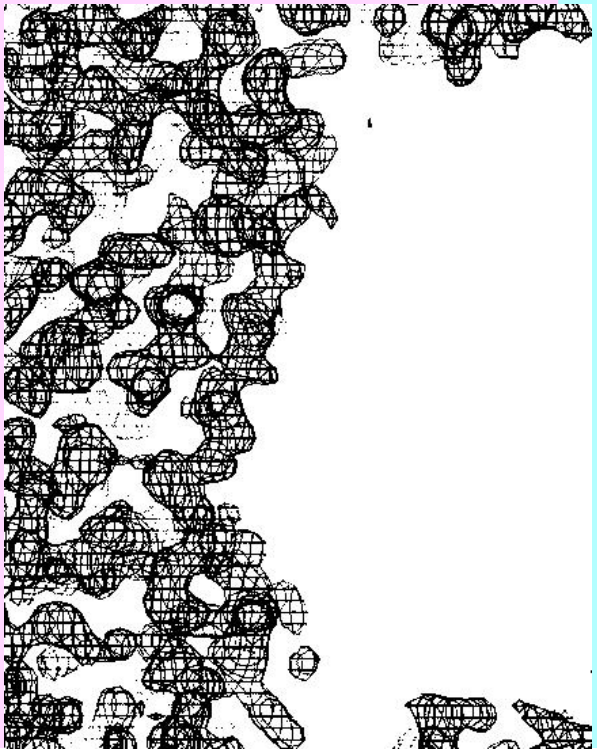
Which solution is best?

Are we on the right track?



If map is good:

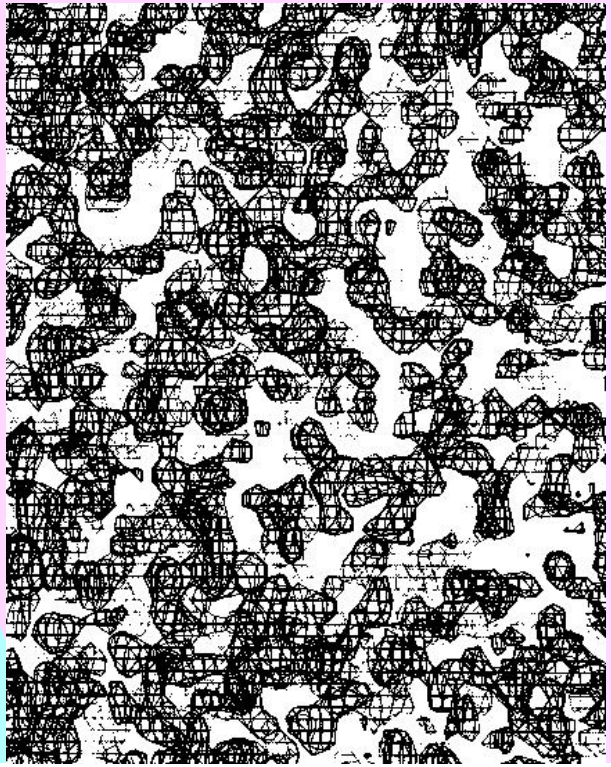
It is easy



Why we need good measures of the quality of an electron-density map:

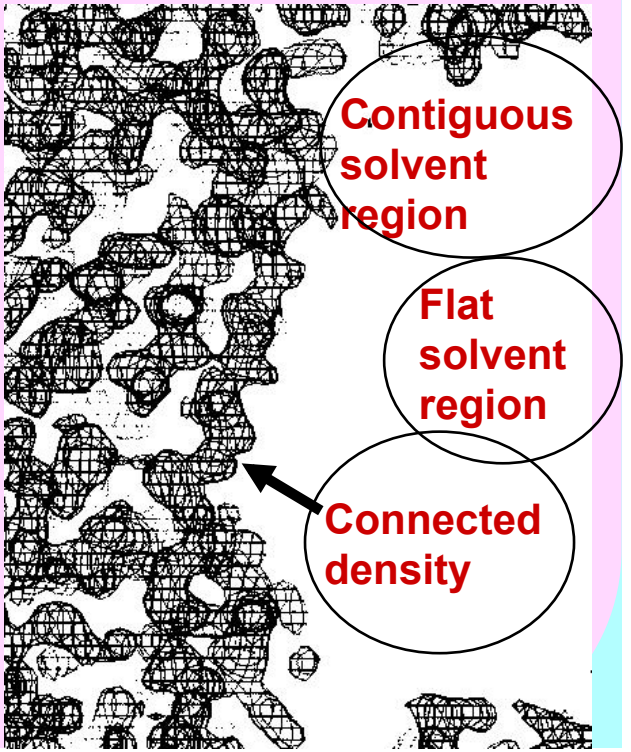
Which solution is best?

Are we on the right track?

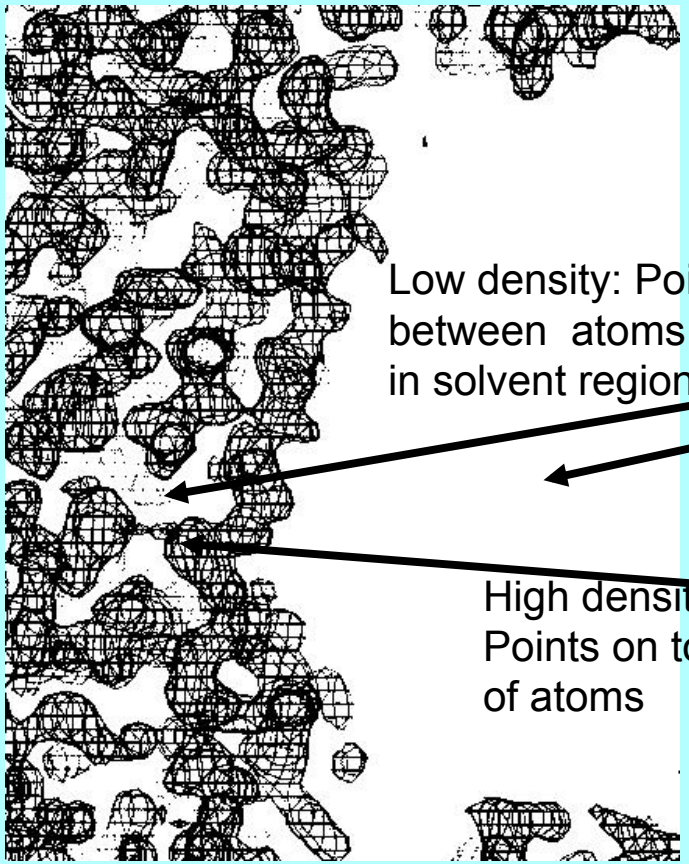


If map is good:

It is easy

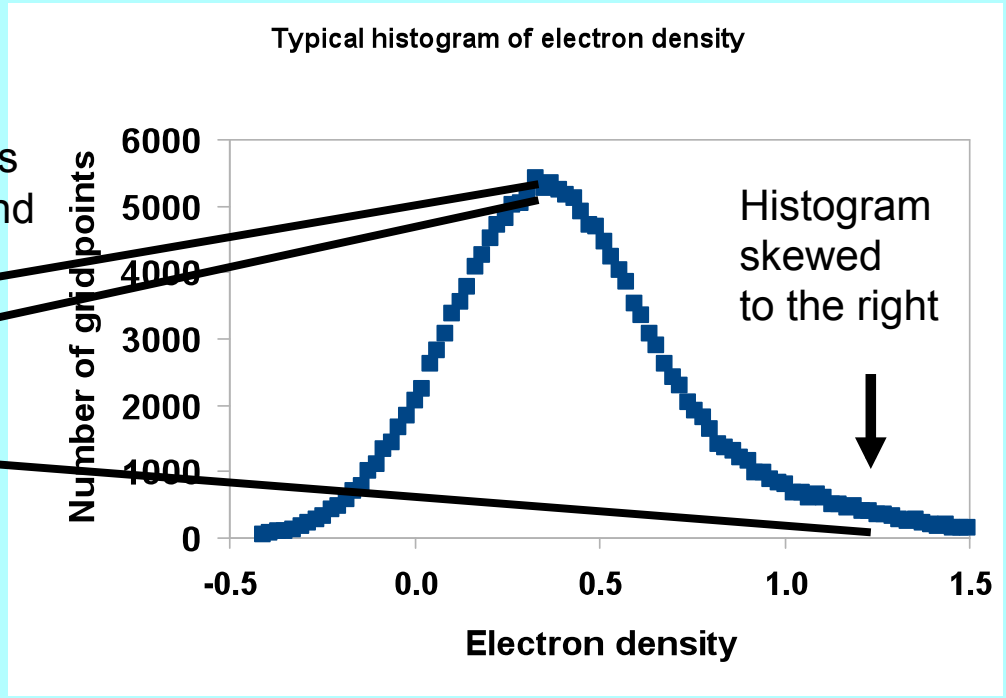


Histogram of electron density values has a positive “skew”

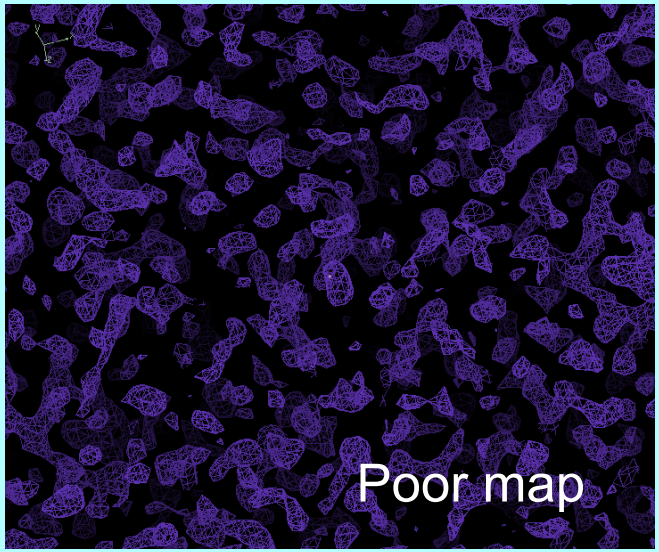


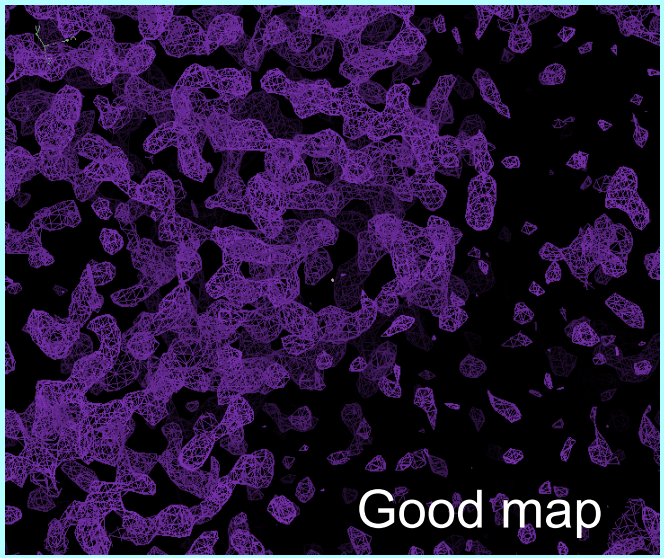
Low density: Points between atoms and in solvent region

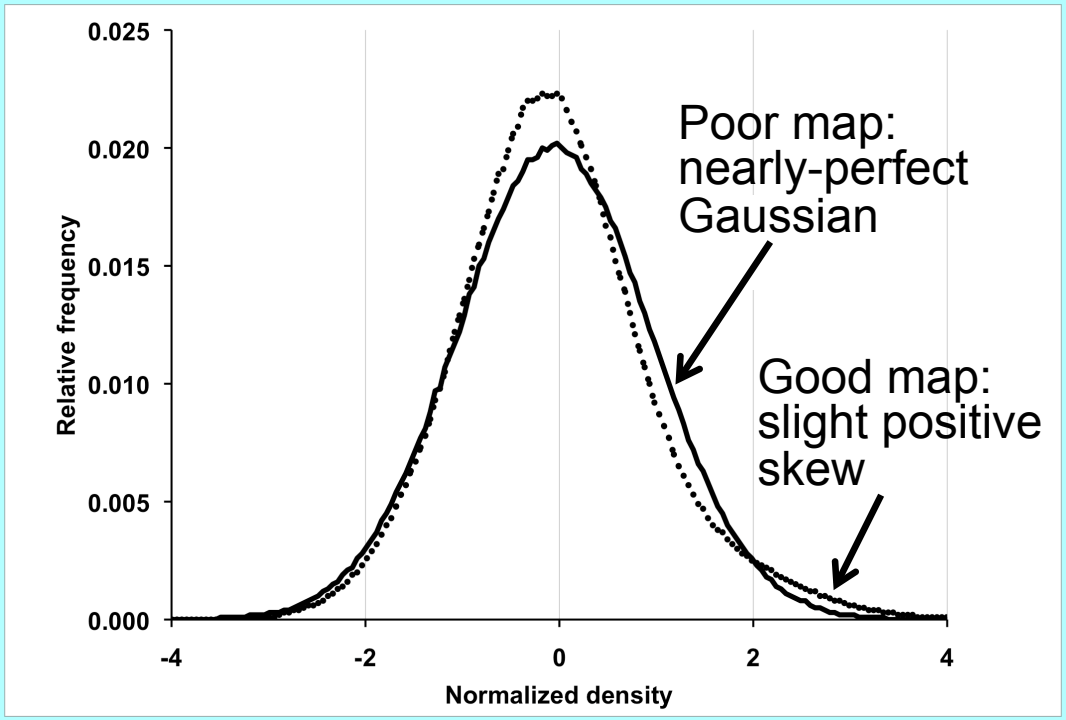
High density: Points on top of atoms



Skew of electron density for poor and good maps







Evaluating electron density maps

Basis	*Good map*	*Random map*
Skew of density (Podjarny, 1977)	Highly skewed (very positive at positions of atoms, zero elsewhere)	Gaussian histogram
Connectivity of regions of high density (Baker, Krukowski, & Agard, 1993)	A few connected regions can trace entire molecule	Many very short connected regions
Correlation of local rms densities (Terwilliger, 1999)	Neighboring regions in map have similar rms densities	Map has uniform rms density
R-factor in 1st cycle of density modification (Cowtan, 1996)	Low R-factor	High R-factor

Which scoring criteria best reflect the quality of a map?

Create real maps

Score the maps with each criteria

Compare the scores with the actual quality of the maps

Creating real maps

247 MAD, SAD, MIR datasets with final model available
(PHENIX library and JCSG publicly-available data)

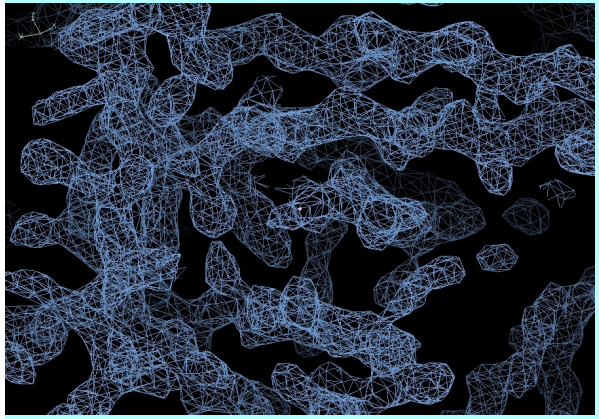
Run AutoSol Wizard on each dataset.

Calculate maps for each solution considered
(opposing hands, additional sites, including various derivatives for MIR)

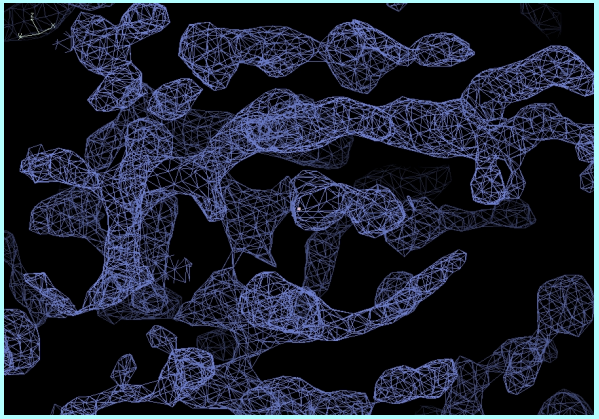
Score maps based on each criteria

Calculate map correlation coefficient (CC) to model map
(no density modification, shift origin if necessary)

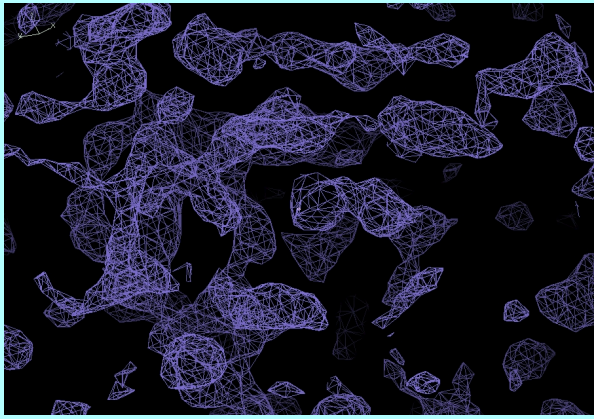
Model map
1VQB, 2.6 Å, SG C2



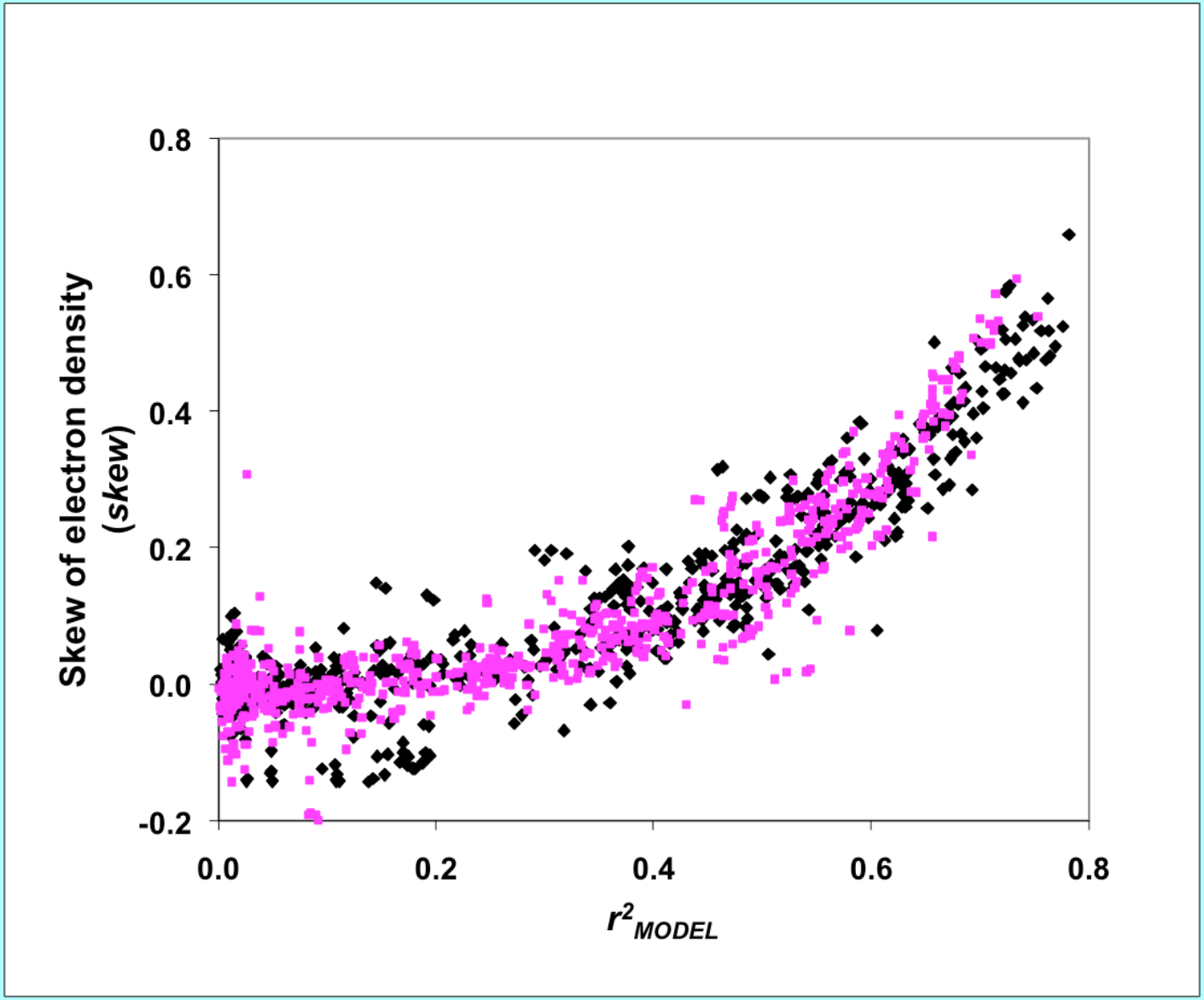
SOLVE MAD map
CC=0.62



Inverse-hand map
CC=0.55

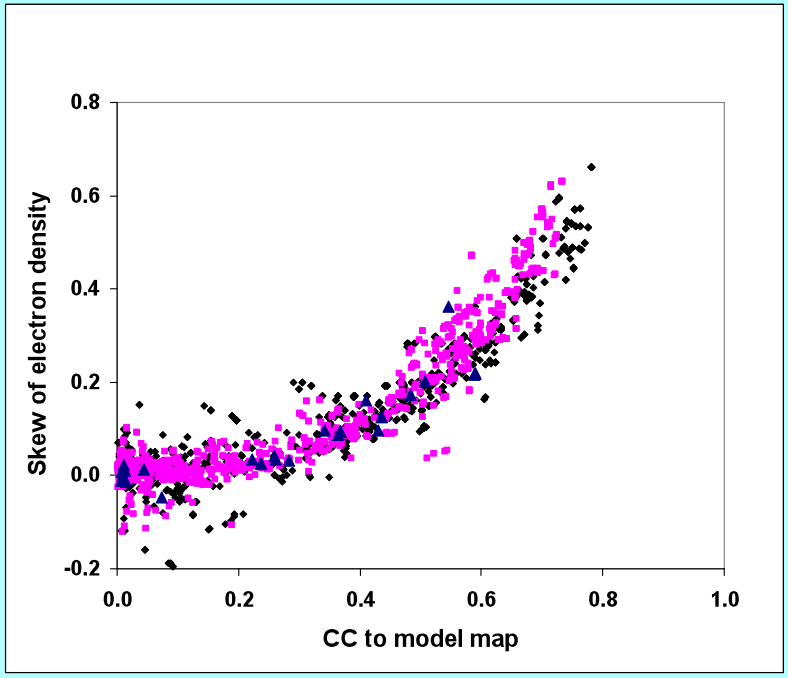


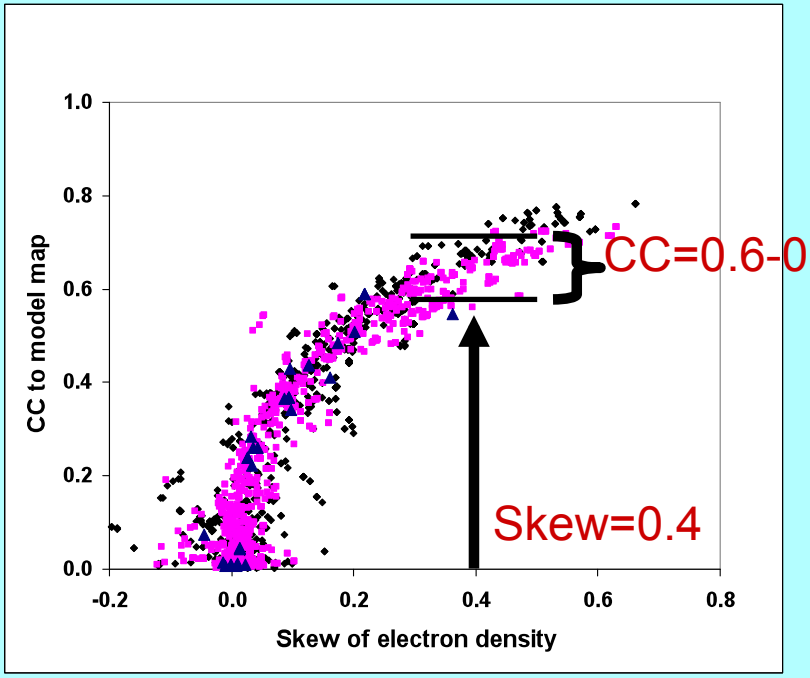
Skew of electron density – positive skew of density values



Using scoring criteria to estimate the quality of a map

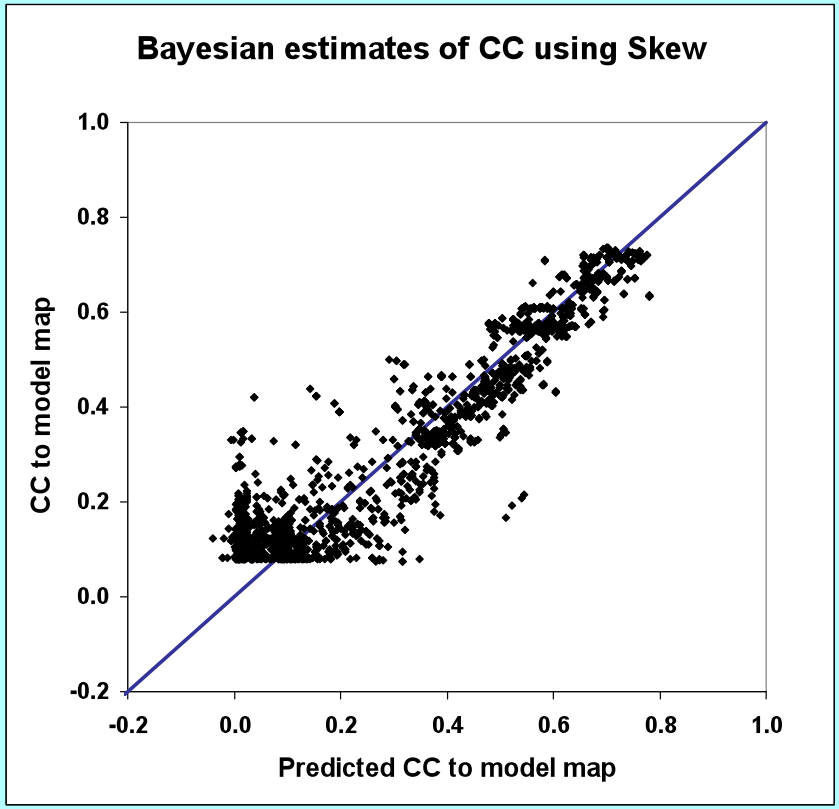
Skew depends on CC → Estimate CC from skew





How accurate are estimates of map quality?

Actual quality



Estimated quality

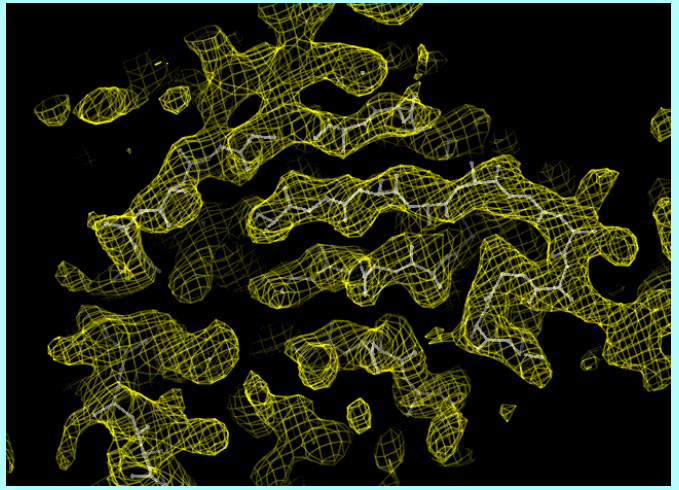
Cross-validated estimates of quality

Estimated map quality in practice

Evaluating solutions to a 2-wavelength MAD experiment
(JCSG Tm3681, 1VPM, SeMet 1.6 Å data)

Data for HYSS	Sites	Estimated CC ± 2SD	Actual CC	
Peak	12	0.73 ± 0.04	0.72	←
Peak (inverse hand)	12	0.11 ± 0.43	0.04	
F_A	12	0.73 ± 0.03	0.72	
F_A (inverse)	12	0.11 ± 0.42	0.04	
Sites from diff Fourier	9	0.70 ± 0.17	0.69	

Statistical density modification (RESOLVE)

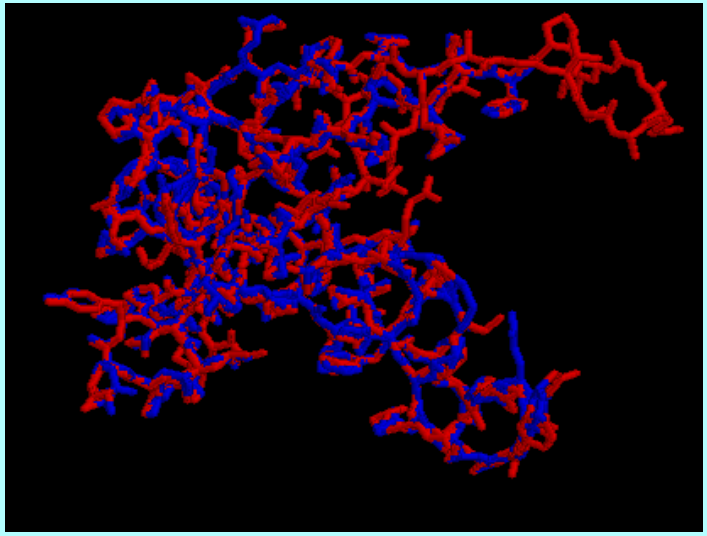


•Principle: phase probability information from probability of the map and from experiment:

•$P(\phi) = P_{\text{map probability}}(\phi)\, P_{\text{experiment}}(\phi)$

•"Phases that lead to a believable map are more probable than those that do not"

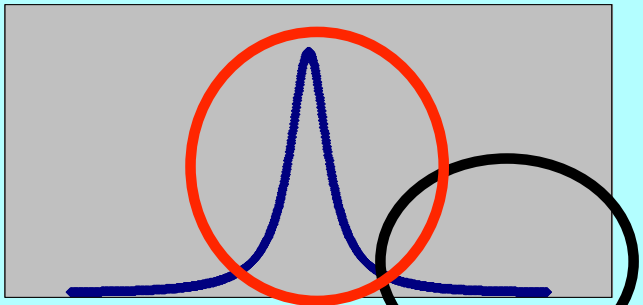
•A believable map is a map that has…
•a relatively flat solvent region
•NCS (if appropriate)
•A distribution of densities like those of model proteins



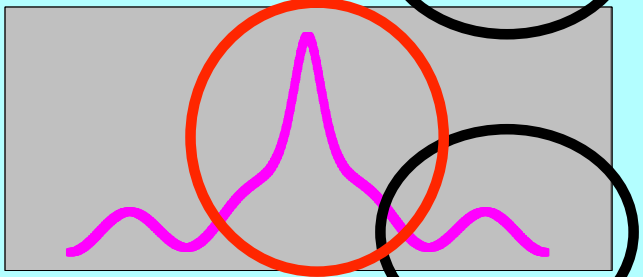
•Method:
•calculate how map probability varies with electron density ρ
•deduce how map probability varies with phase ϕ
•combine with experimental phase information

Map probability phasing: Getting a new probability distribution for each phase given estimates of all others

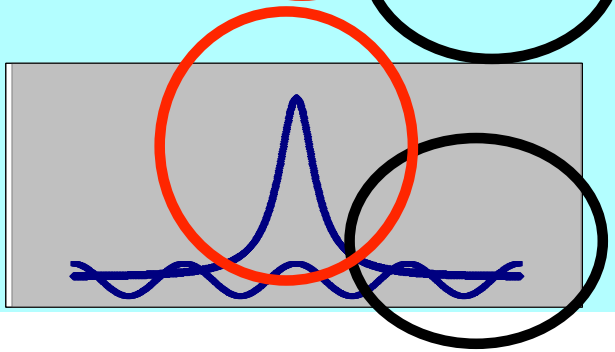
1. Identify expected features of map (flat far from center)
2. Calculate map with current estimates of all structure factors except one (k)
3. Test all possible phases ϕ for structure factor k (for each phase, calculate new map including k)
4. Probability of phase ϕ estimated from agreement of map with expectations
5. Phase probability of reflection k from map is independent of starting phase probability because reflection k is omitted from the map



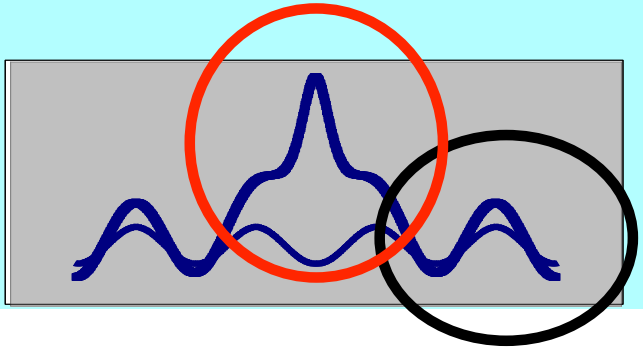
A function that is (relatively) flat far from the origin



Function calculated from estimates of all structure factors but one (k)



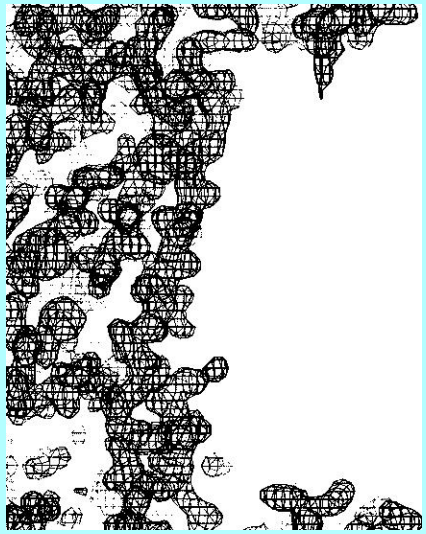
Test each possible phase of structure factor k. P(ϕ) is high for phase that leads to flat region



A map-probability function – allowing different weighting of information from different parts of the map

Log-probability of the map is sum over all points in map of local log-probability

$$LL^{MAP}(\{\mathbf{F_h}\}) \approx \frac{\mathrm{N_{REF}}}{\mathrm{V}} \int_{\mathbf{V}} LL(\rho(\mathbf{x}, \{\mathbf{F_h}\}))\mathrm{d}^3\mathbf{x}$$



A map with a flat (blank) solvent region is a likely map

Local log-probability is believability of the value of electron density (ρ(x)) found at this point

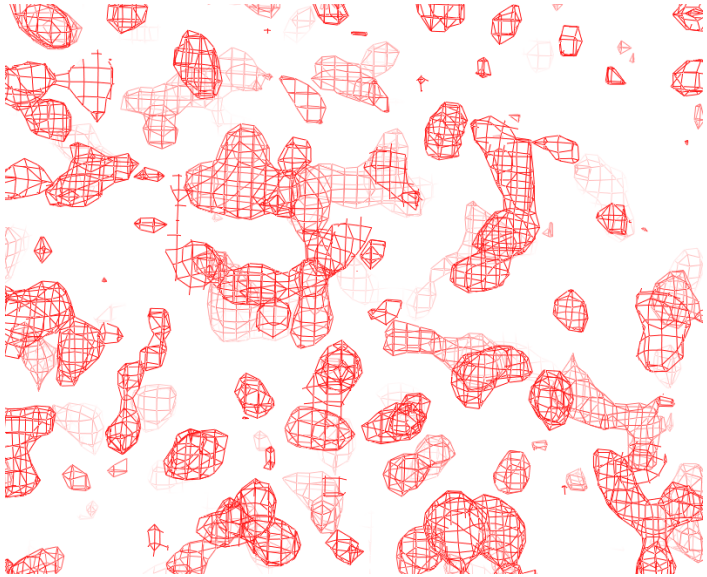
$$LL(\rho(\mathbf{x}, \{\mathbf{F_h}\})) = \ln[p(\rho(\mathbf{x})|PROT)p_{PROT}(\mathbf{x}) + p(\rho(\mathbf{x})|SOLV)p_{SOLV}(\mathbf{x})]$$

If the point is in the PROTEIN region, most values of electron density (ρ(x)) are believable

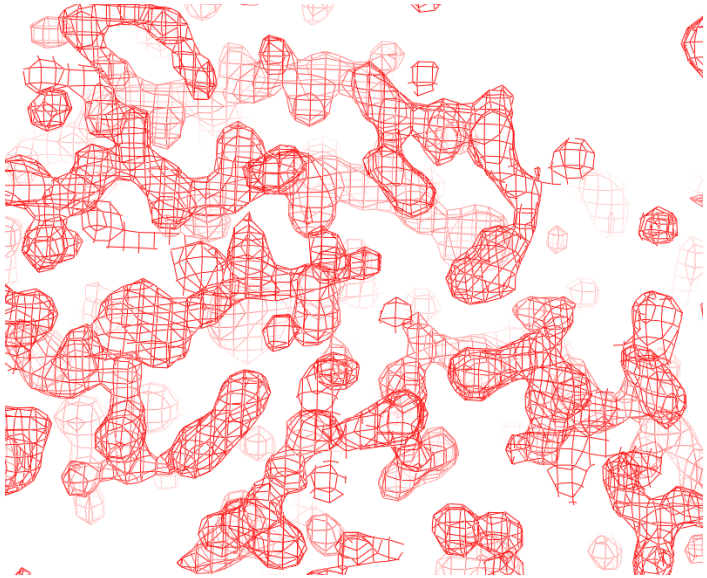
If the point is in the SOLVENT region, only values of electron density near zero are believable

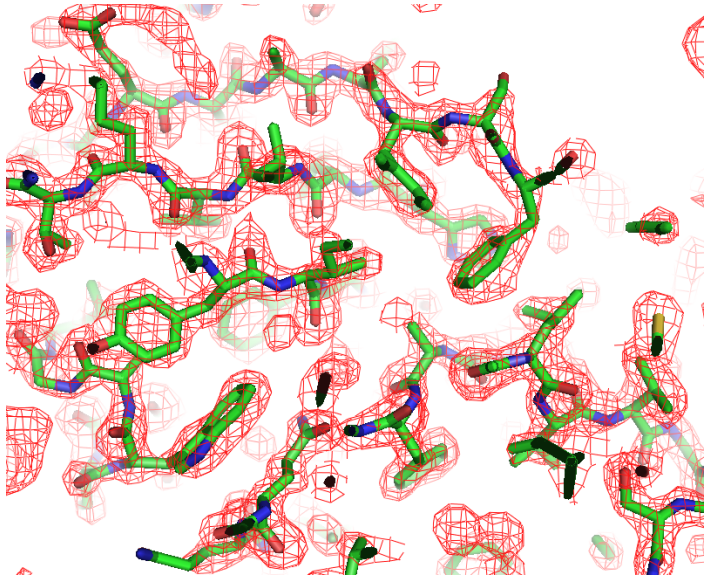
Statistical density modification
(nsf-N SAD map , 2Å, no NCS, 50% solvent)

Phaser SAD map
(CC=0.43)



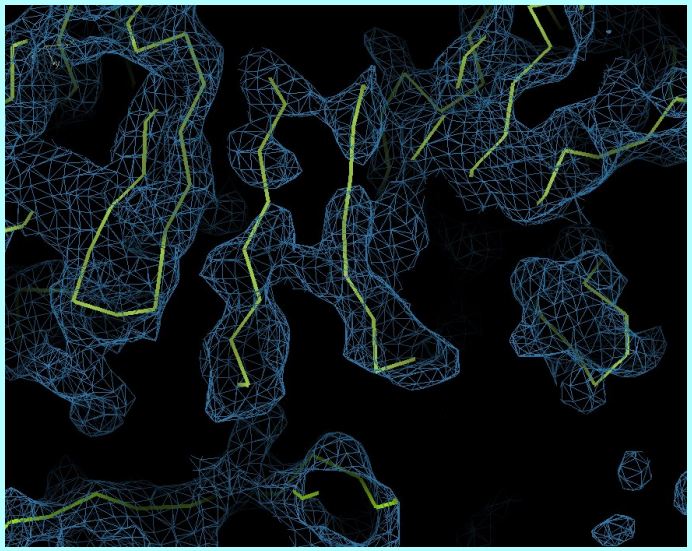
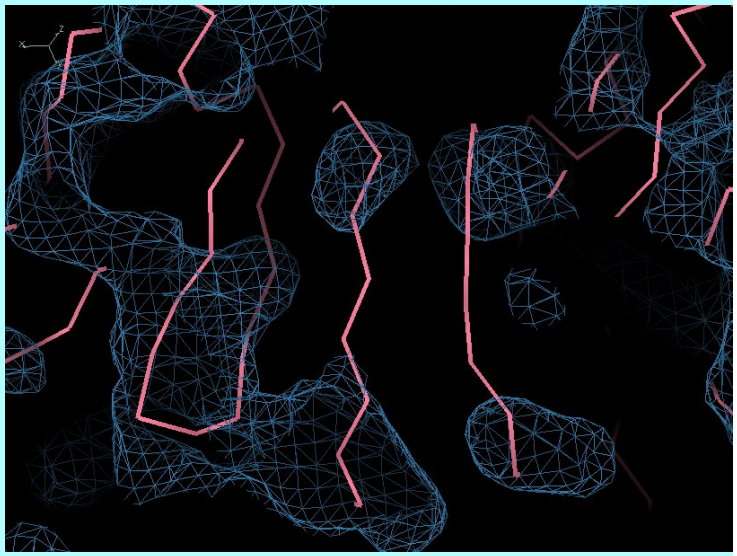
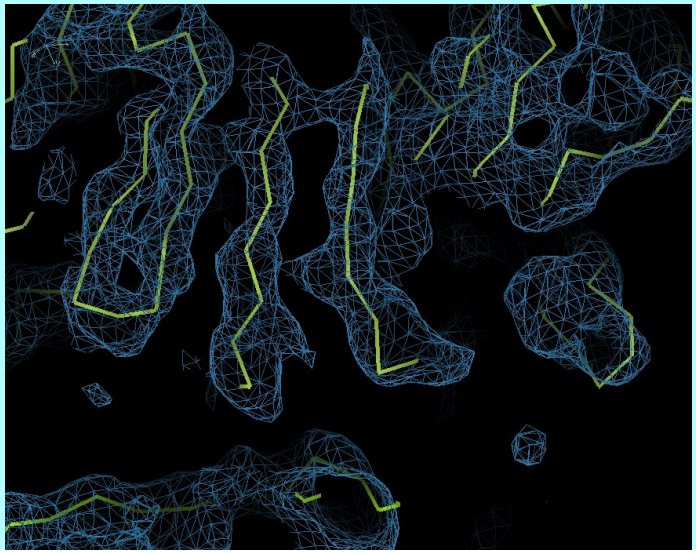
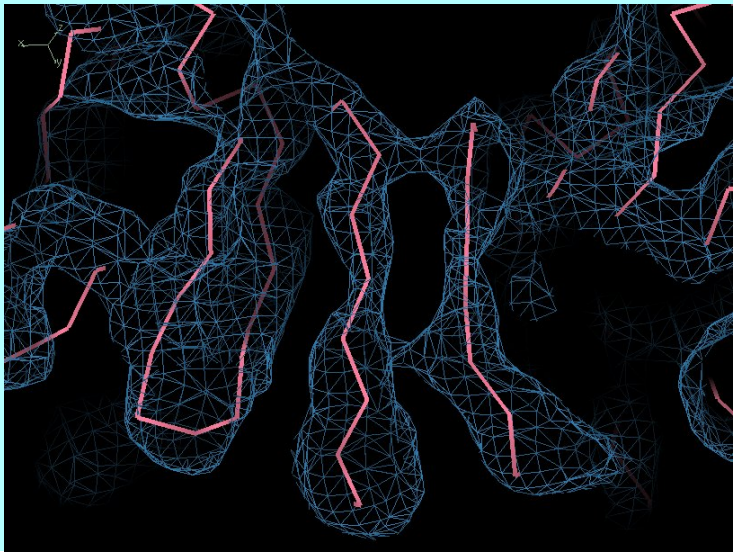
Phaser +RESOLVE
(CC=0.79)



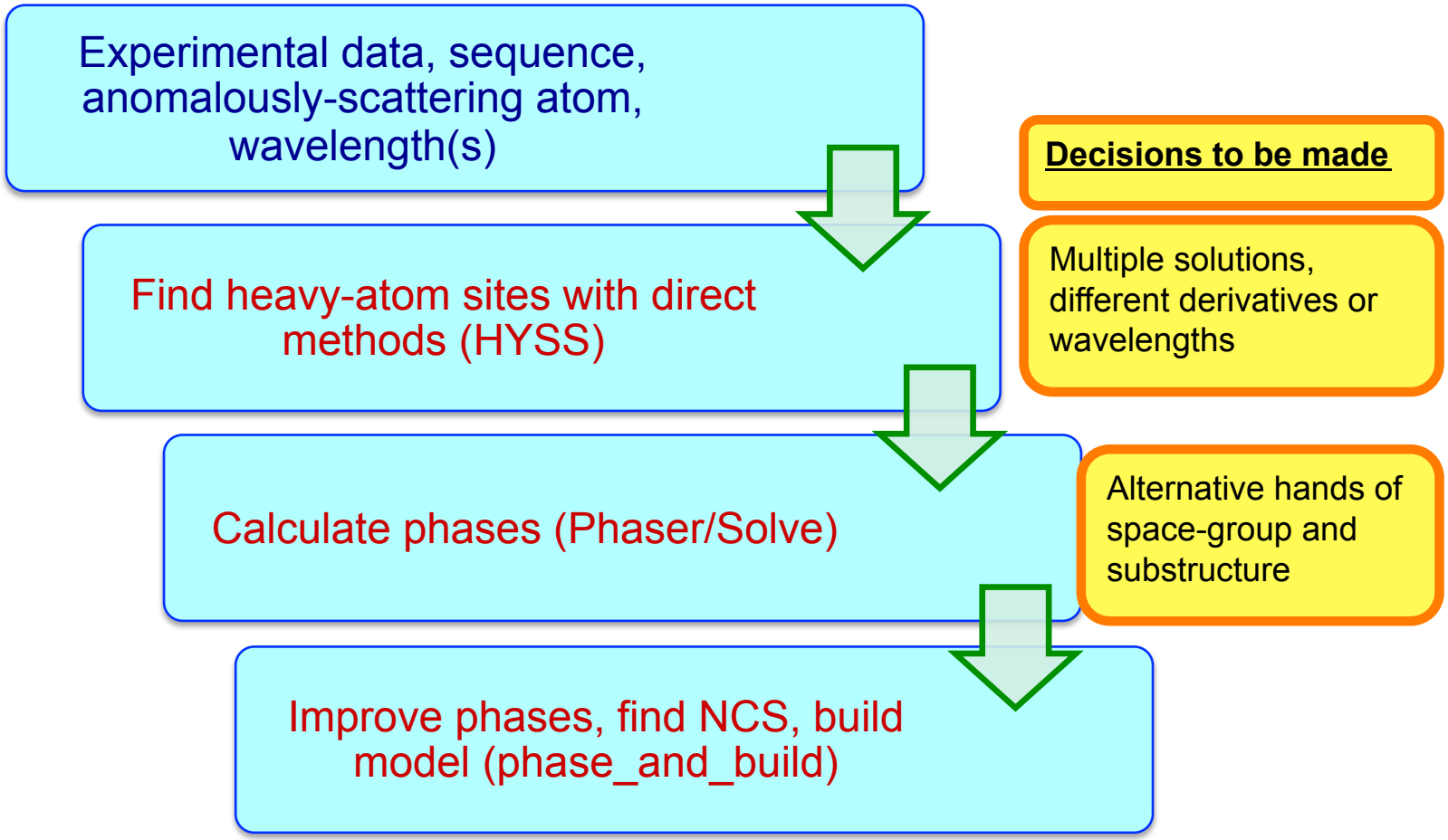


Statistical density modification with cross-crystal averaging

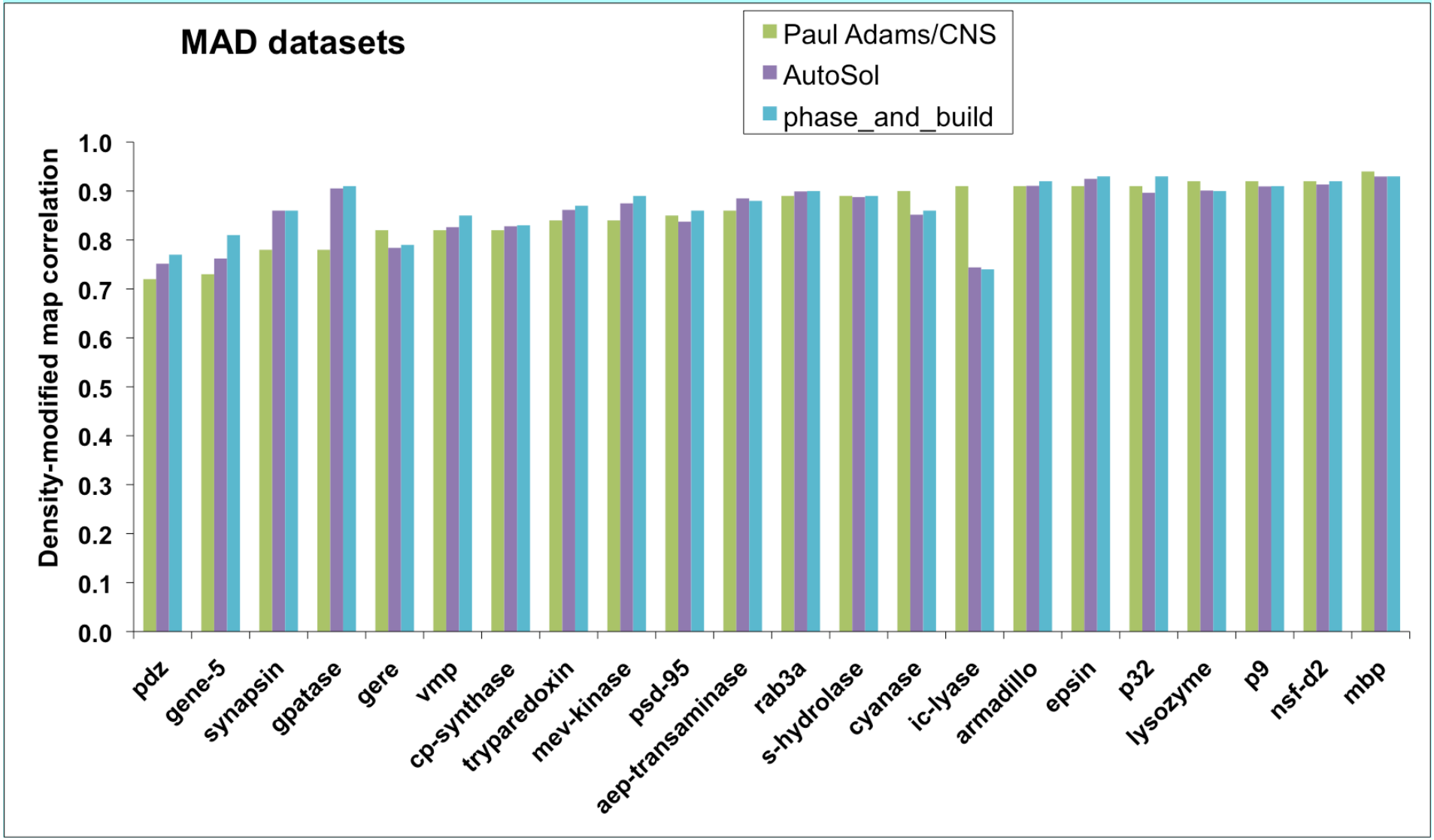
Cell receptor at 3.5/3.7 Å. Data courtesy of J. Zhu

	Crystal 1 (4 copies)	Crystal 2 (2 copies)
RESOLVE density modification		
PHENIX Multi-crystal averaging		

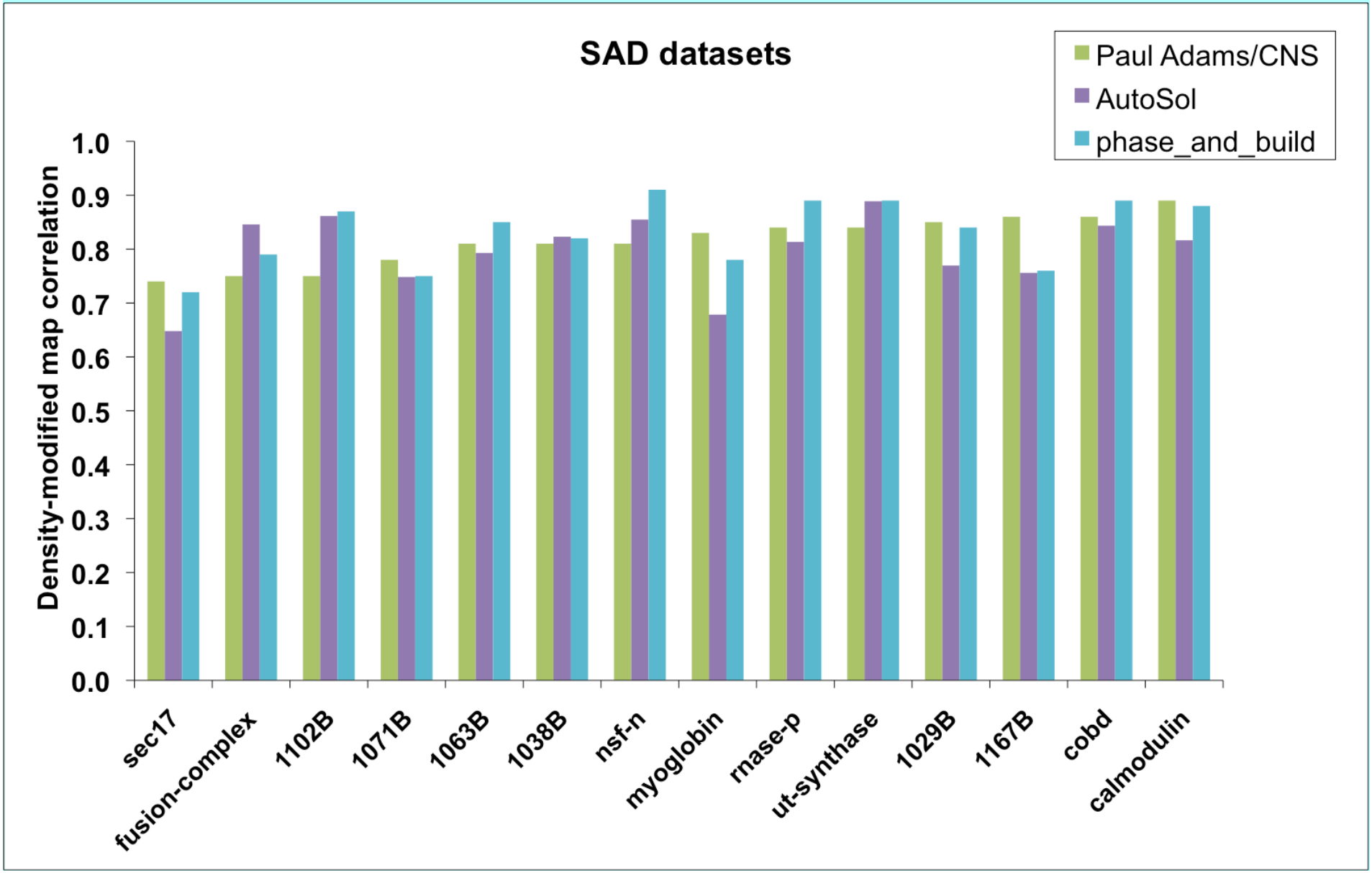
Structure solution with phenix.autosol



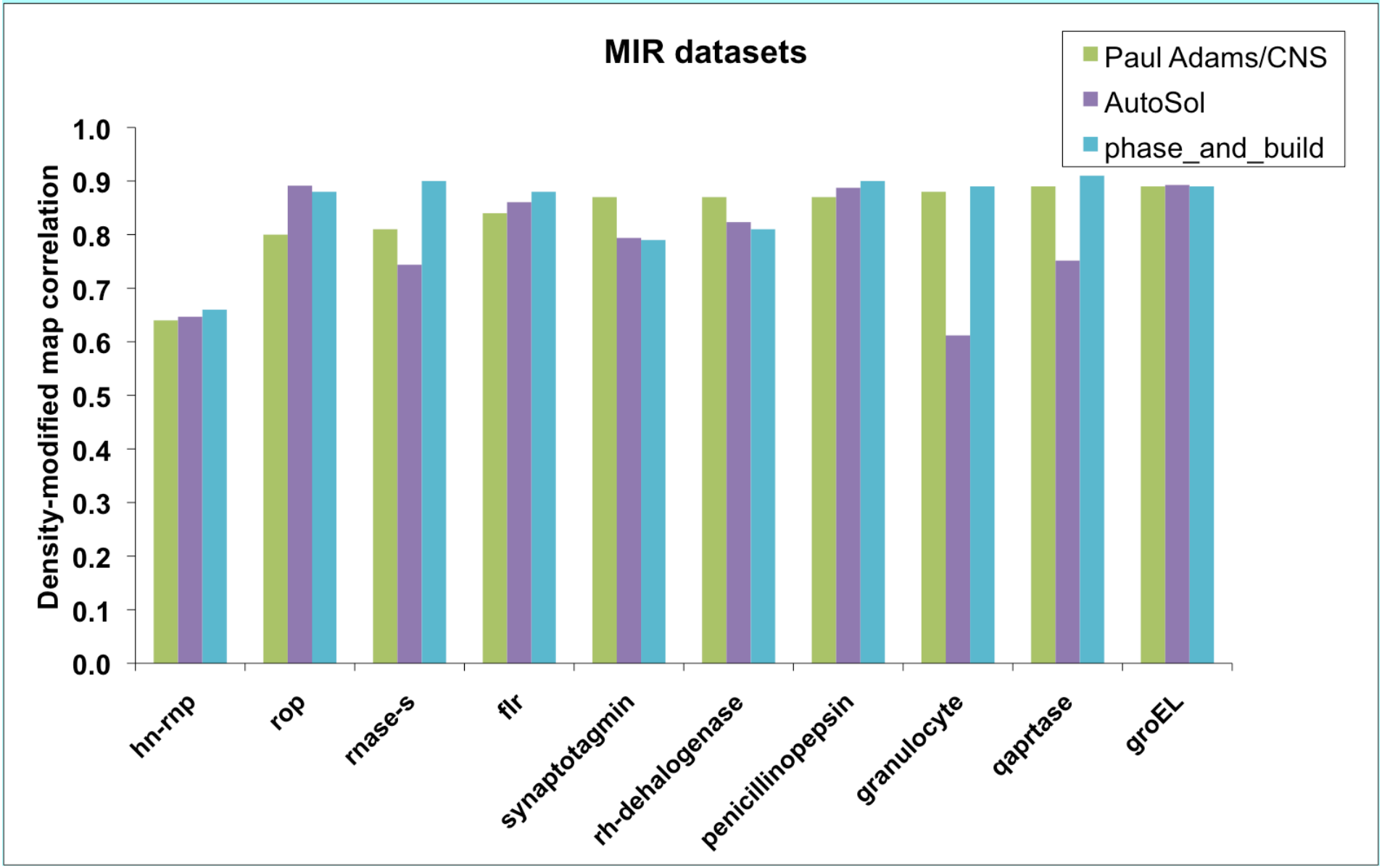
AutoSol – fully automatic tests with structure library
(MAD datasets, HYSS search, SOLVE)
RESOLVE/ phase_and_build maps



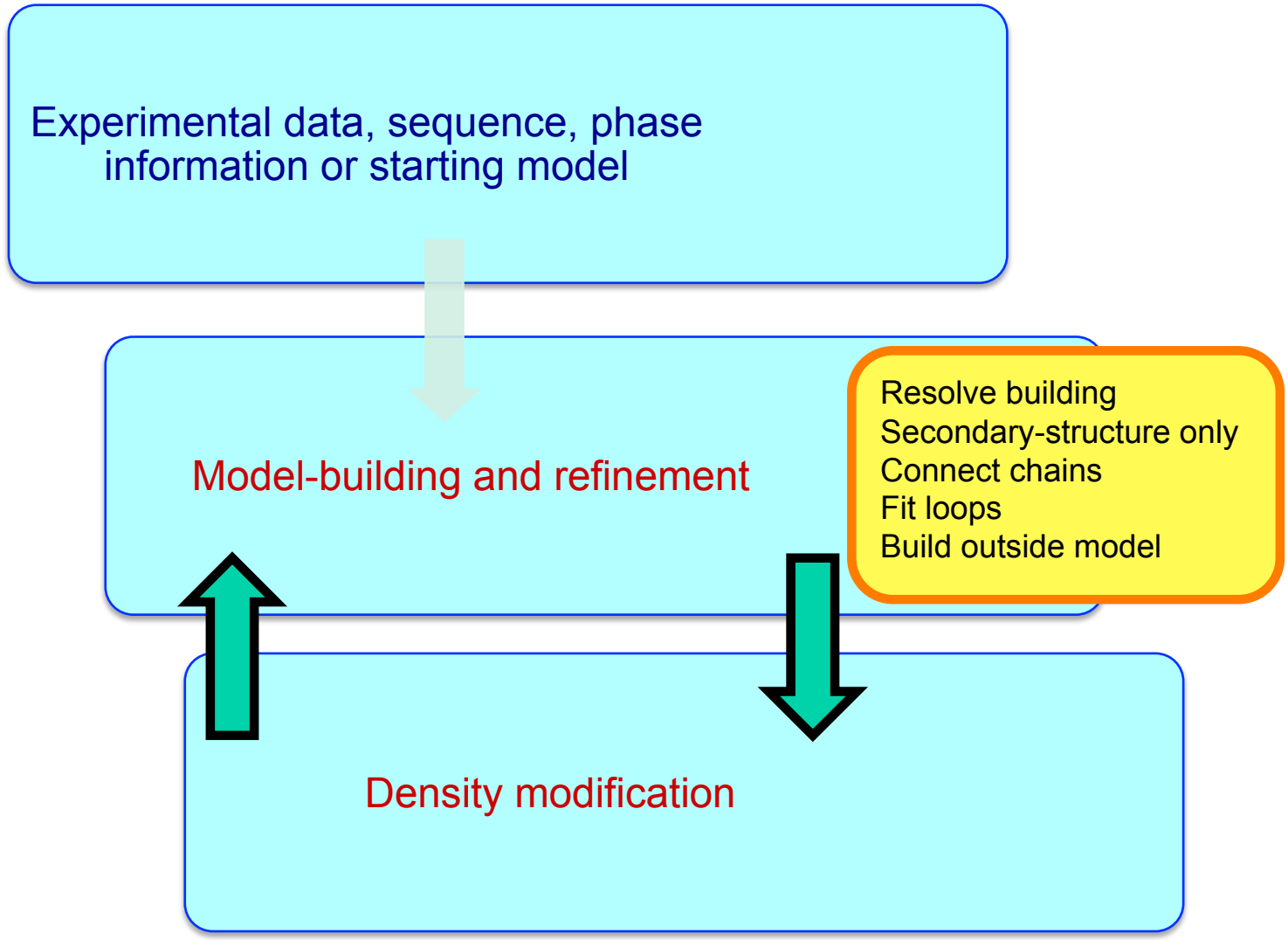
AutoSol – fully automatic tests with structure library
(SAD datasets, HYSS, Phaser)
RESOLVE/ phase_and_build maps



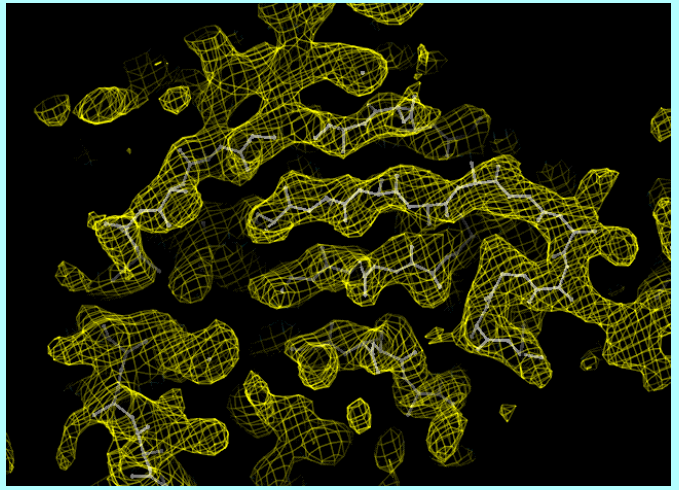
AutoSol – fully automatic tests with structure library
(SAD datasets, HYSS, Phaser)
RESOLVE/ phase_and_build maps



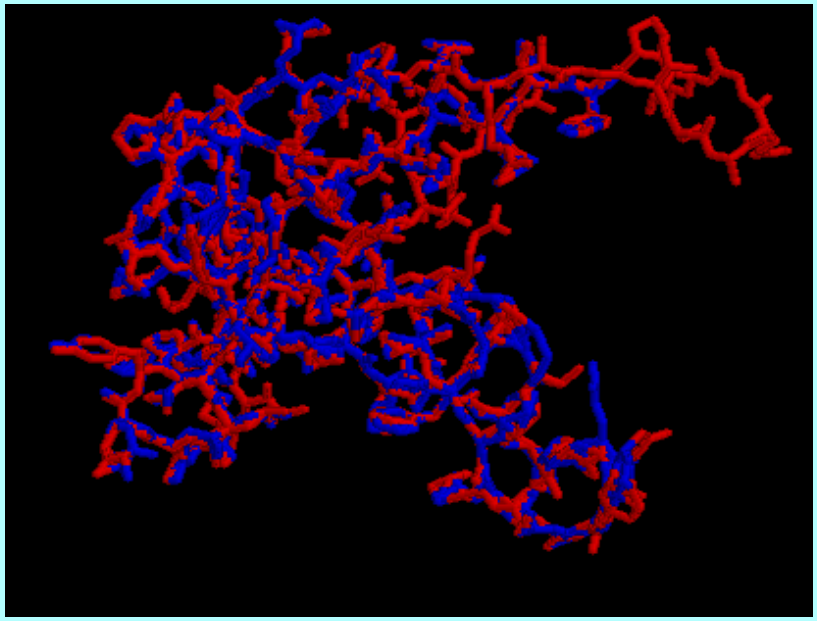
Iterative density modification, model-building and refinement with phenix.autobuild



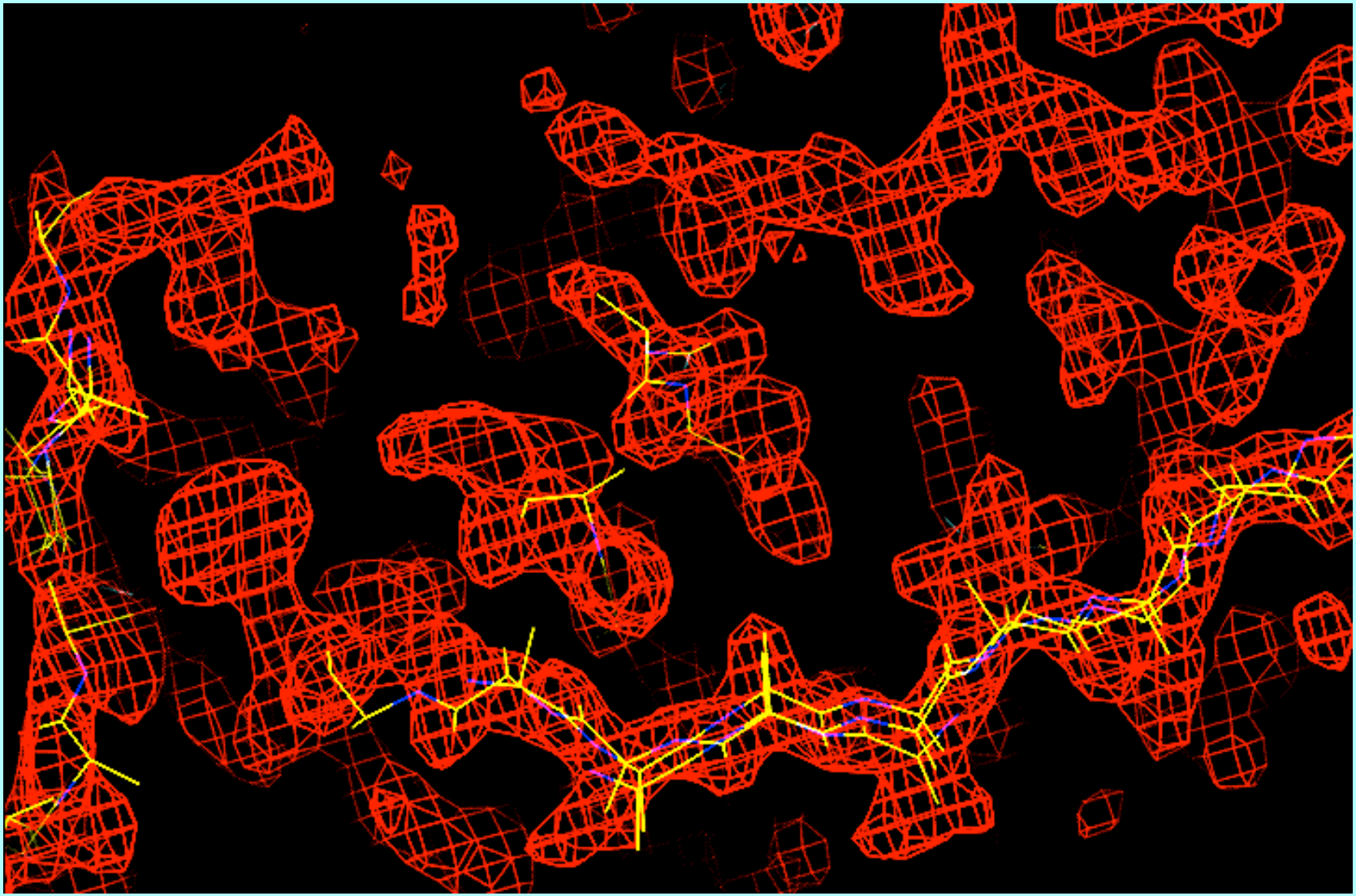
RESOLVE model-building at moderate resolution



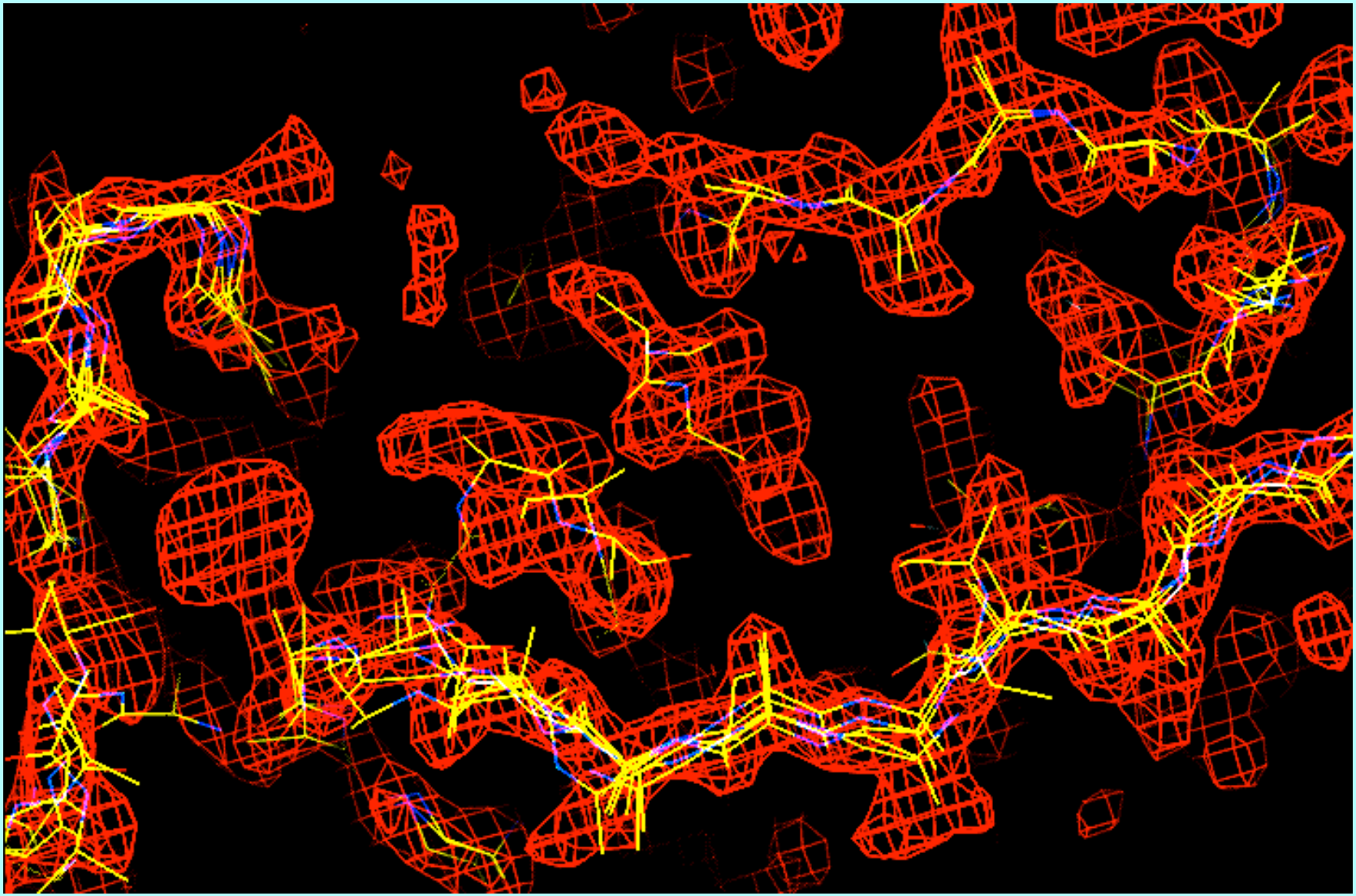
- FFT-based identification of helices and strands
- Extension with tripeptide libraries
- Probabilistic sequence alignment
- Automatic molecular assembly



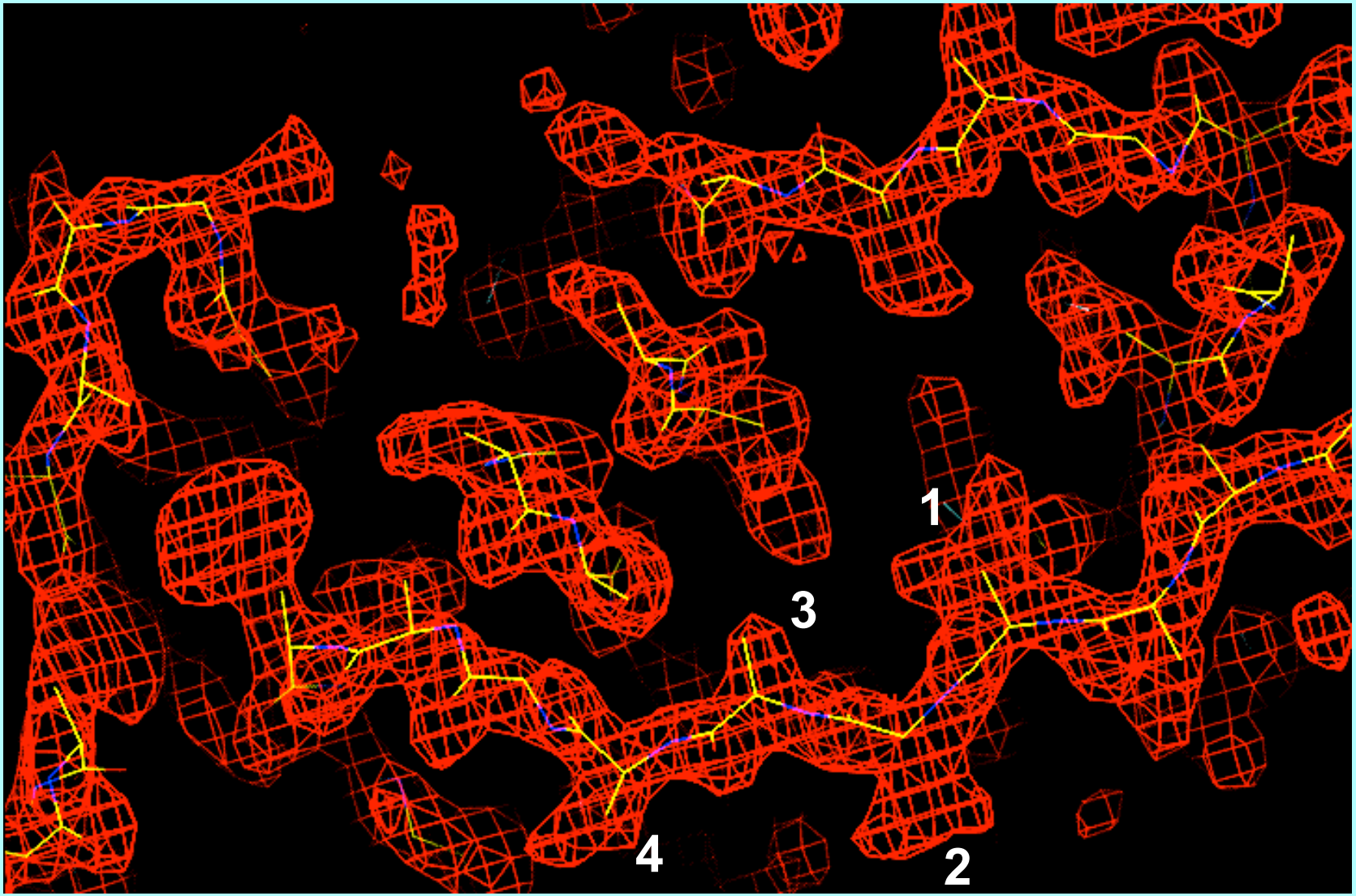
Initial model-building – strand fragments



Chain extension
(result: many overlapping fragments)



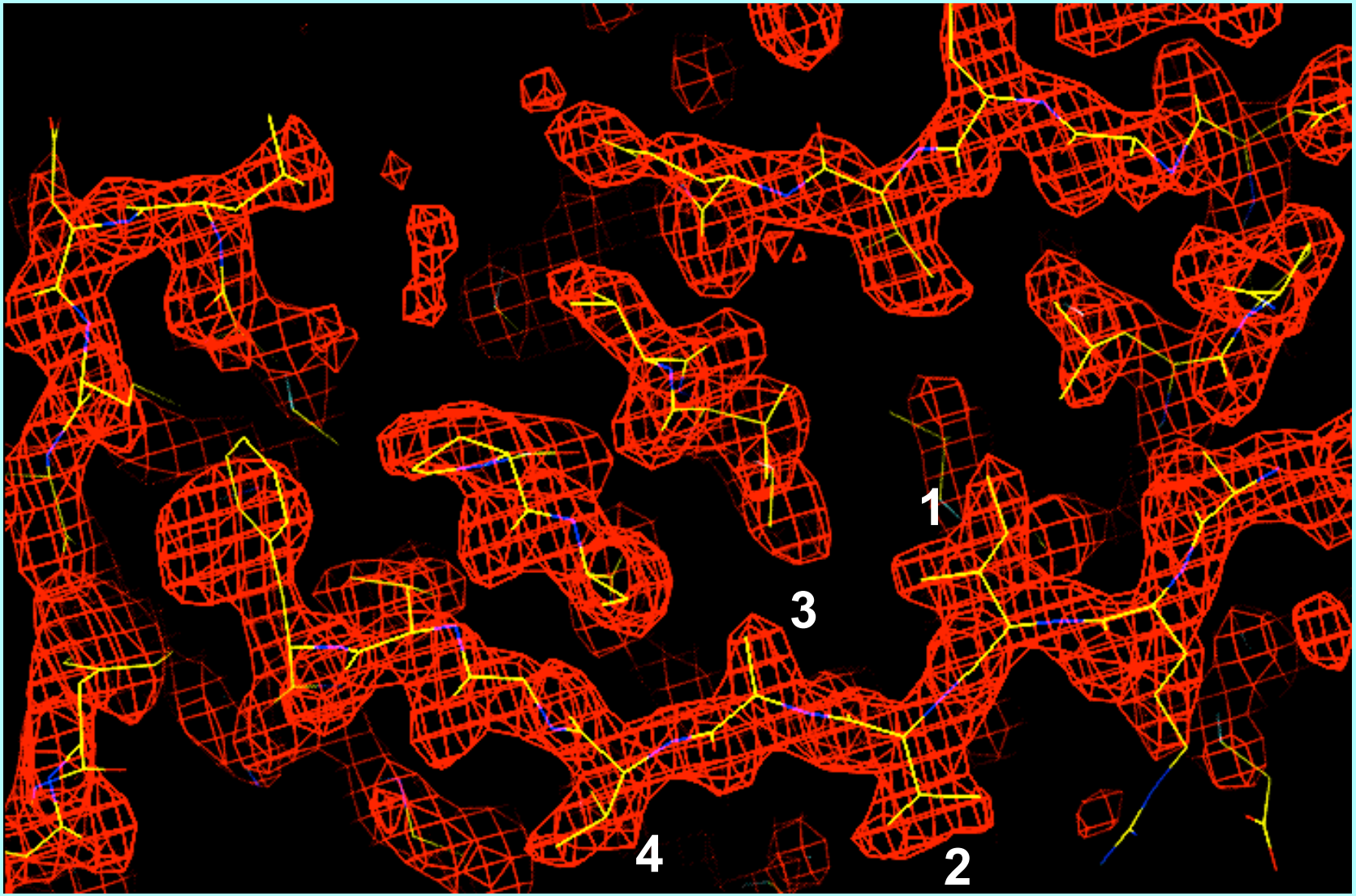
Main-chain as a series of fragments
(choosing the best fragment at each location)



Side-chain template matching to identify sequence alignment to map (IF5A data)
Relative probability for each amino acid at each position
(Correct amino acids in bold)

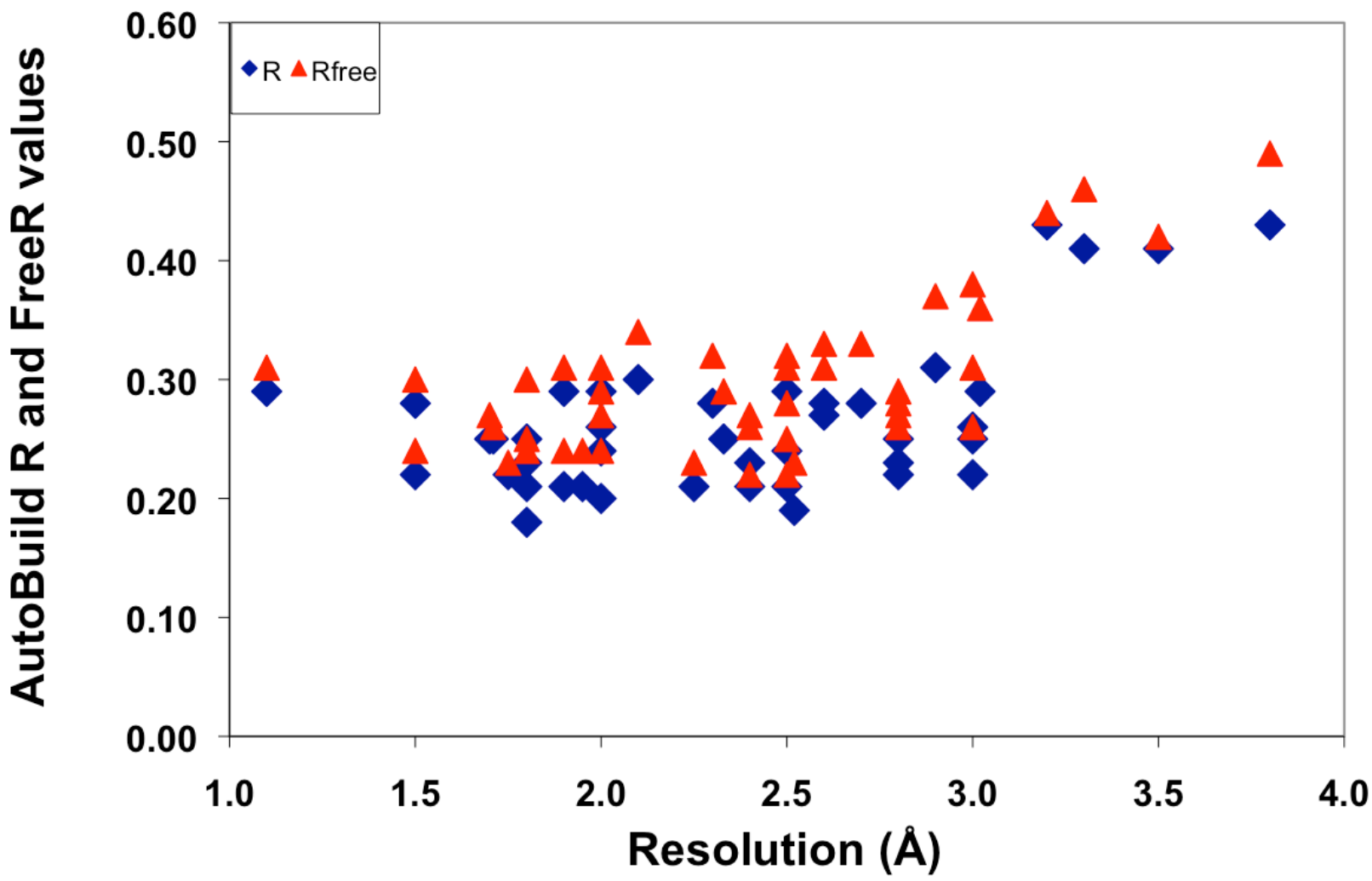
#	G	A	S	V	I	L	M	C	F	Y	K	R	W	H	E	D	Q	N	P	T
1	6	5	4	18	**18**	6	1	1	1	2	6	2	2	1	9	6	1	0	1	4
2	4	11	14	**37**	5	2	0	2	0	0	2	3	0	0	1	2	0	0	0	6
3	11	**23**	5	12	5	3	2	0	1	3	7	3	1	0	5	3	2	0	2	2
4	7	9	6	**16**	8	5	2	0	1	3	8	4	1	0	7	6	2	0	3	4
5	**31**	7	3	7	4	2	1	0	1	3	5	4	1	0	6	2	2	0	11	1
6	1	3	3	**41**	14	8	0	0	0	0	2	1	0	0	2	4	0	0	1	9
7	0	0	0	0	0	0	0	0	**15**	63	1	0	17	1	0	0	0	0	0	0
8	2	3	6	23	10	6	2	1	0	1	4	3	0	0	5	**16**	1	0	1	6
9	**96**	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0

Addition of side-chains to fixed main-chain positions



AutoBuild – tests with structure library

Fully automated iterative model-building, final R/Rfree



Rapid building of models for regions containing regular secondary-structure

Helices:

Identification: rods of density at low resolution

Strands:

Identification: β structure as nearly-parallel pairs of tubes

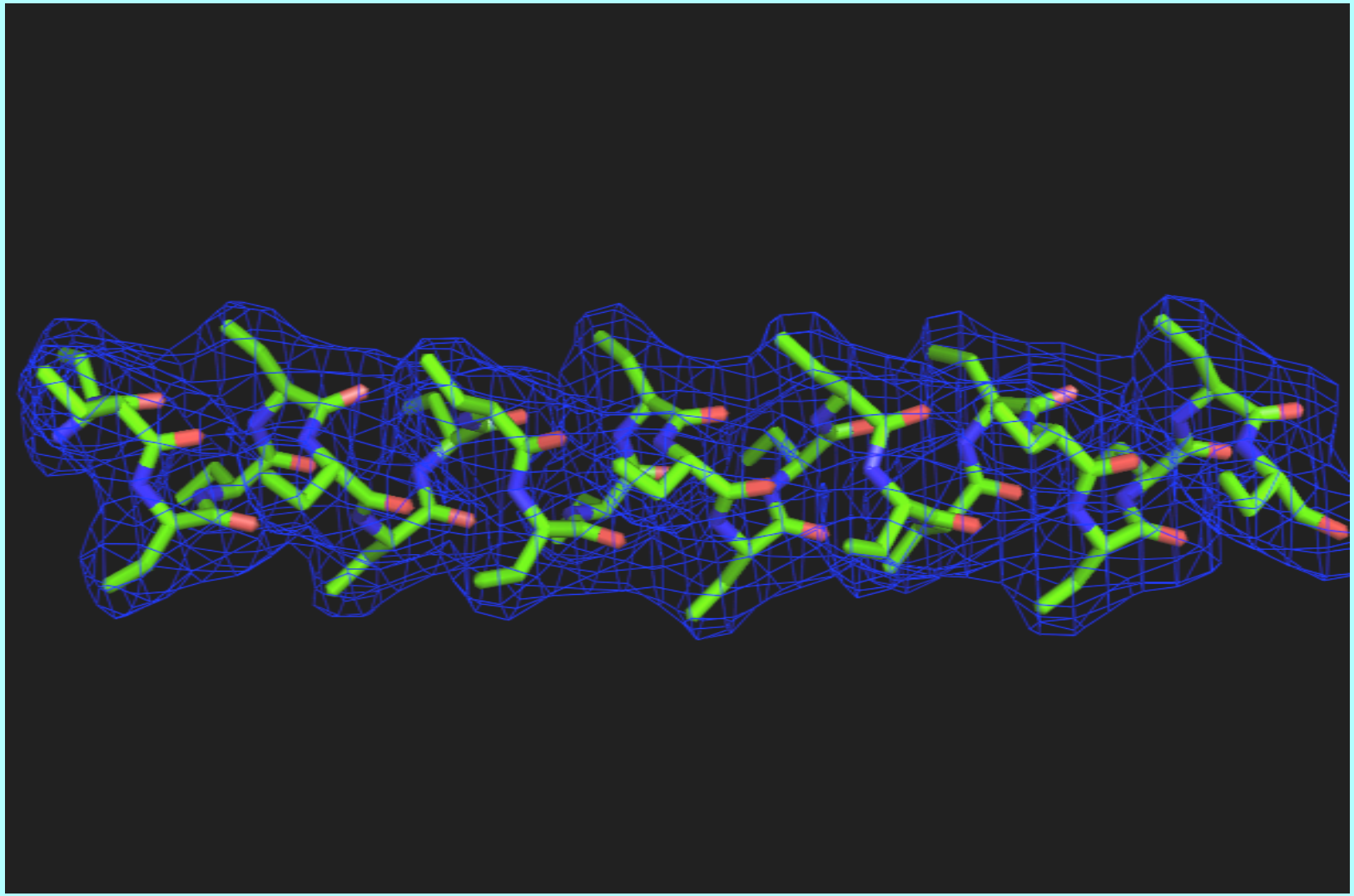
Any protein chains (trace_chain):

Identification: Cα positions consistent with density and geometry of protein chains

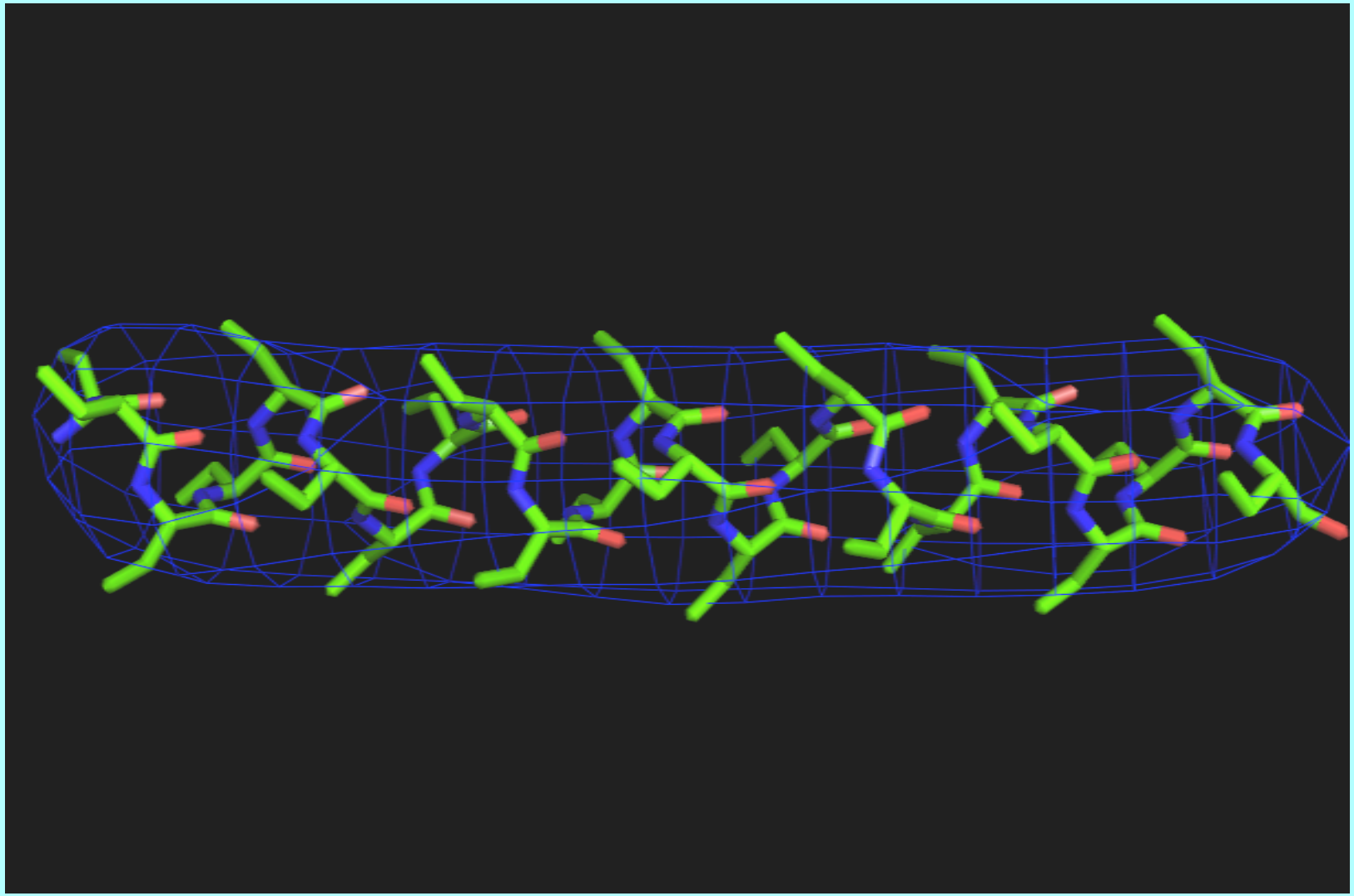
RNA/DNA:

Identification: match of density to averaged A or B-form template

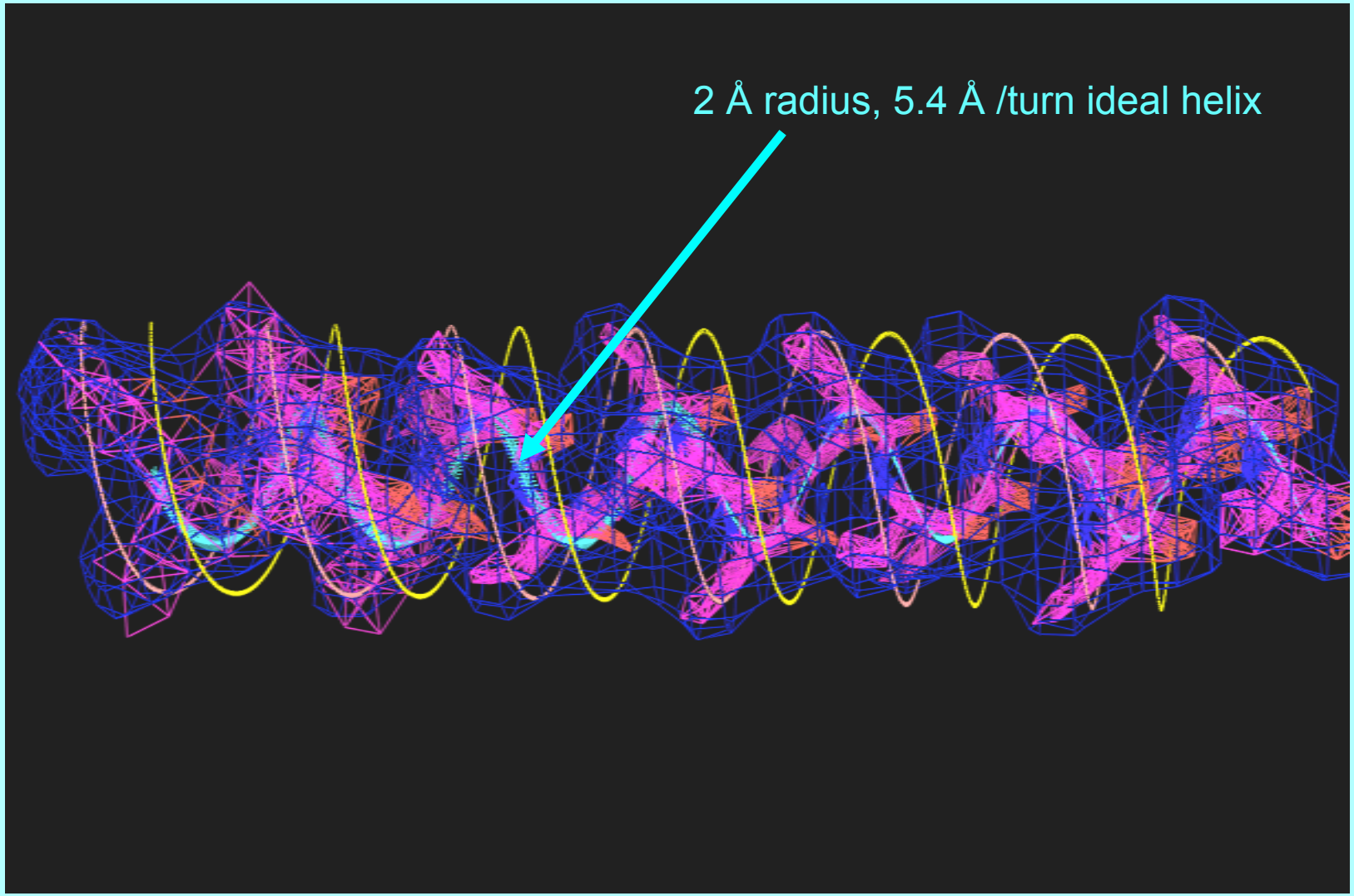
Model α-helix; 3 Å map



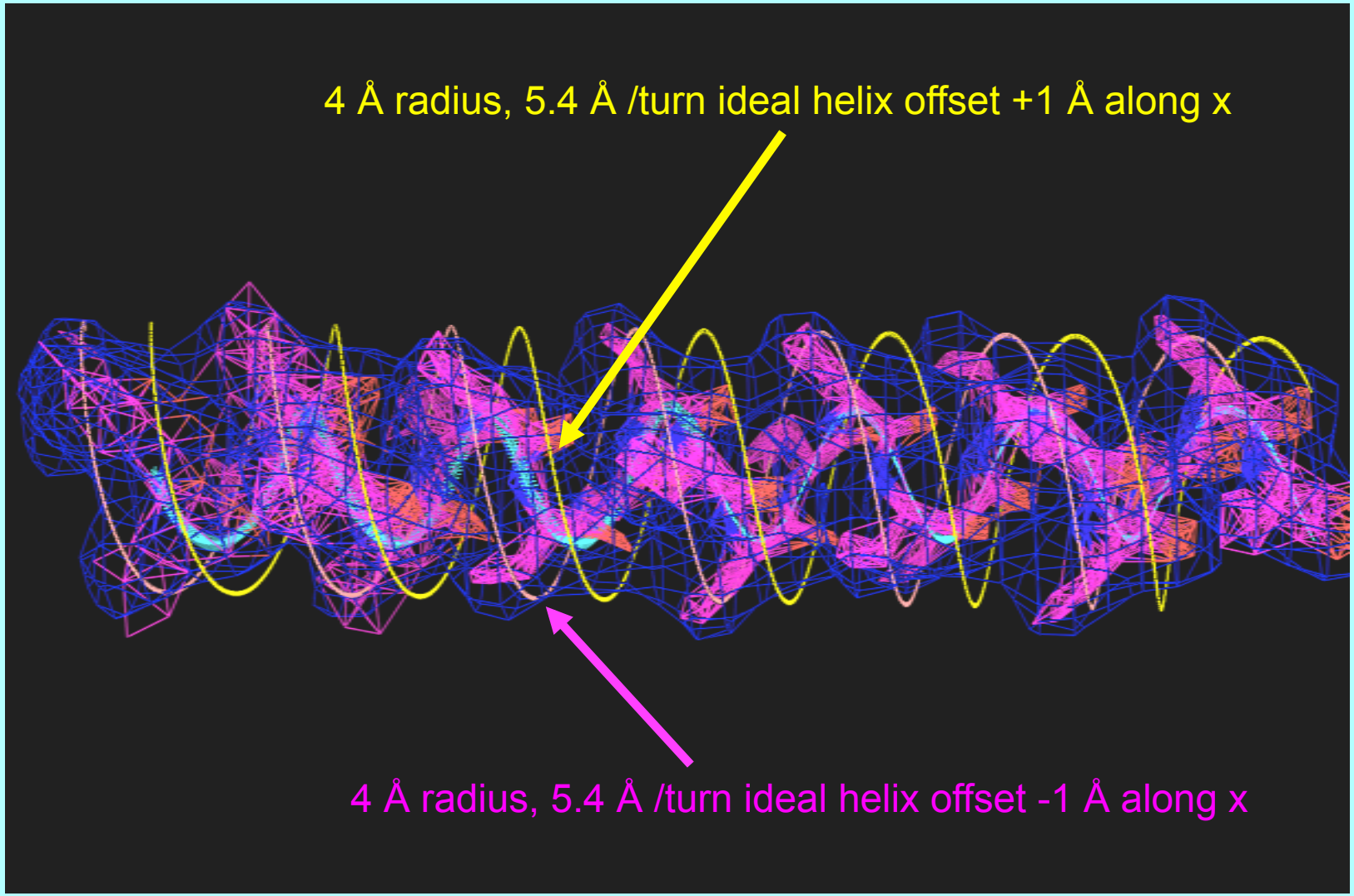
Model α-helix; 7 Å map



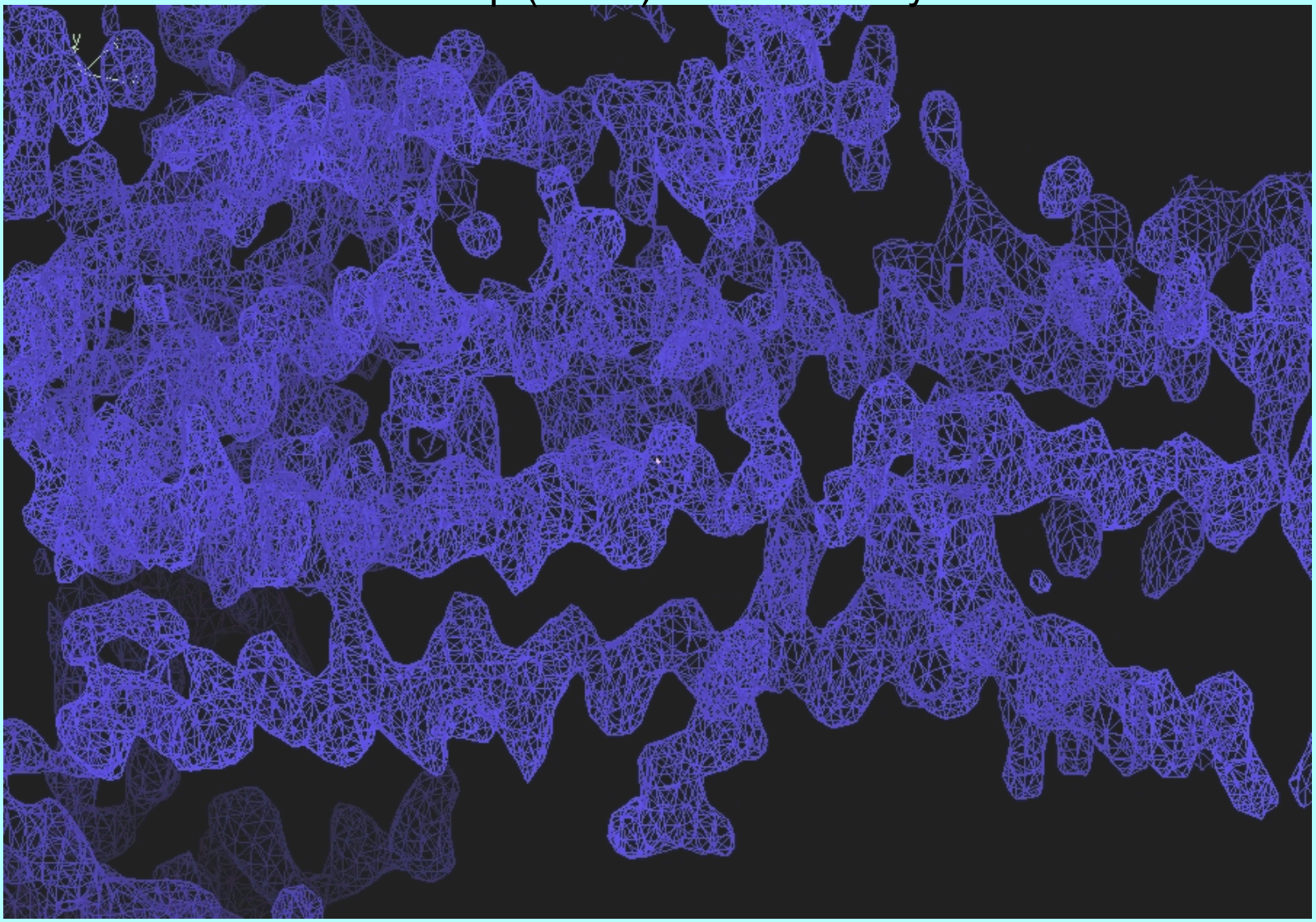
Trace main-chain with ideal helix, allowing curvature



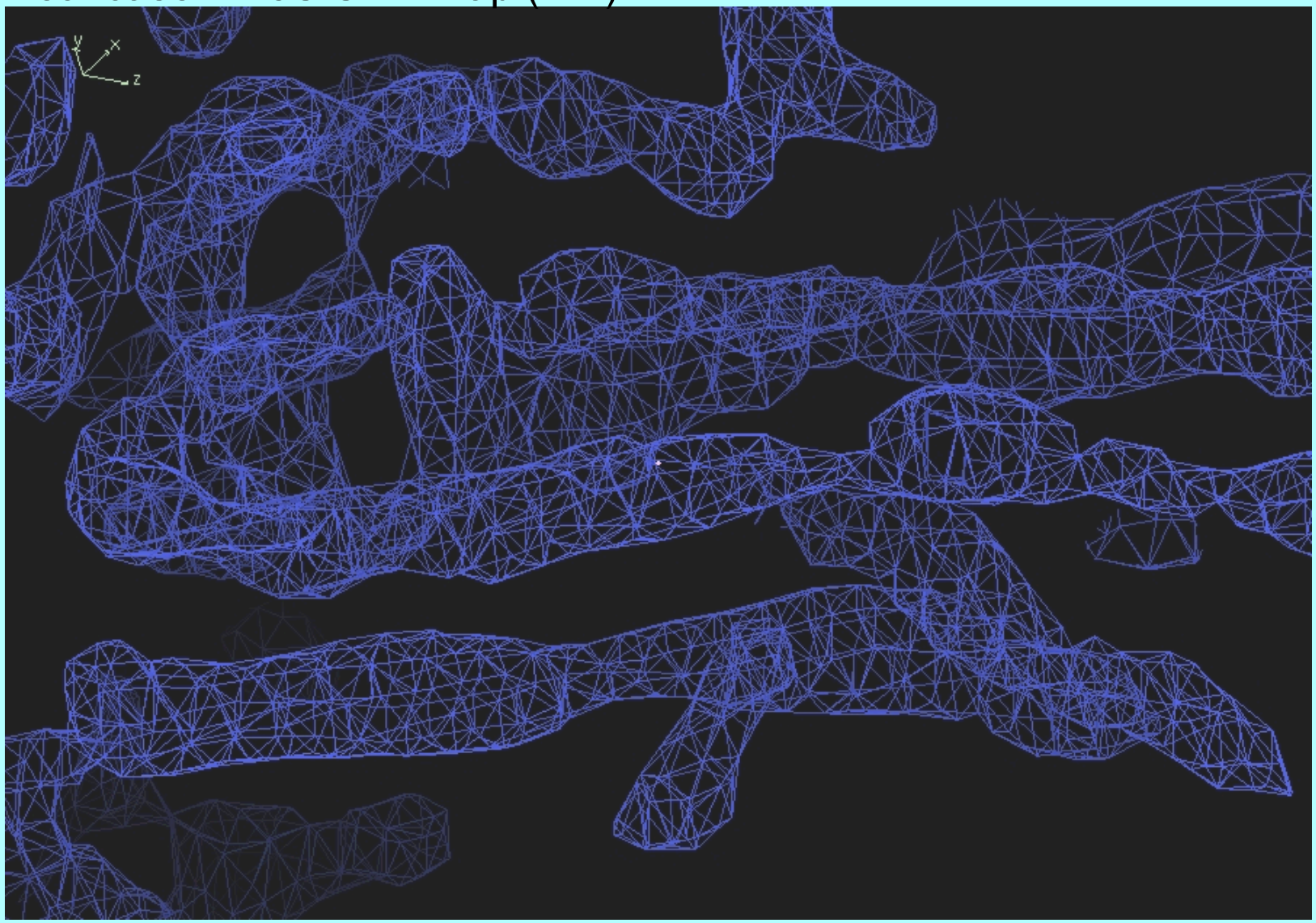
Identify direction and Cα position from overlap with 4 Å radius helices offset +/- 1 Å from main-chain



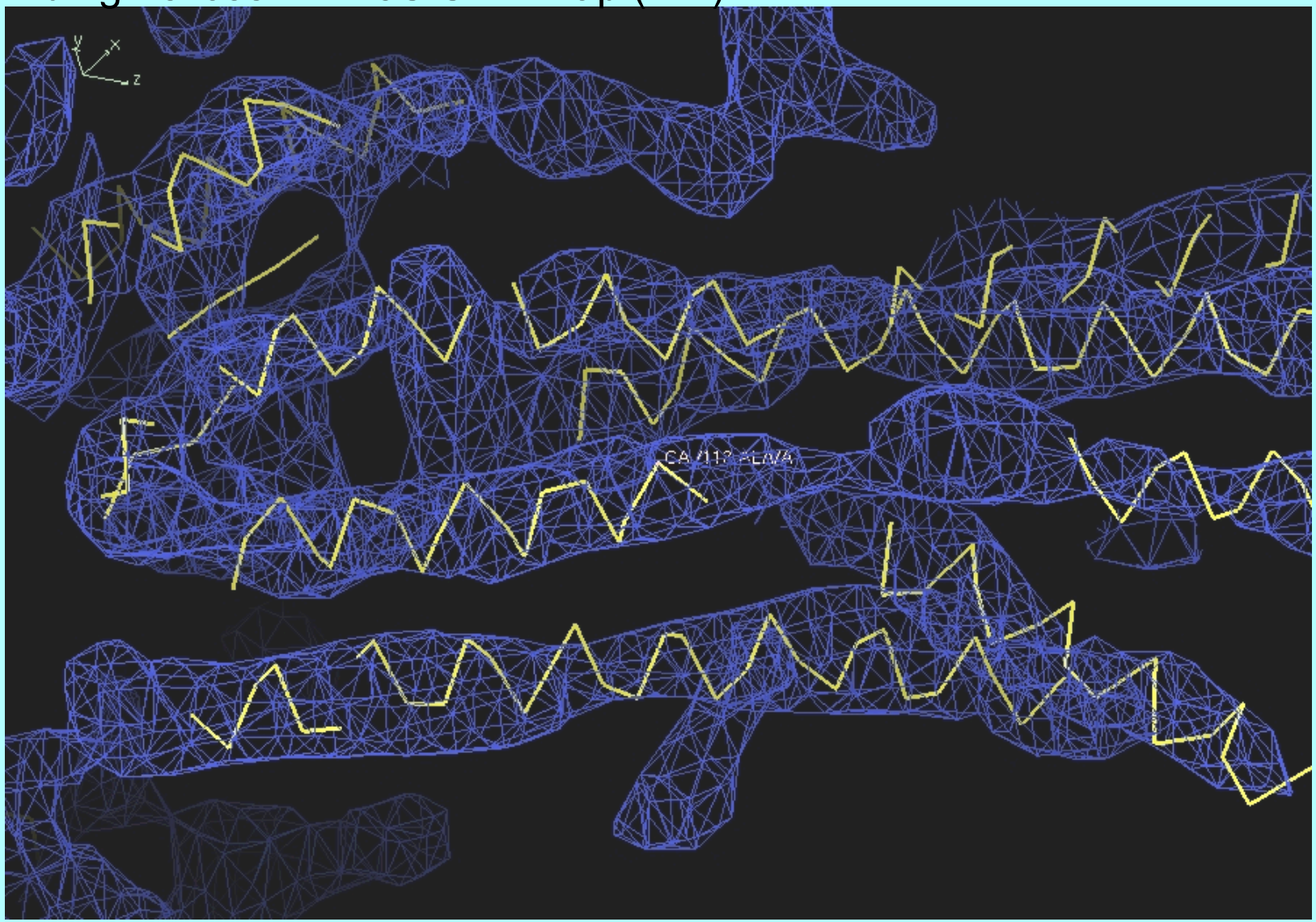
A real case: 1T5S SAD map (3.1 Å) Data courtesy of P. Nissen



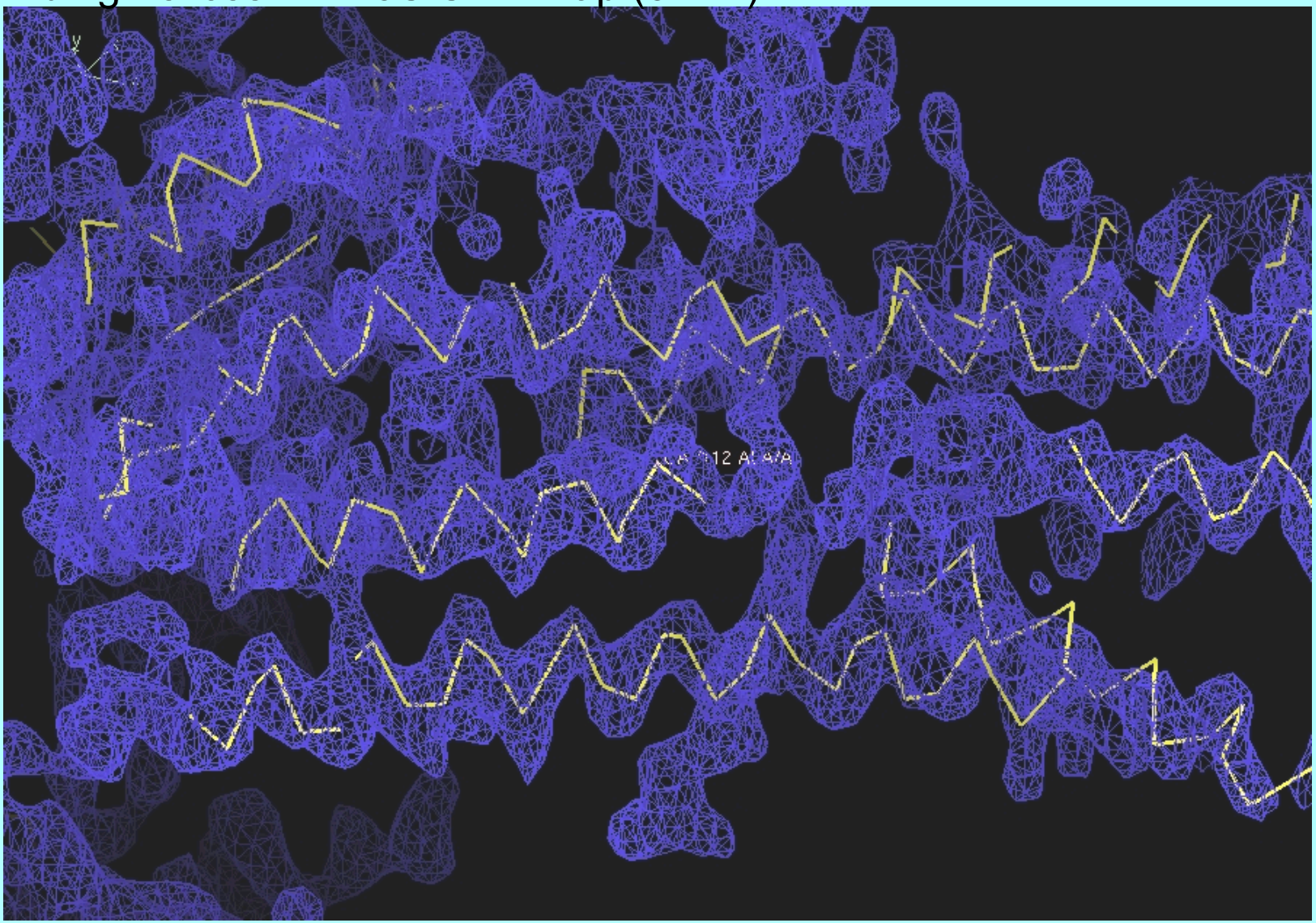
A real case: 1T5S SAD map (7 Å)



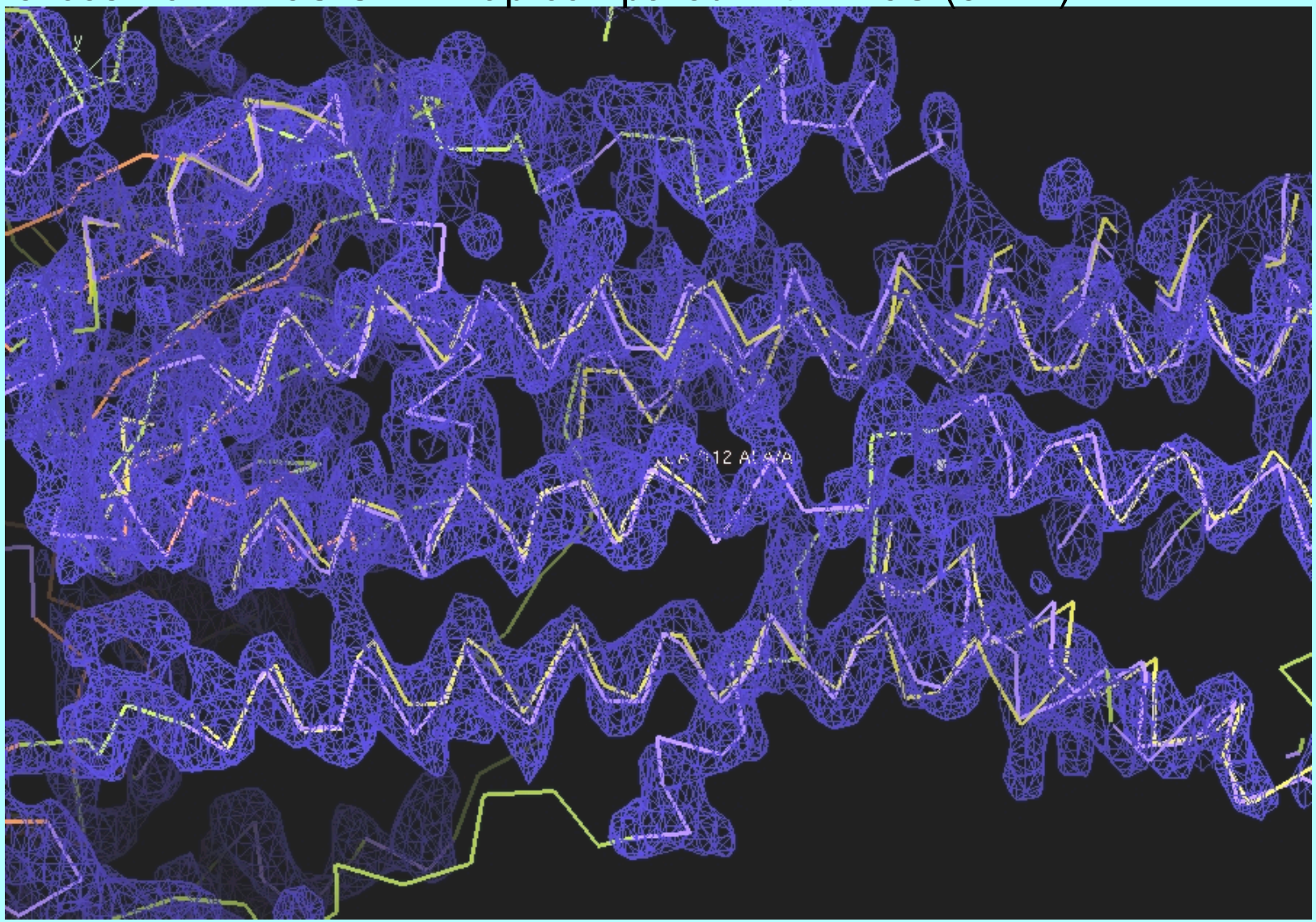
Finding helices in 1T5S SAD map (7 Å)

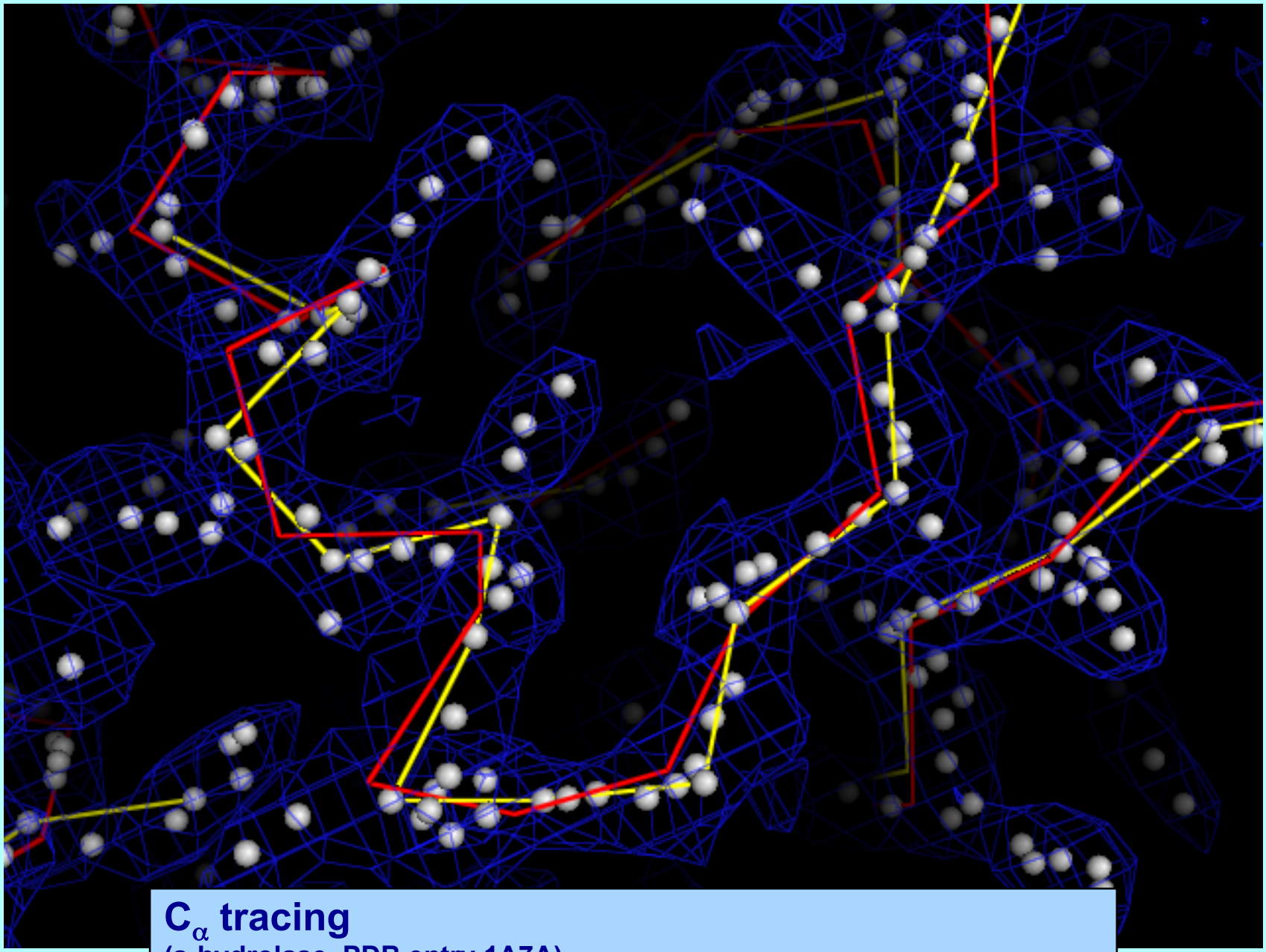


Finding helices in 1T5S SAD map (3.1 Å)



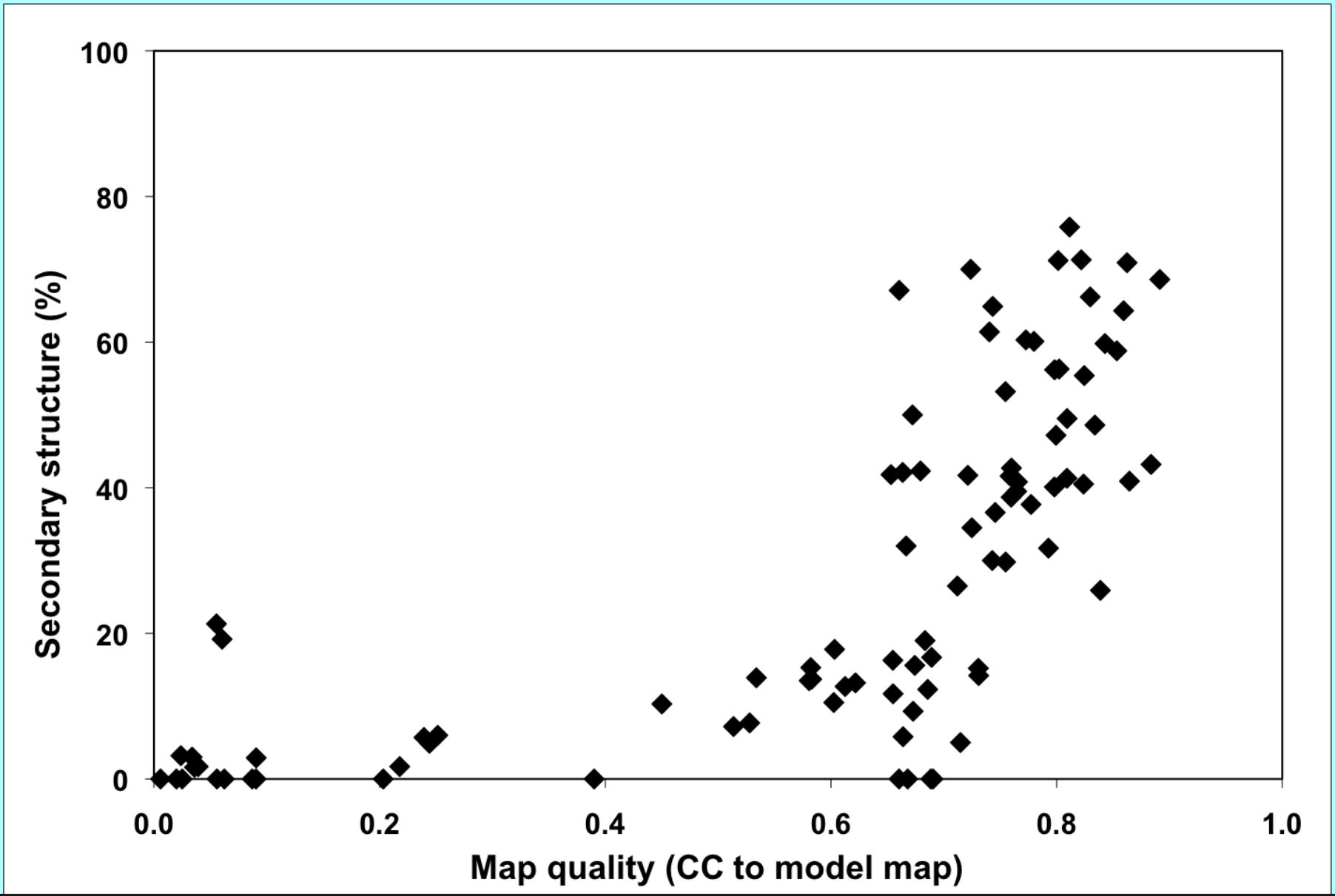
Helices from 1T5S SAD map compared with 1T5S (3.1 Å)





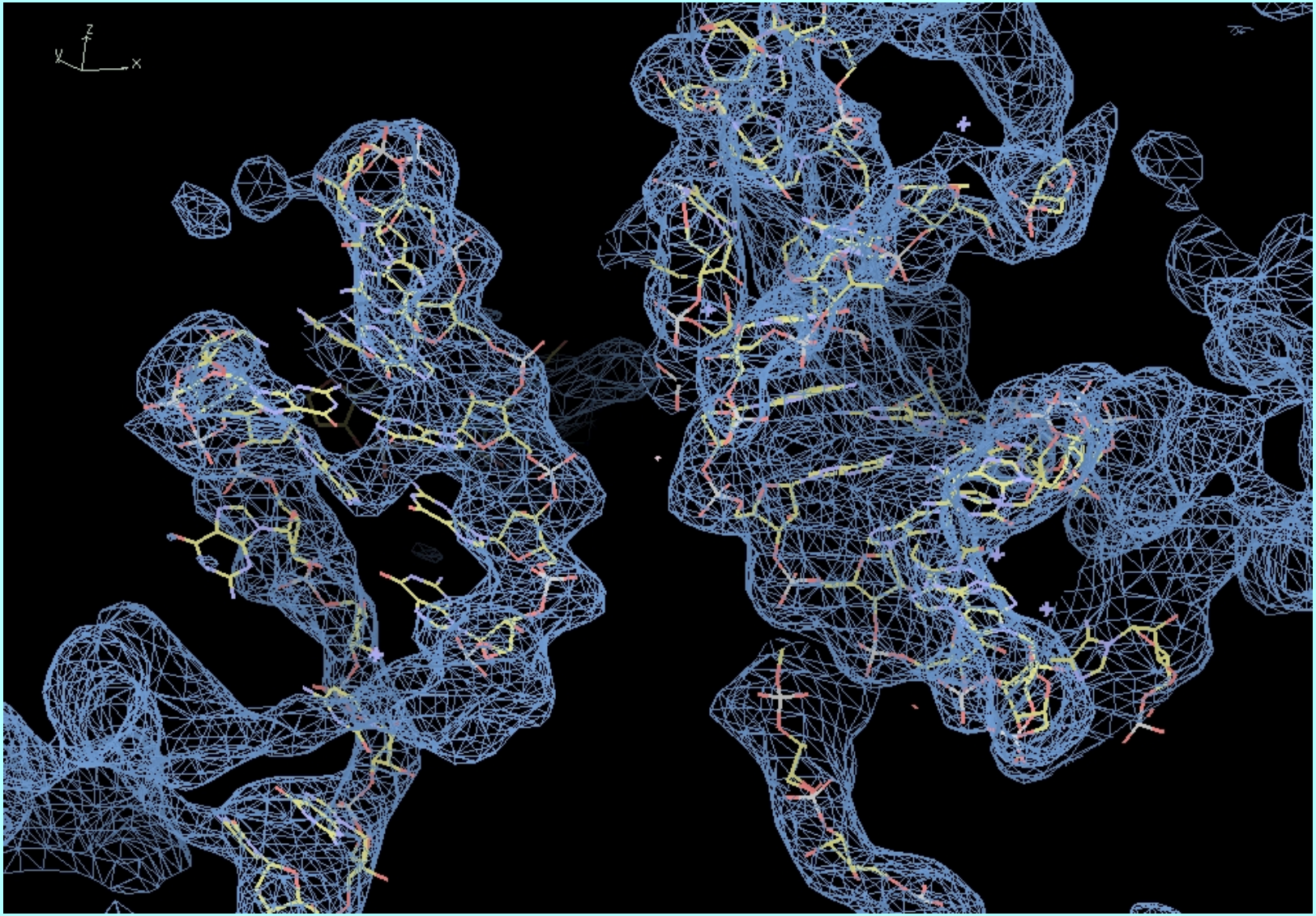
C_α tracing
(s-hydrolase, PDB entry 1A7A)

C_α tracing
(mevalonate kinase, PDB entry 1KKH, 9 sec)



Using secondary structure content to evaluate map quality

Building RNA
Group II intron at 3.5 Å. Data courtesy of J. Doudna



Rapid phase improvement and model-building with phenix.phase_and_build

First improve the map

NCS identification from density

Iterative rapid model-building and density modification

Then build a full model

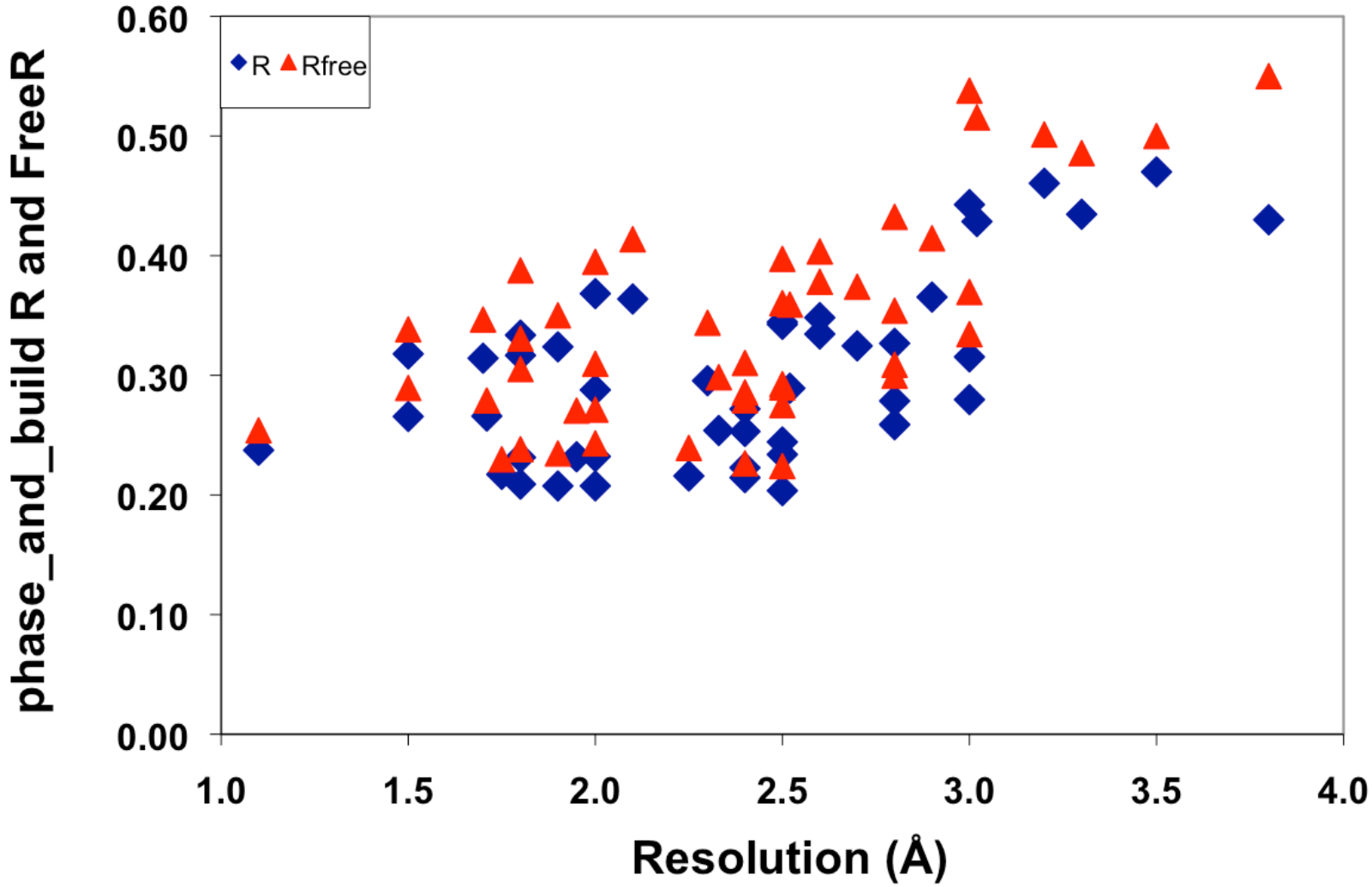
Model-building and refinement with NCS

Comprehensive sequence assignment

Loop fitting

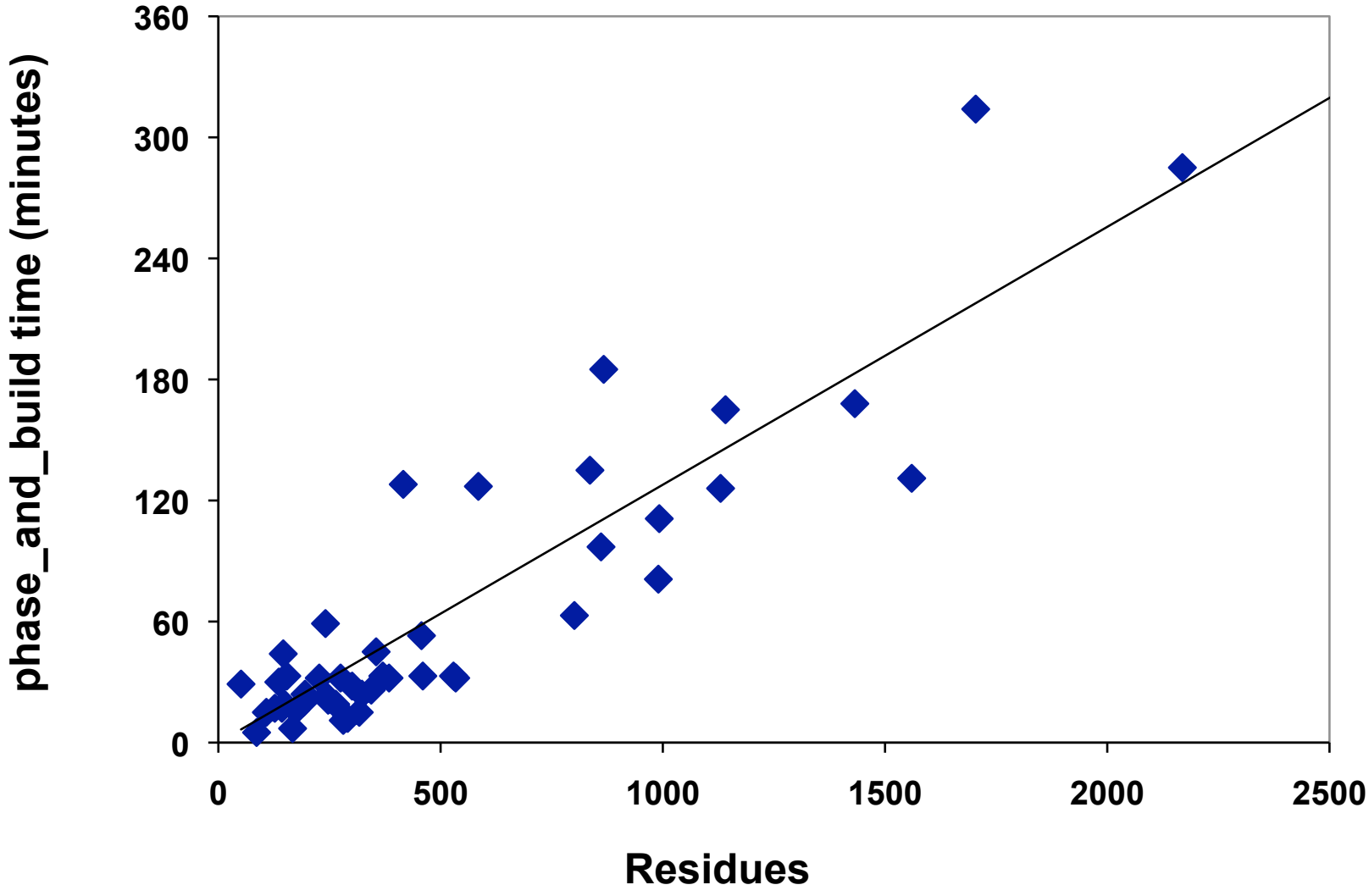
phenix.phase_and_build – tests with structure library

Final R/Rfree



phase_and_build – tests with structure library

One cycle (approx 500 residues/hour)



What can you do with automated procedures for structure solution and model-building?

If a task is modular and automated…

you can run it many times

…checking different space groups, datasets to use

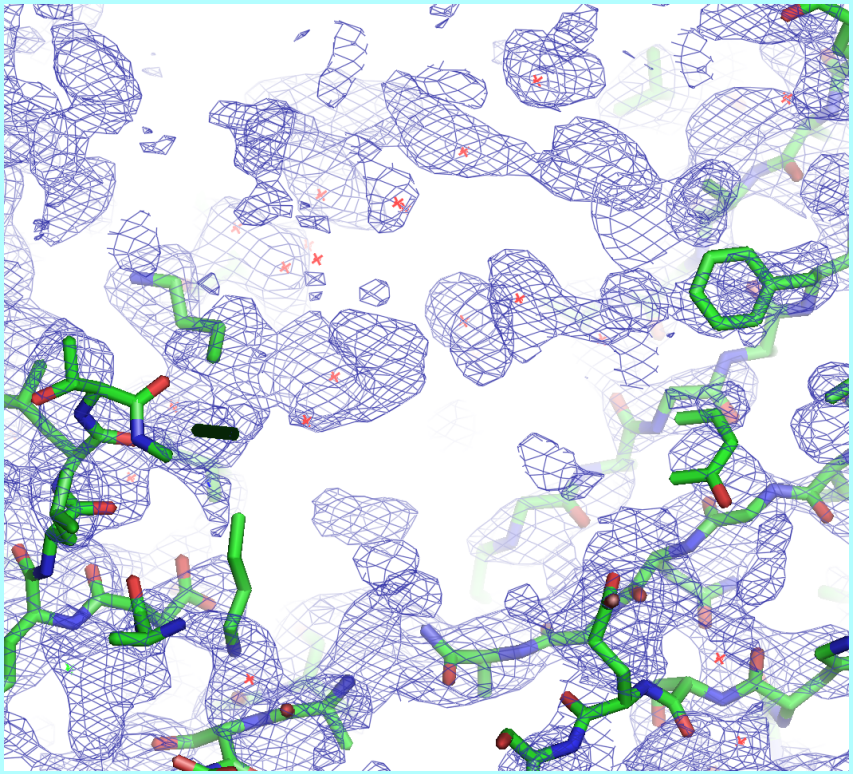
…checking if your model is biasing your map

…checking if you always get the same model

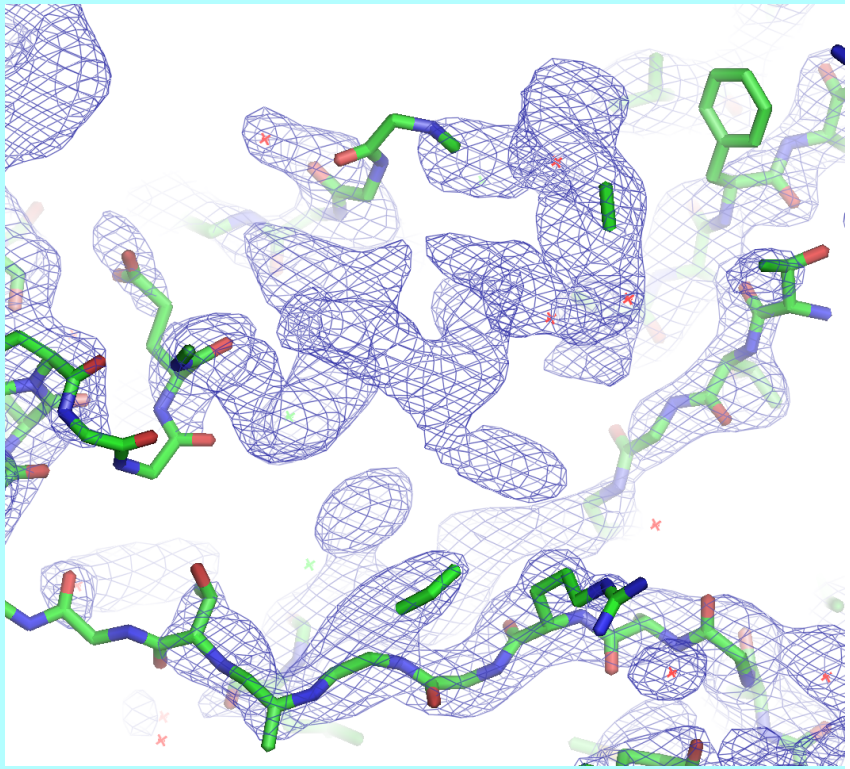
Iterative-Build OMIT procedure

"Is the density in my map biased by the model?"

2mFo-DFc omit map



After building outside OMIT region 10 cycles



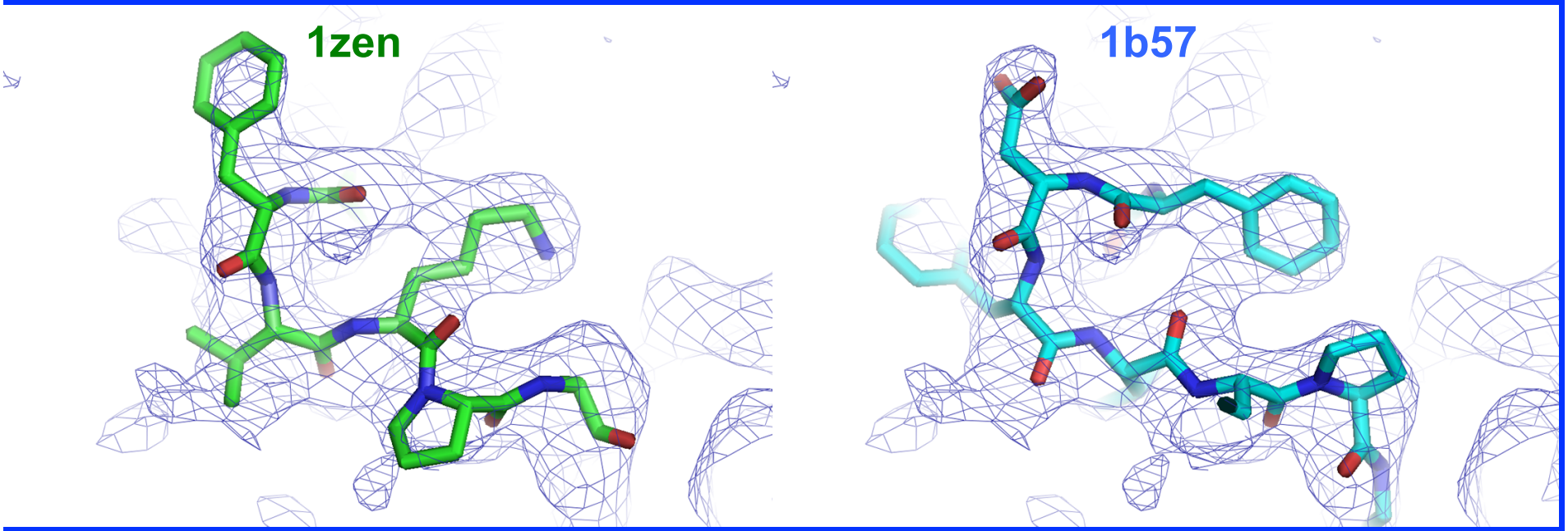
1HP7 molecular replacement with 1AS4

R/Rfree after initial refinement: 0.41/0.48

Iterative-Build OMIT procedure

"Removing model bias"

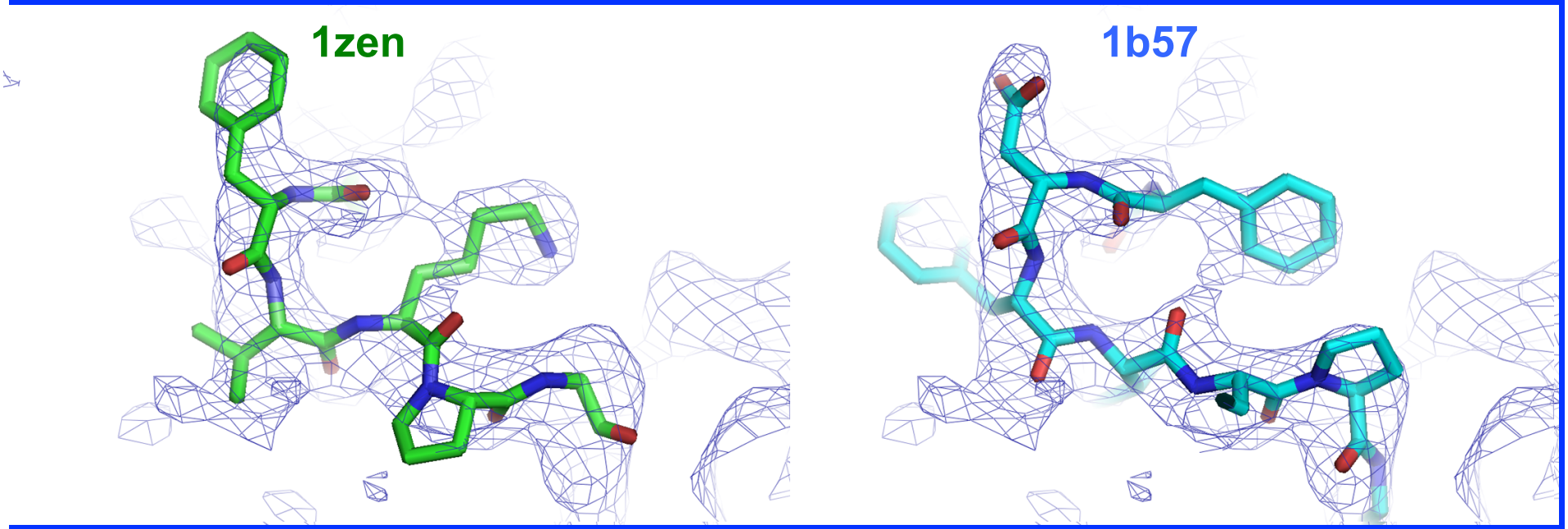
2mFo-DFc map
Phased with 1zen model



Iterative-Build OMIT procedure

"Removing model bias"

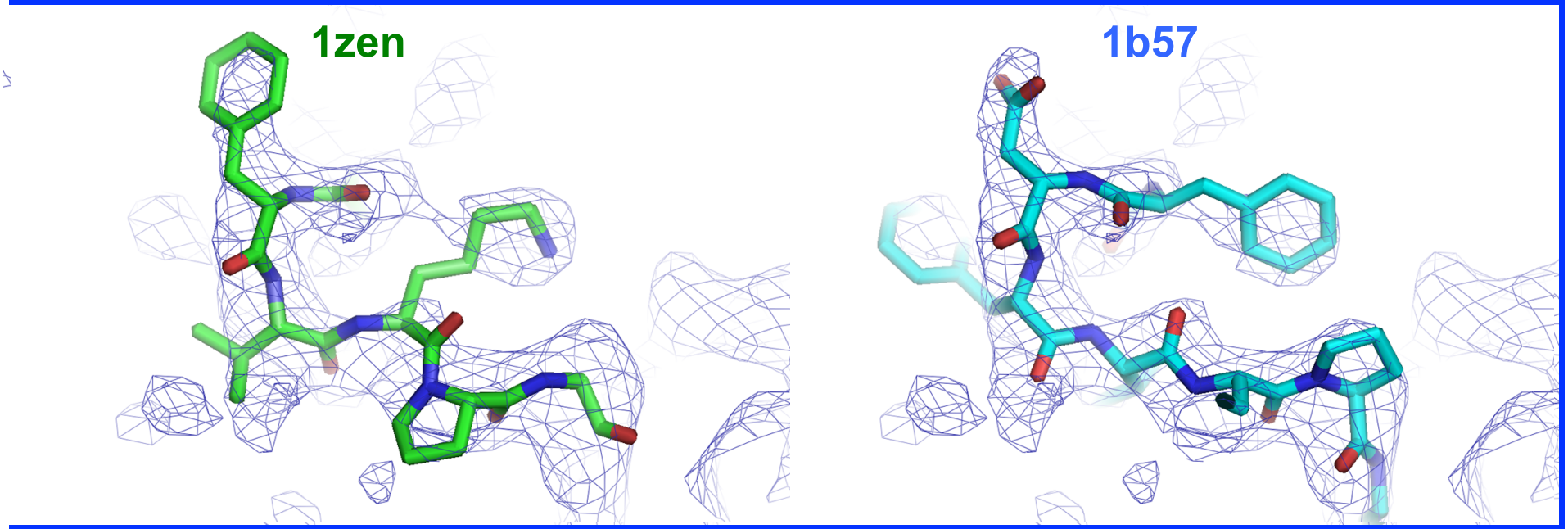
2mFo-DFc omit map
Phased with 1zen model



Iterative-Build OMIT procedure

"Removing model bias"

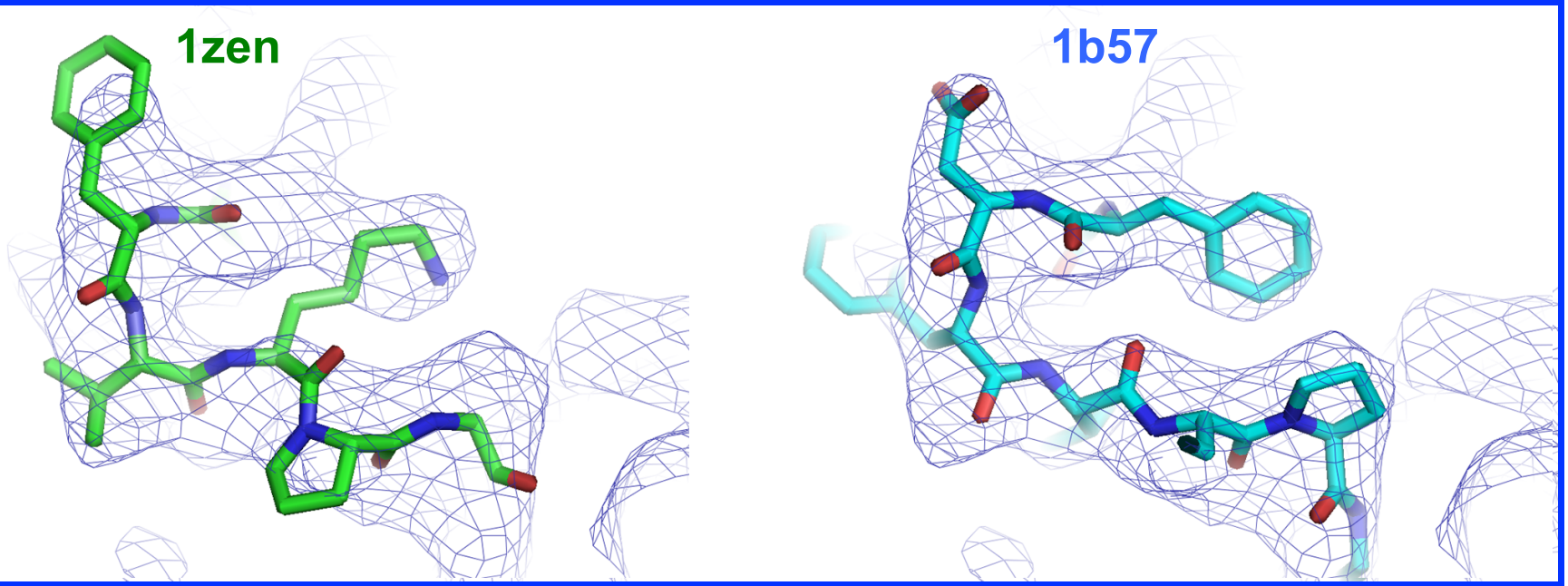
2mFo-DFc SA-omit map
Phased starting with 1zen model



Iterative-Build OMIT procedure

"Removing model bias"

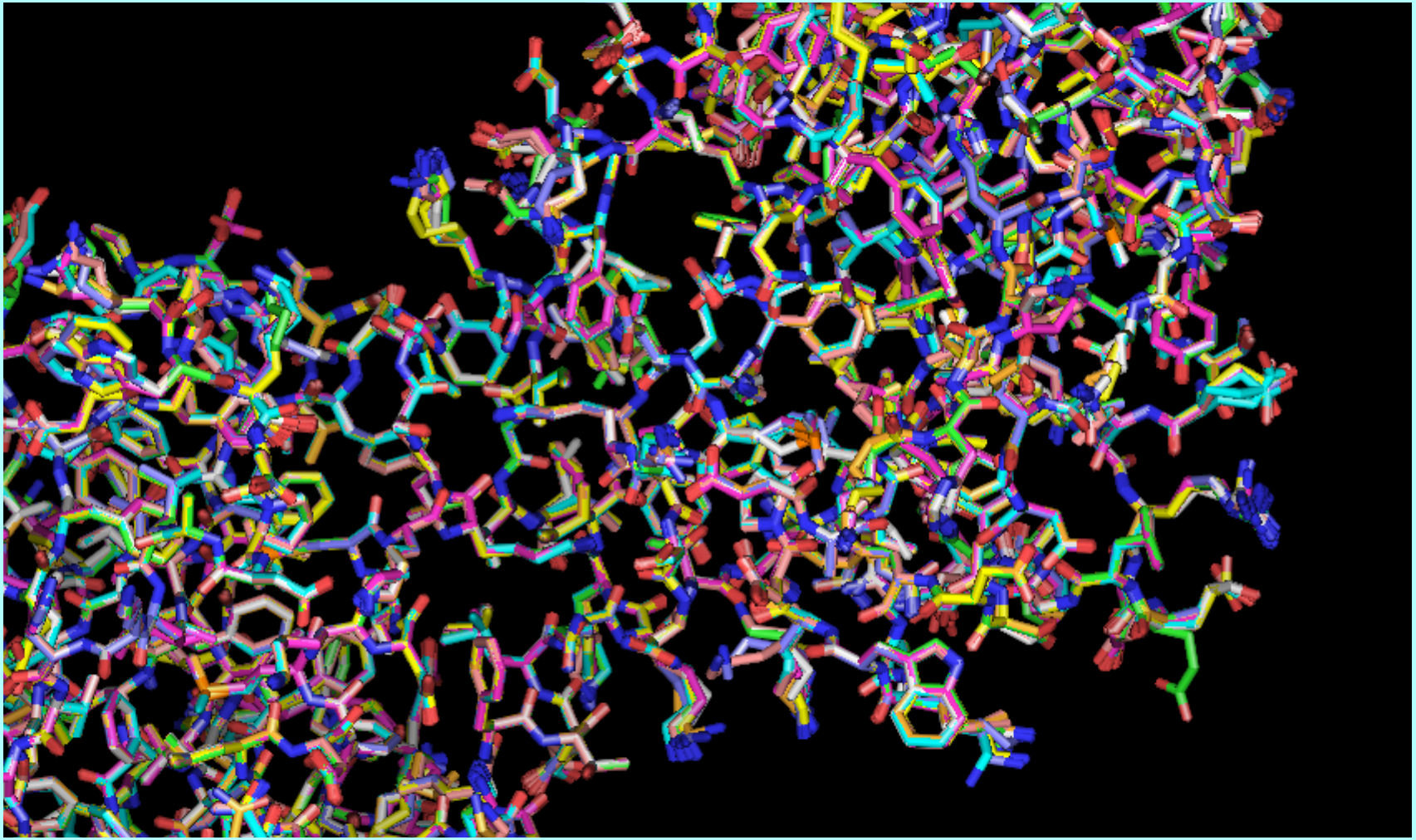
2mFo-DFc iterative-build omit map
Phased starting with 1zen model



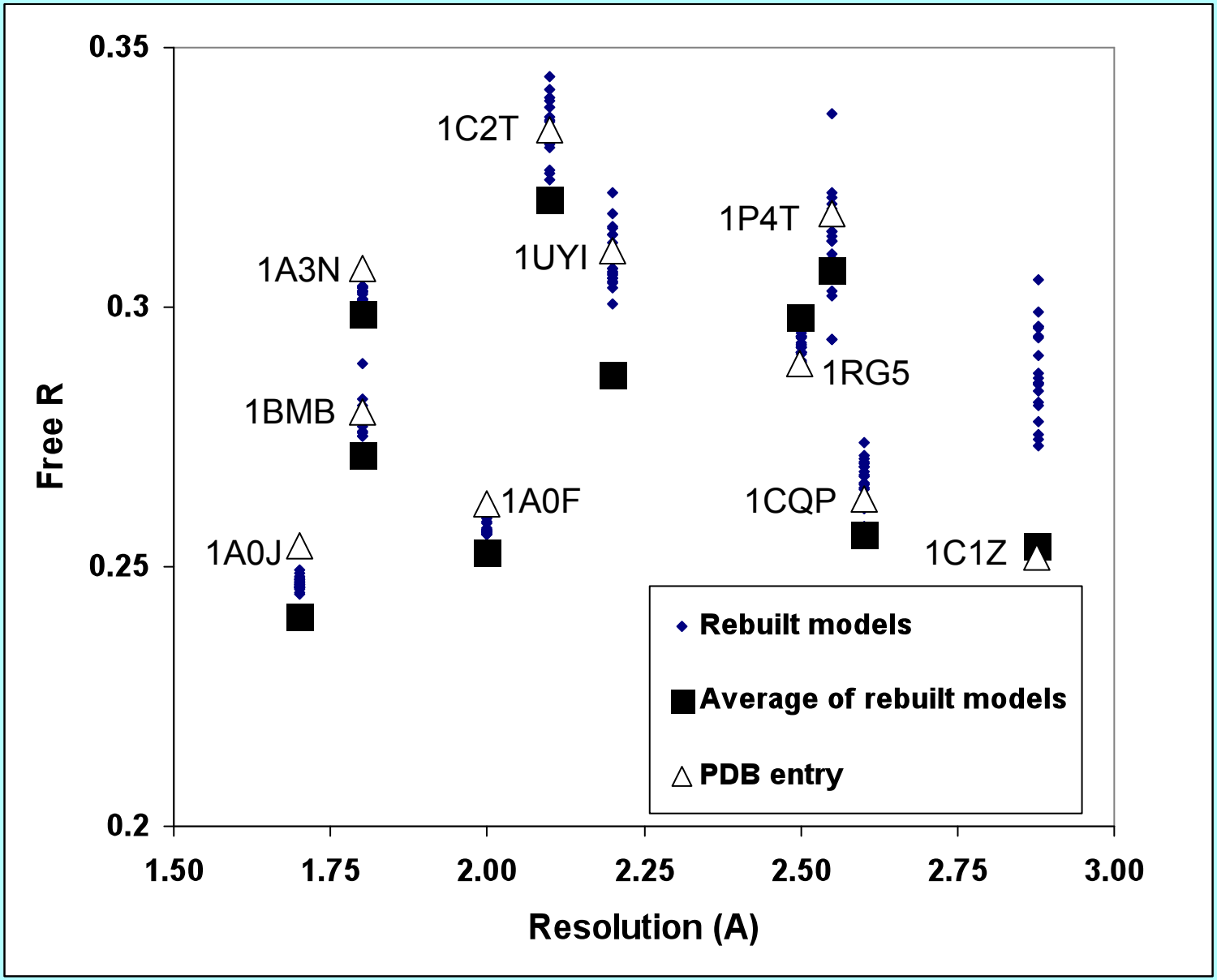
Multiple-model representation of uncertainties

20 models built for 1CQP, no waters, Dmin=2.6 A R=0.19-0.20; Rfree=0.26-0.27

The variation among models is a lower bound on their uncertainty

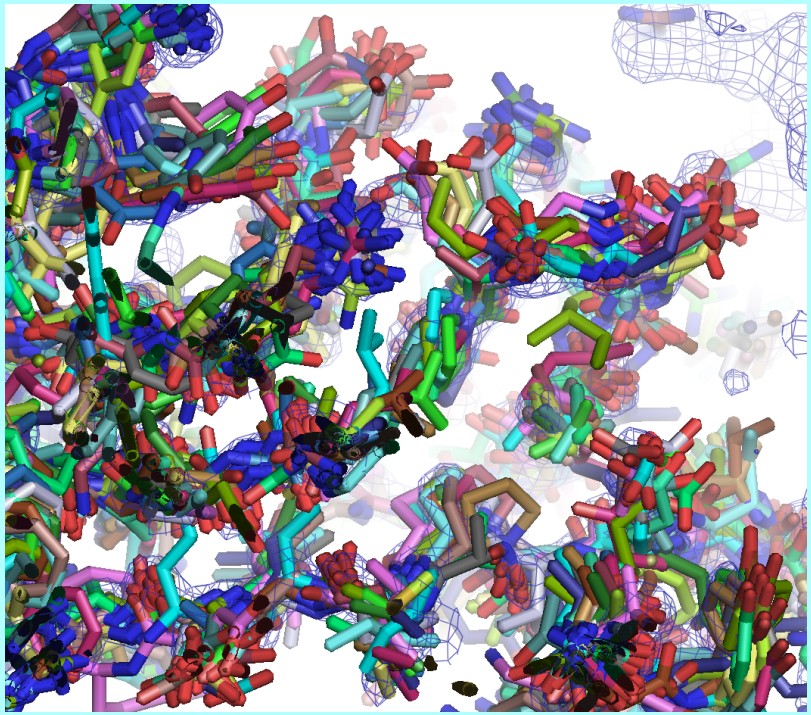


Building 20 models for each of 10 structures

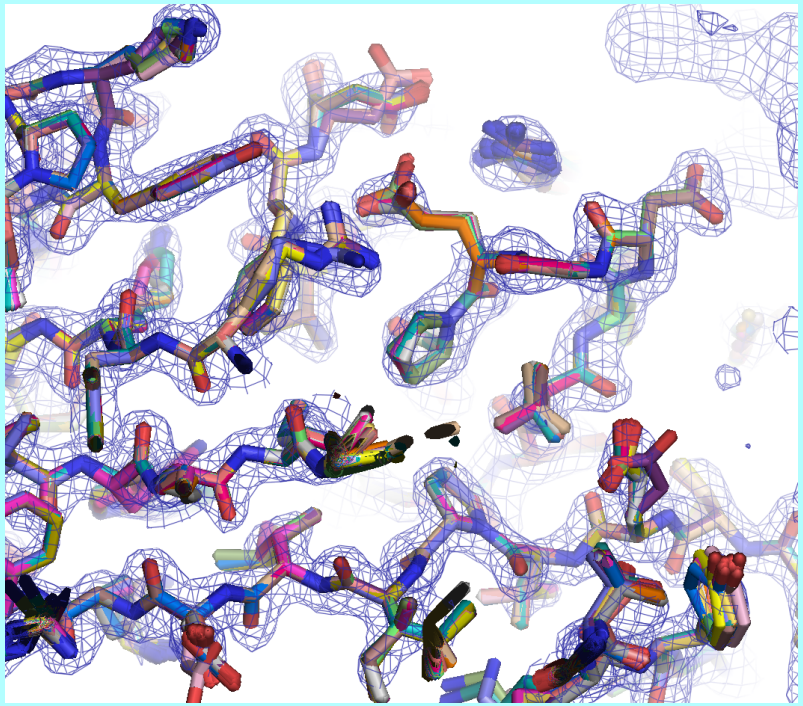


->The RMSD among models tells us (a lower bound on) the uncertainty in our models

(It is not the RMSD of true structures in the crystal)



Rebuild with 4.5 Å data



Rebuild with 1.75 Å data

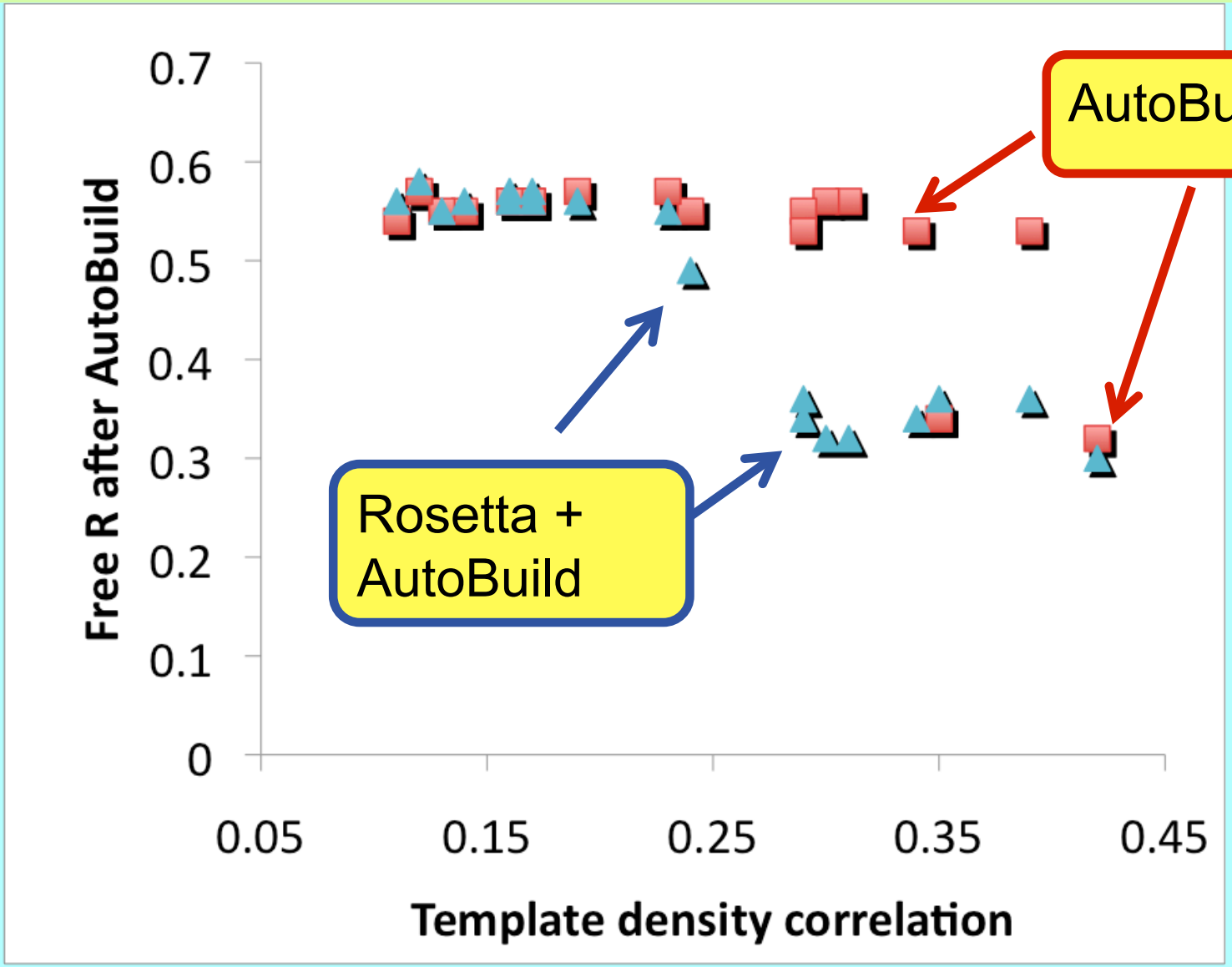
Complementarity of PHENIX and Rosetta model-building

(Randy Read, David Baker, Frank DiMaio)

Characteristic	Crystallographic model-building (PHENIX)	Structure-modeling (Rosetta)
Optimization	Interpretation of patterns of density	Creating physically plausible models
Model-building approach	Density search for regular secondary structure	Ab-initio modeling or homology modeling
Fragment libraries	3-residue fragment library	3- and 9-residue libraries
Model-building target	Fit to density	Rosetta force field with density term
Refinement target	Structure-factor likelihood refinement target	Rosetta force field with density term

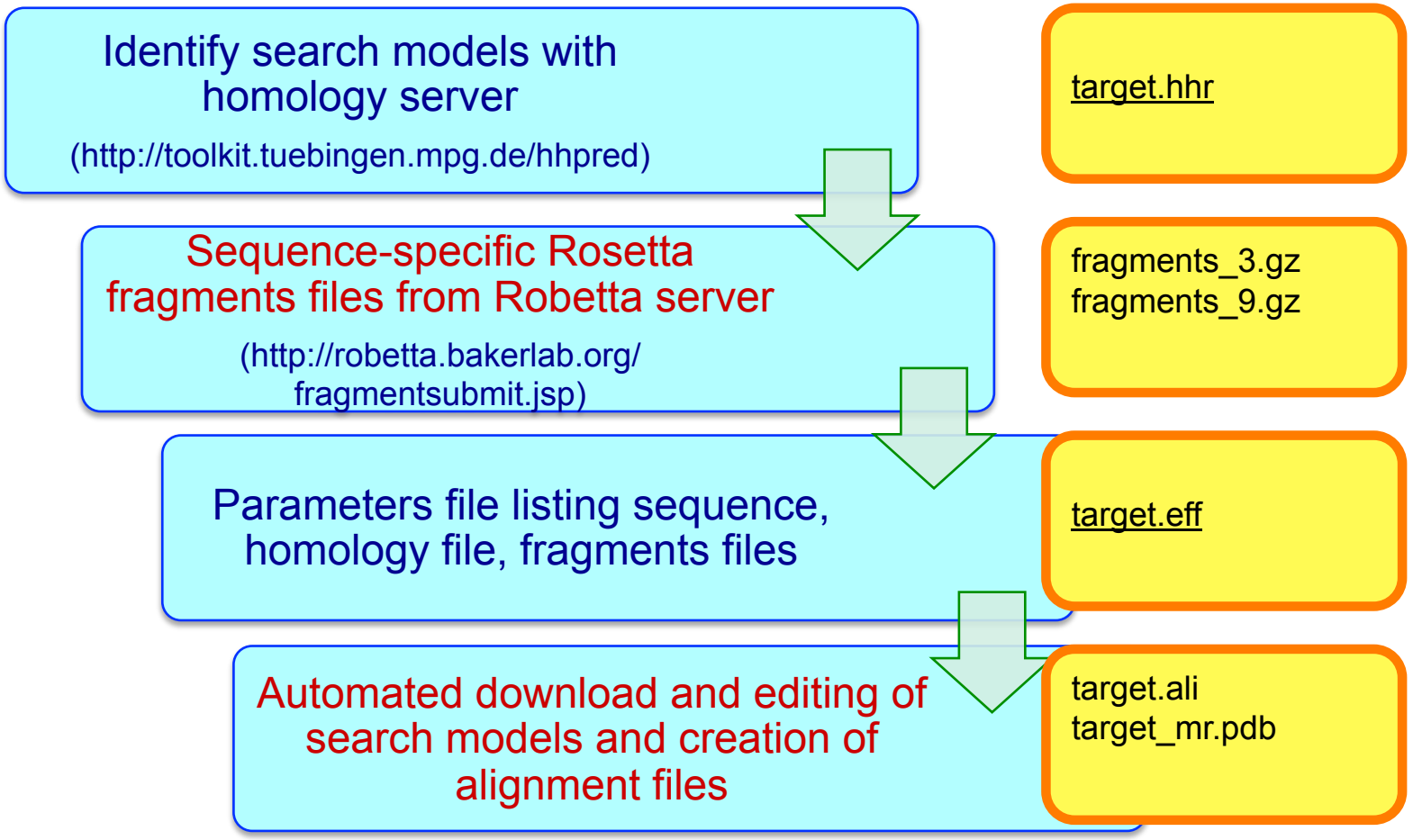
Combining structure-modeling with crystallographic model-building

20 templates for 1XVQ from PDB (optimally superimposed)



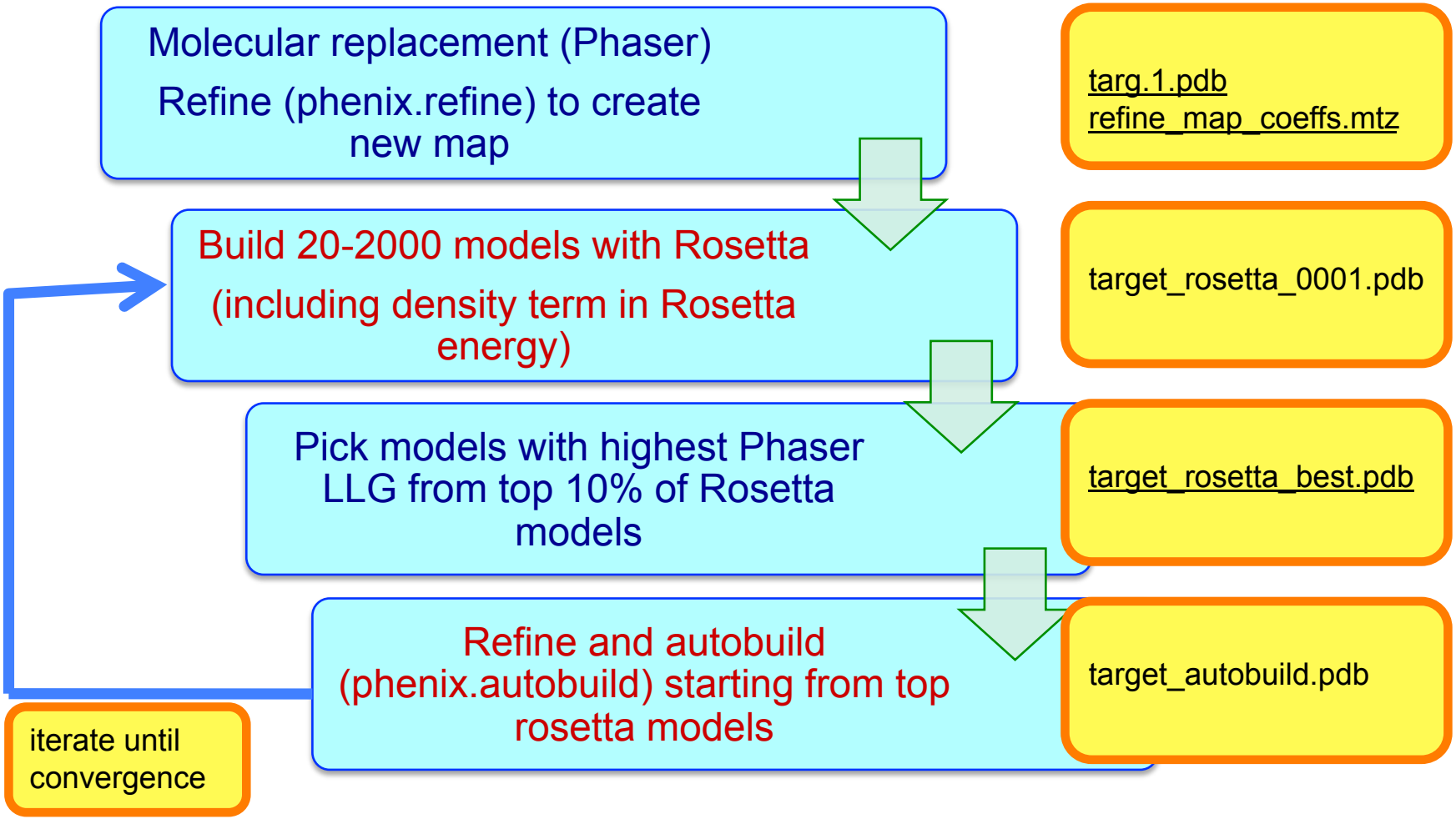
Molecular replacement using distant homology models with PHENIX and Rosetta (phenix.mr_rosetta)

Setup and model preparation



Molecular replacement using distant homology models with PHENIX and Rosetta (phenix.mr_rosetta)

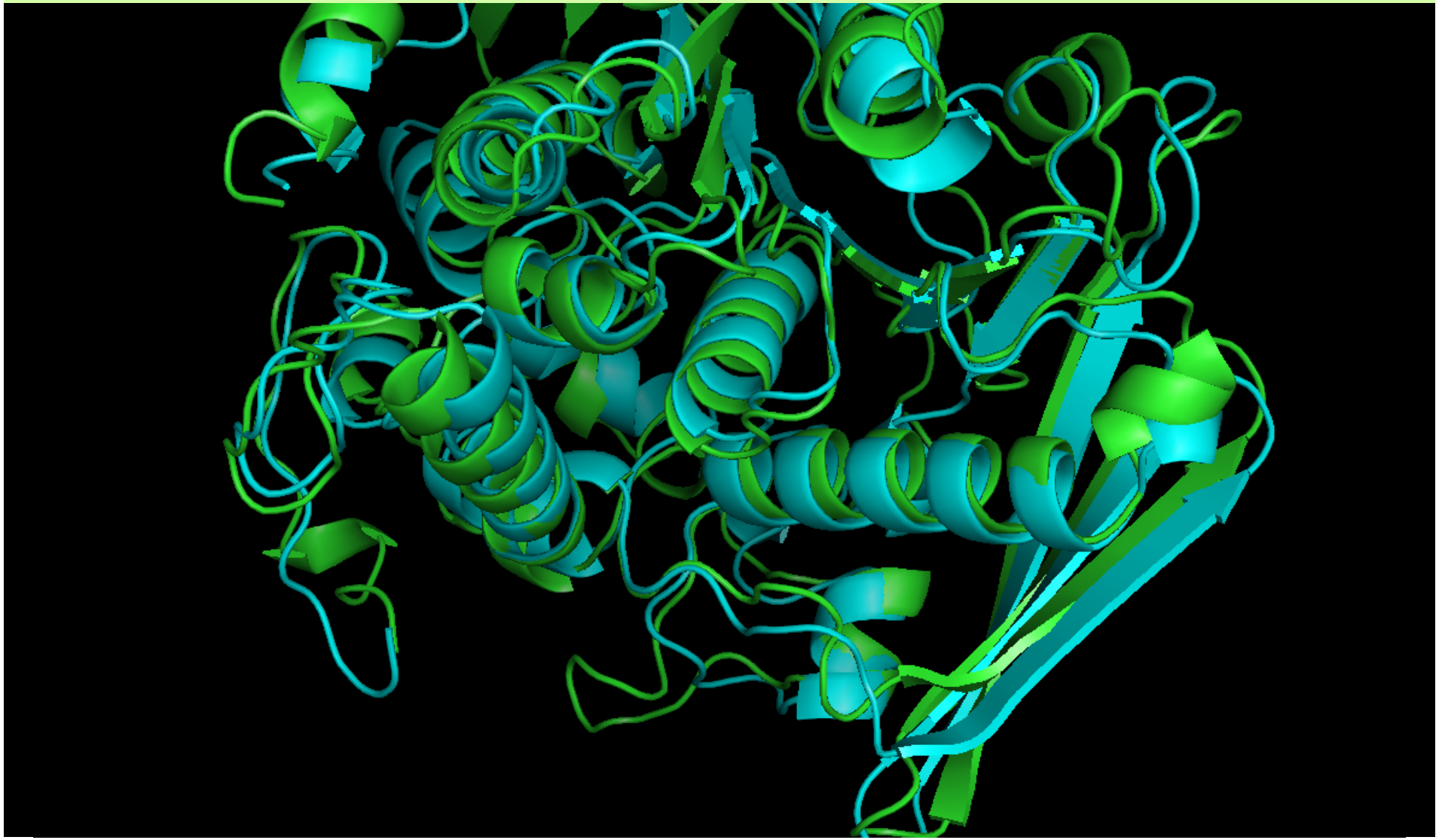
Molecular replacement and model-building



Structure determination of cab55348 (using template supplied by user)

1.9 A, 28% sequence identity (AutoMR alone fails with R/Rfree=0.47/0.53)

MR model: blue, Final model: green

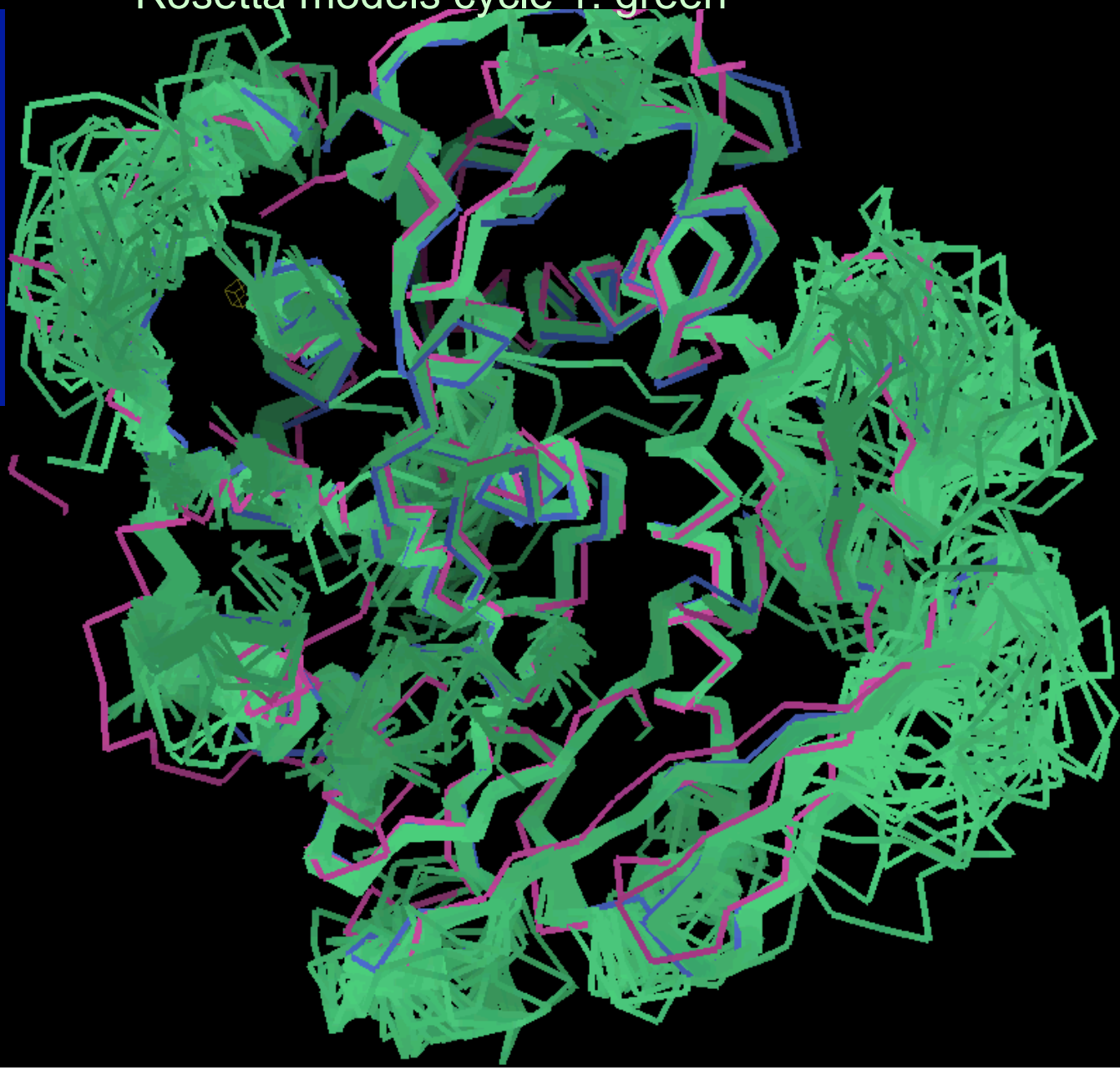


MR model : blue

Final model: pink

Rosetta models cycle 1: green

Sample Rosetta models in cycles 1 and 2,

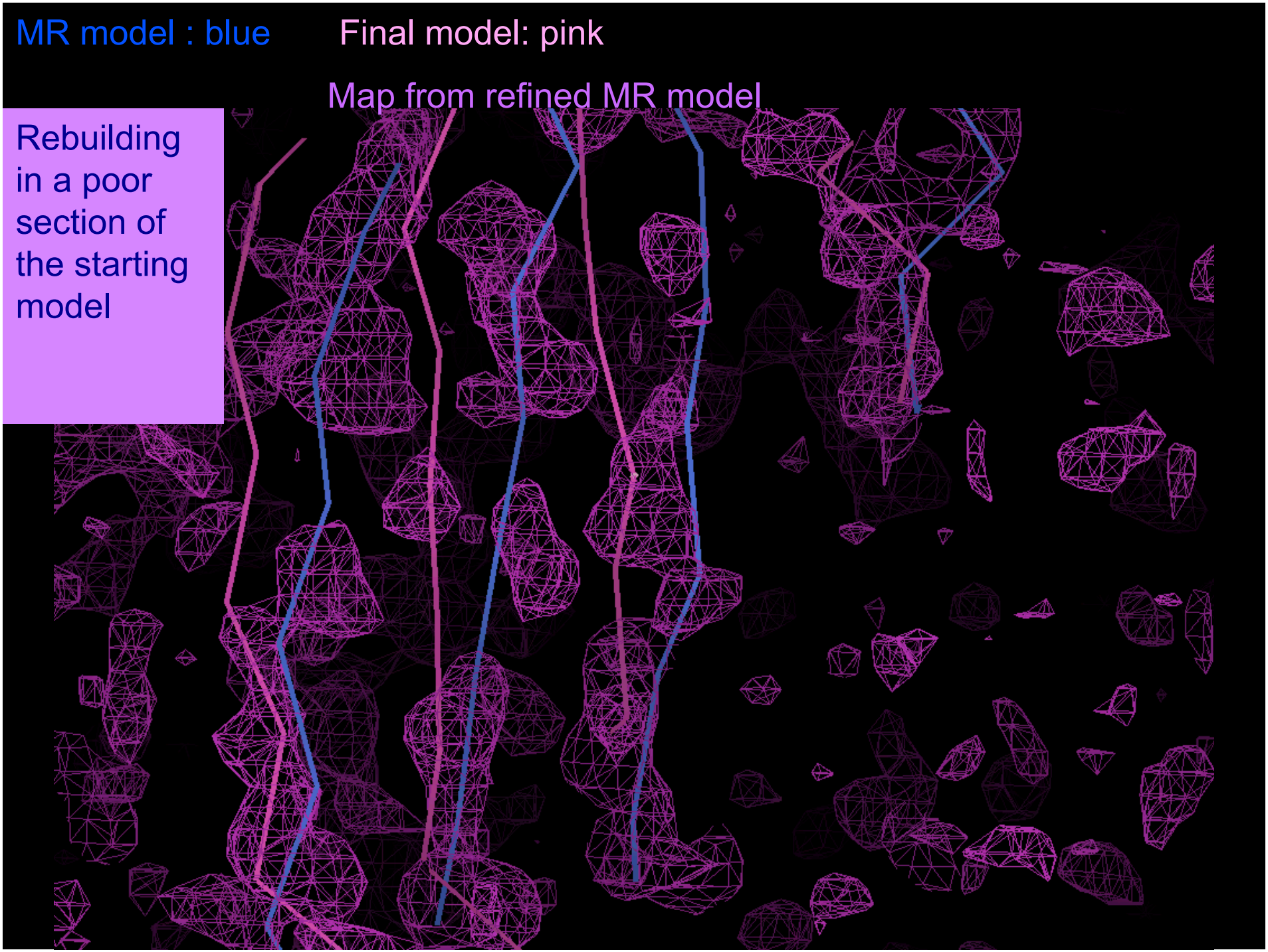


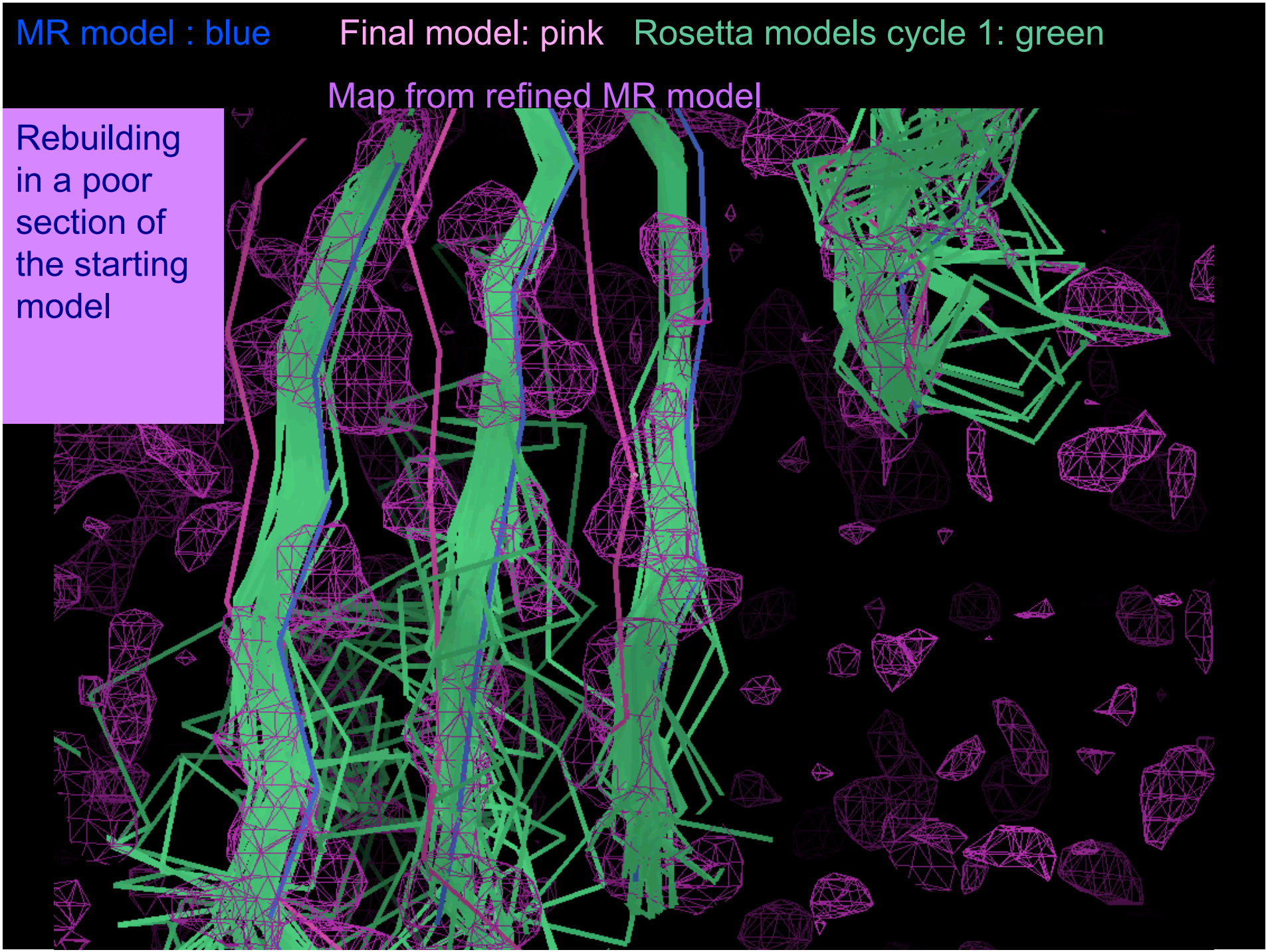
MR model : blue

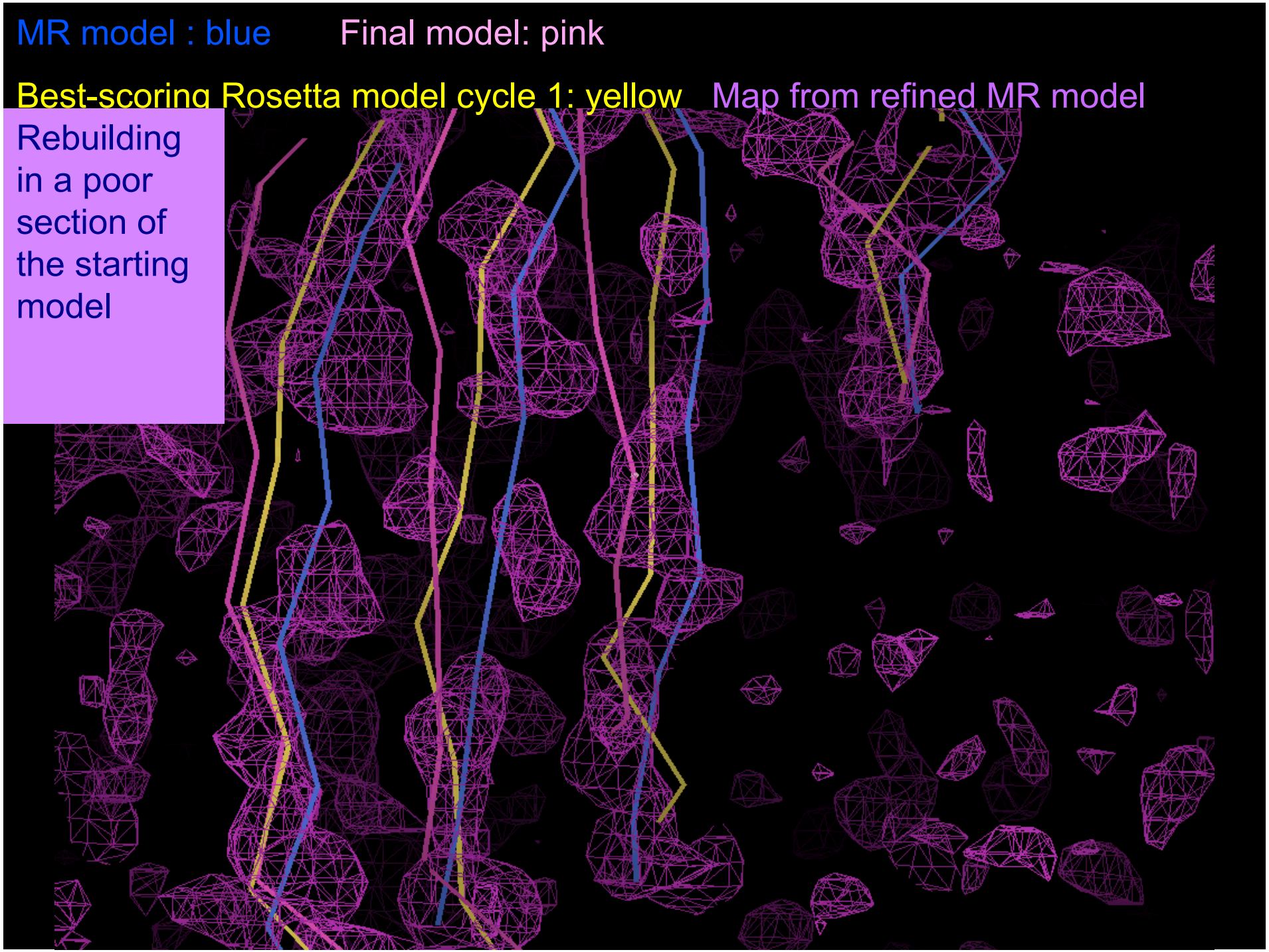
Final model: pink

Map from refined MR model

Rebuilding in a poor section of the starting model



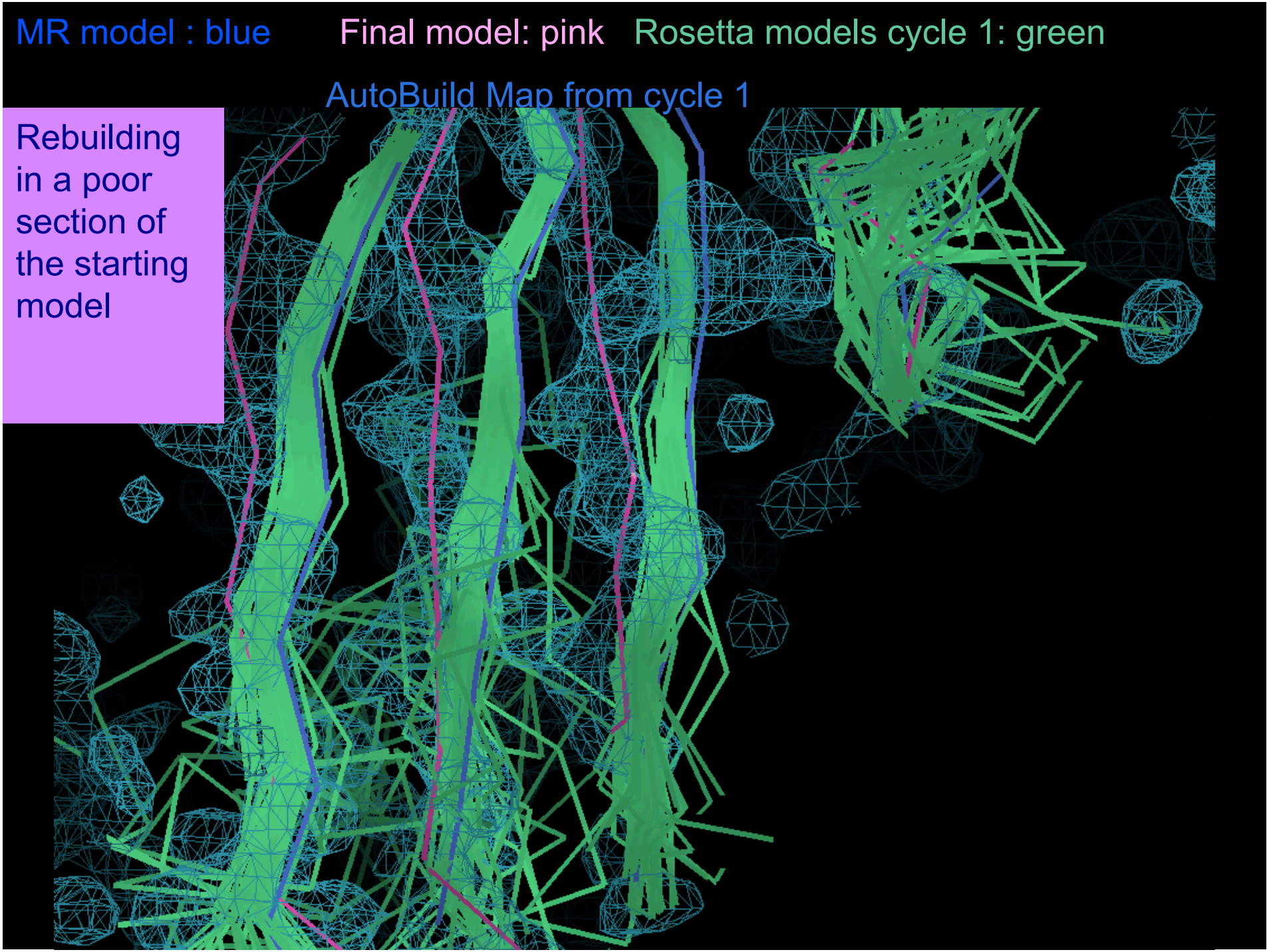

MR model : blue
Final model: pink
Rosetta models cycle 1: green
Map from refined MR model
Rebuilding in a poor section of the starting model


MR model : blue
Final model: pink
Best-scoring Rosetta model cycle 1: yellow
Map from refined MR model
Rebuilding in a poor section of the starting model

MR model : blue Final model: pink Rosetta models cycle 1: green

AutoBuild Map from cycle 1

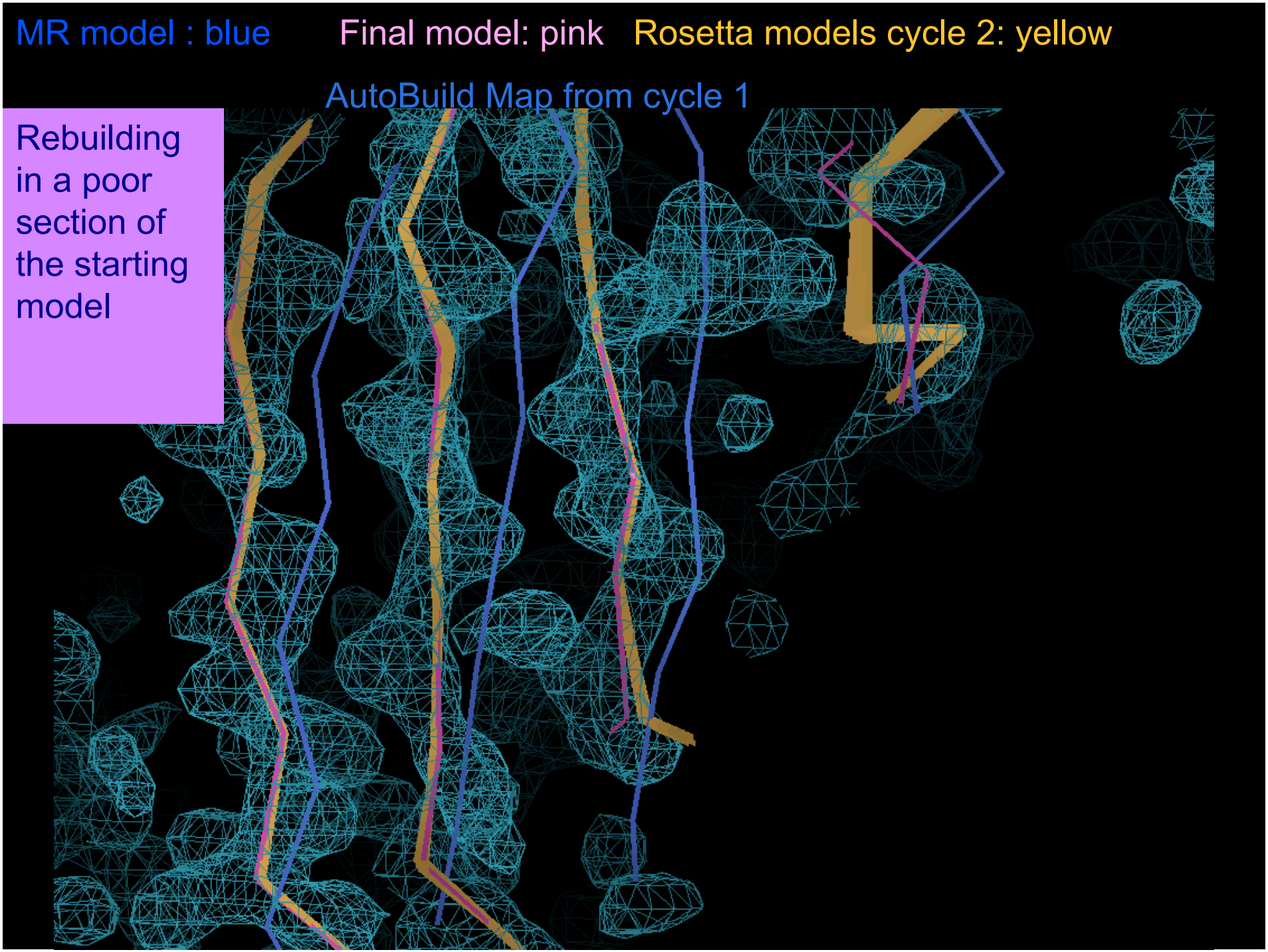
Rebuilding in a poor section of the starting model



MR model : blue Final model: pink Rosetta models cycle 2: yellow

AutoBuild Map from cycle 1

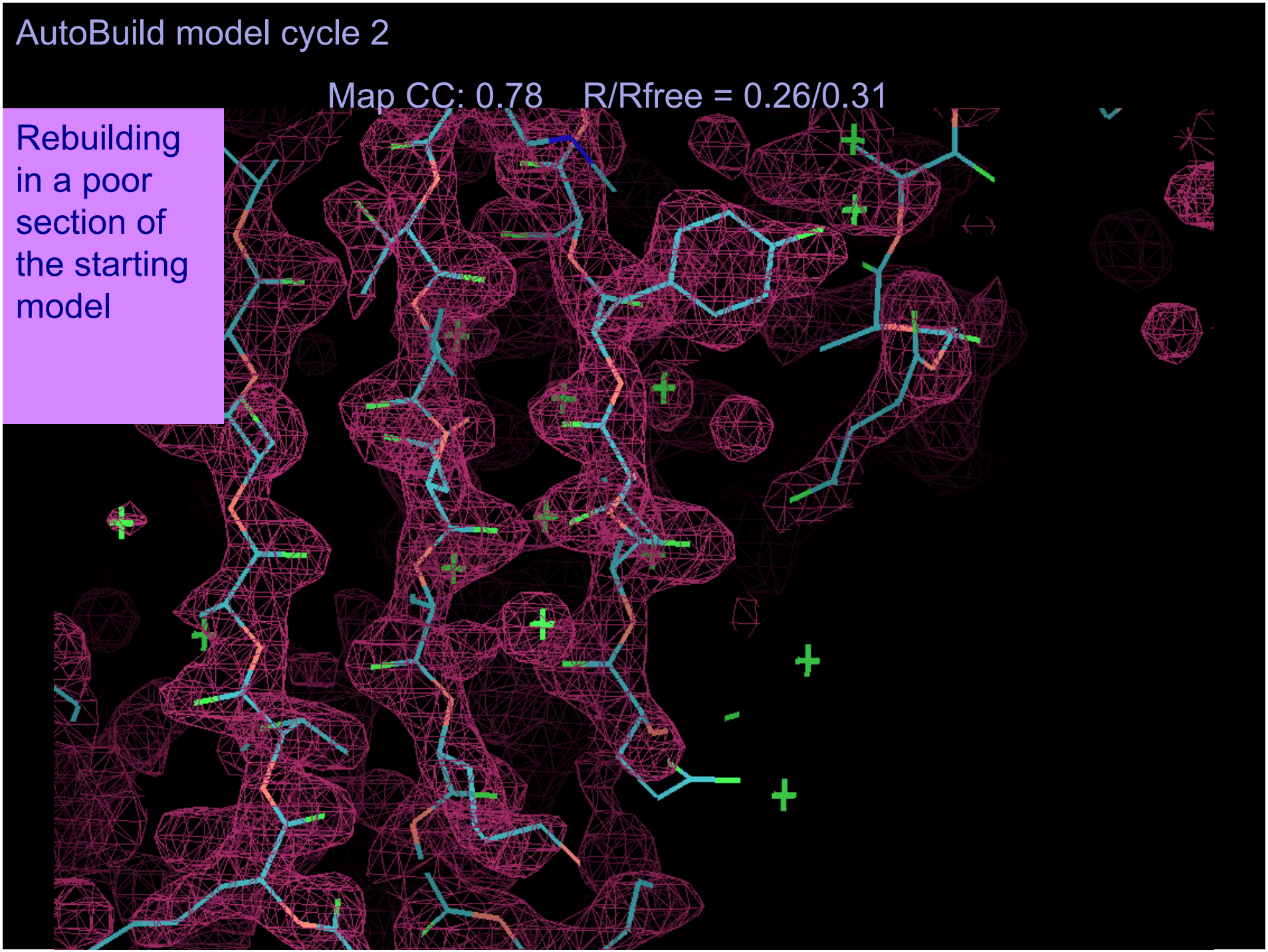
Rebuilding in a poor section of the starting model



AutoBuild model cycle 2

Map CC: 0.78 R/Rfree = 0.26/0.31

Rebuilding in a poor section of the starting model



Wizards

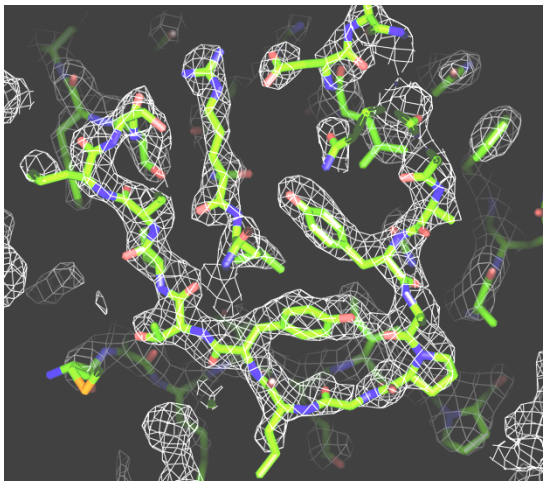
Phenix

•AutoSol Wizard: Structure solution (MIR/MAD/SAD) with HYSS/Phaser/Solve/Resolve

•AutoBuild Wizard: Iterative density modification, model-building and refinement with Resolve/phenix.refine/Elbow; model rebuilding in place; touch-up of model; simple OMIT; SA-OMIT; Iterative-build OMIT; OMIT around atoms in a PDB file; protein, RNA, DNA model-building

•LigandFit Wizard: automated fitting of flexible ligands

•AutoMR Wizard: Phaser molecular replacement followed by automatic rebuilding



MODEL-BUILDING TOOLS

Phenix

- phenix.find_ncs: Find and evaluate NCS from density, heavy-atom sites, or model
- phenix.apply_ncs: Apply NCS operators to a single chain
- phenix.build_one_model: Resolve rapid model-building with real-space refinement
- phenix.phase_and_build: Improve map by model-building and refinement, then build full model
- phenix.find_helices_strands: Trace chain or build secondary structure from a map
- phenix.mr_rosetta: Combine Rosetta structure-modeling with Phenix

REFINEMENT AND TOOLS

Phenix

- phenix.refine: fully automatic/fully flexible refinement, SA-refinement, NCS identification, TLS, torsion-angle refinement, twin refinement

- phenix.xtriage: twinning, twin laws, anisotropy, anomalous signal, outliers, space group

- phenix.builder: ligand structures and CIF definitions from SMILES, PDB....

- phenix.ligand_identification: identify ligand density with class-specific libraries

- phenix.validation, phenix.model_vs_data, phenix.real_space_correlation, phenix.get_cc_mtz_mtz: Molprobity and density analysis of structures and density maps

- phenix.pdbtools, phenix.reflection_file_editor: manipulate PDB and mtz files

- ...and many more: see phenix.doc and **www.phenix-online.org**

The PHENIX Project

Phenix

Lawrence Berkeley Laboratory

Paul Adams, Ralf Grosse-Kunstleve, Pavel Afonine, Nat Echols, Nigel Moriarty, Jeff Headd, Nicholas Sauter, Peter Zwart

Los Alamos National Laboratory

Tom Terwilliger, Li-Wei Hung

Cambridge University

Randy Read, Airlie McCoy, Gabor Bunkoczi, Rob Oeffner

Duke University

Jane & David Richardson, Vincent Chen, Chris Williams, Bryan Arendall, Laura Murray

An NIH/NIGMS funded Program Project