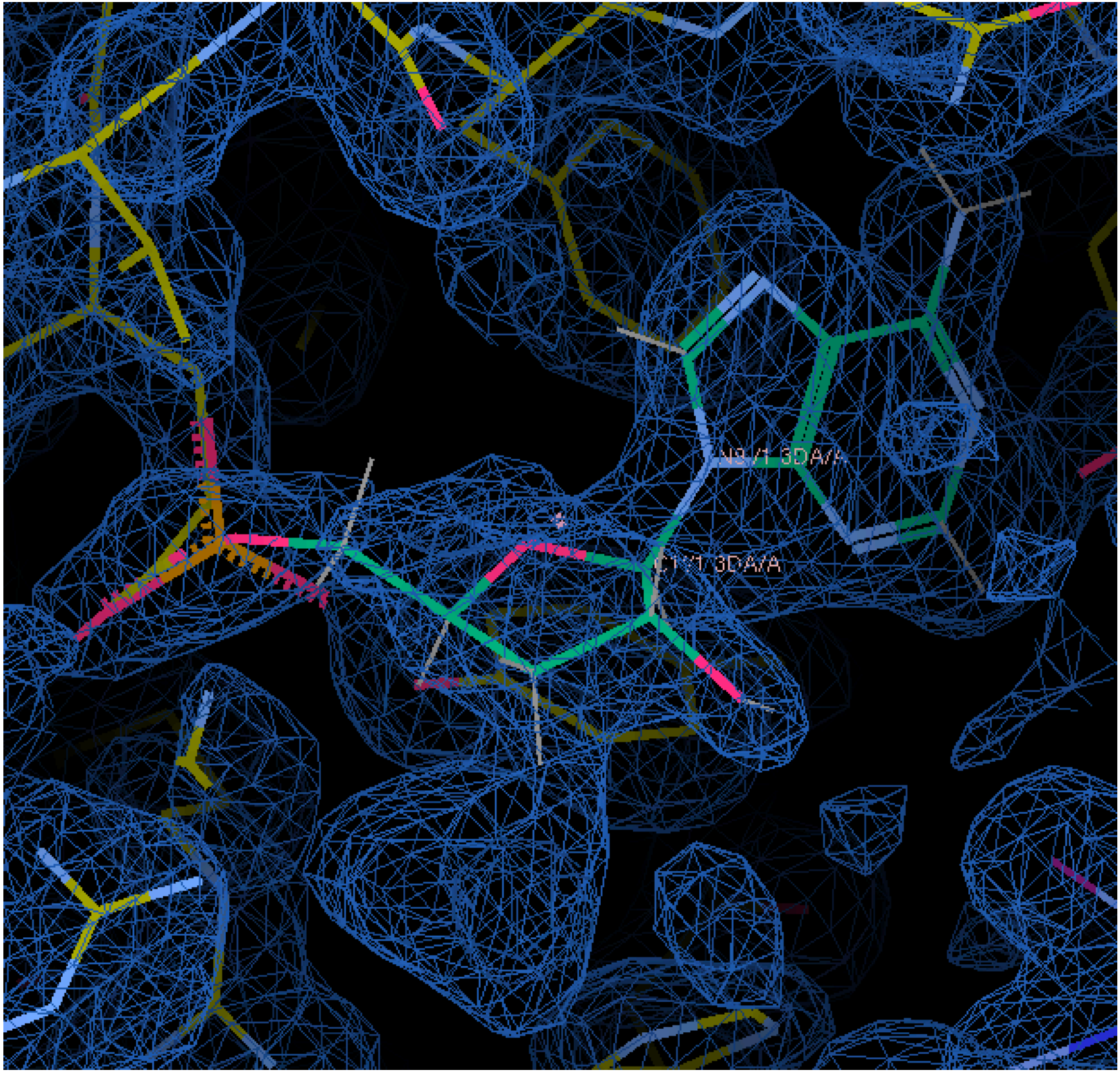
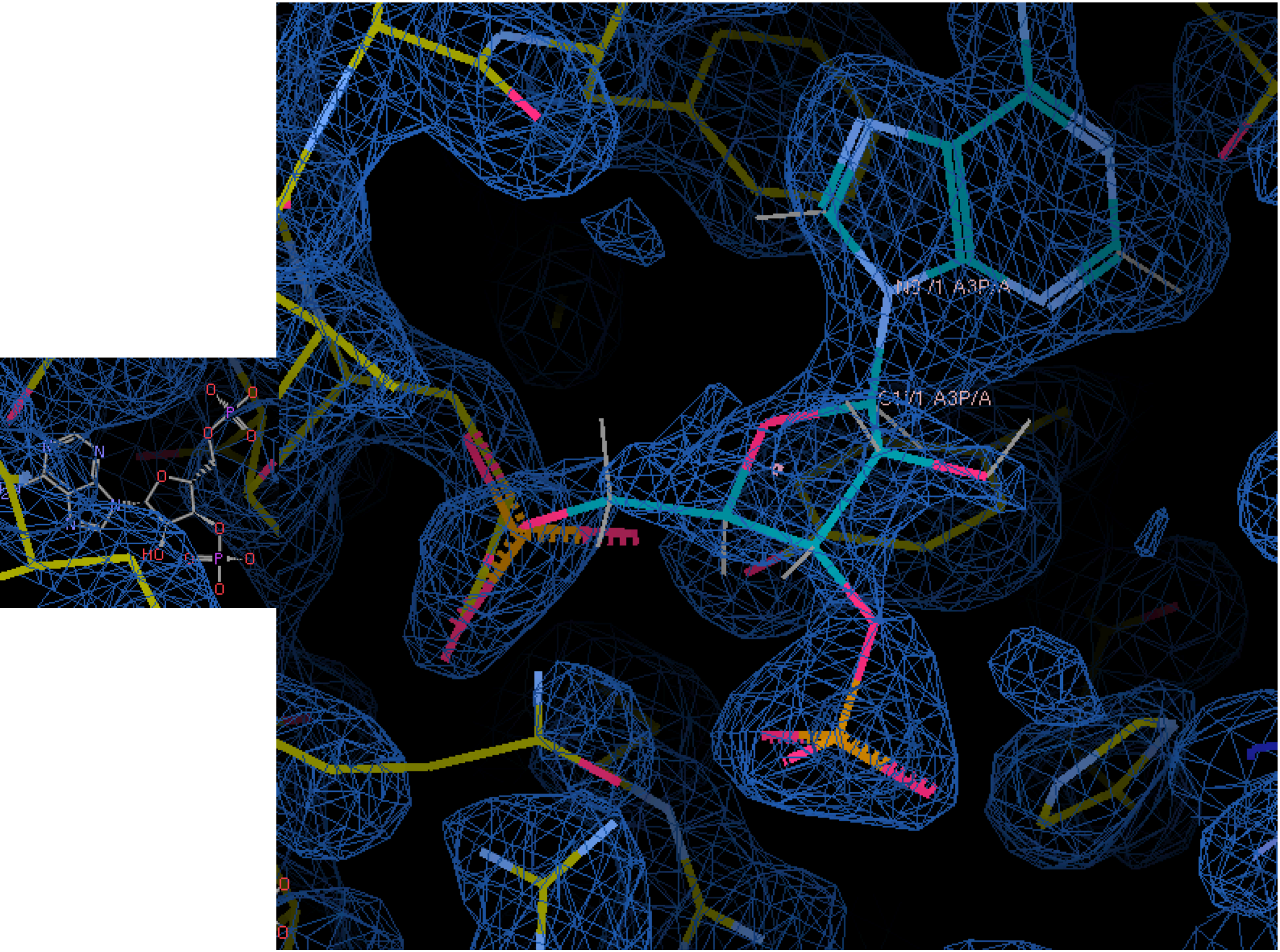
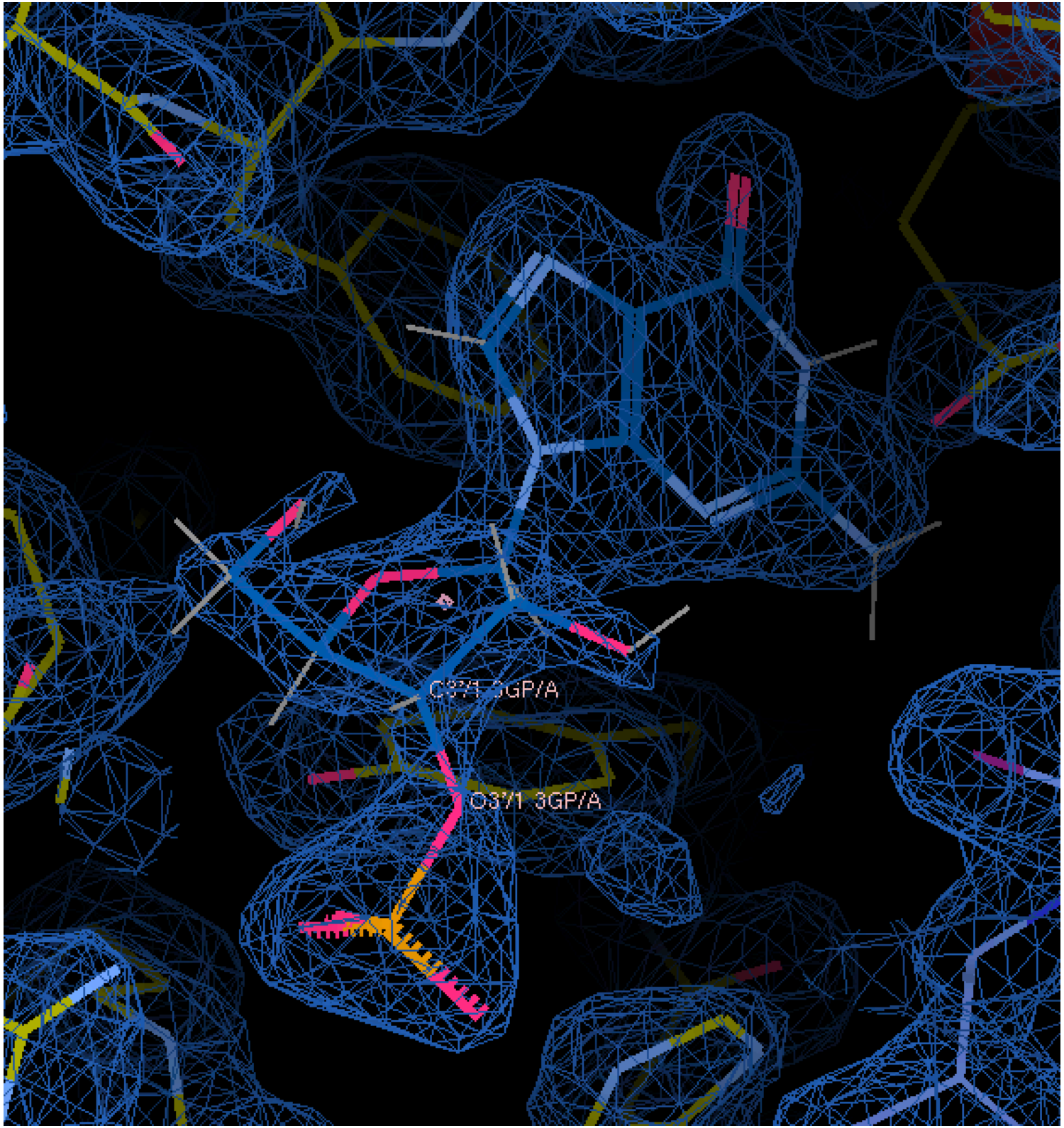
# Protein Structure Determination '20 -- Lecture 10

In class exercises:
Ramachandran plot
Superposition
Crystal contacts


N9 /1 3DA/A
C1 /1 3DA/A


A3P/A
A3P/A


C3'/1 3GP/A
O3'/1 3GP/A

# Model Validation

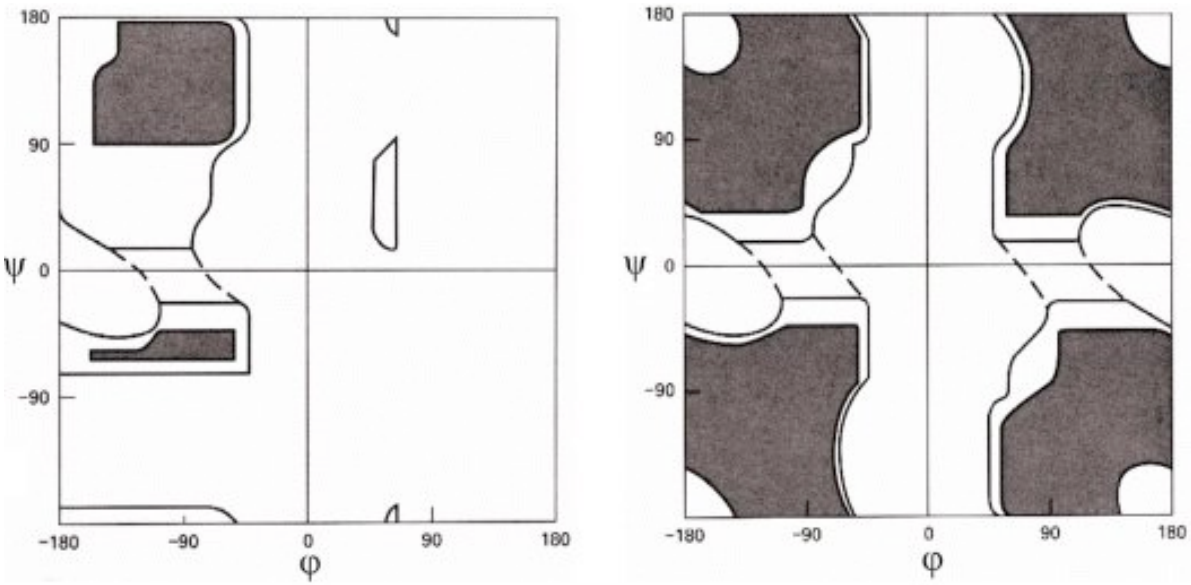
# Model quality metrics

- Resolution
- R-factor (free R-factor)
- Overall B-factor (approx Wilson B)
- Quality of density (holes in rings, H bumps)
- Ramachandran plot
- Rotamers
- other stereochemistry (bond lengths, angles, chirality)
- B-factors
- Rsym, completeness (measure quality of data, not of model, but related)

# Ramachandran Plot matches stats of good Xray structures

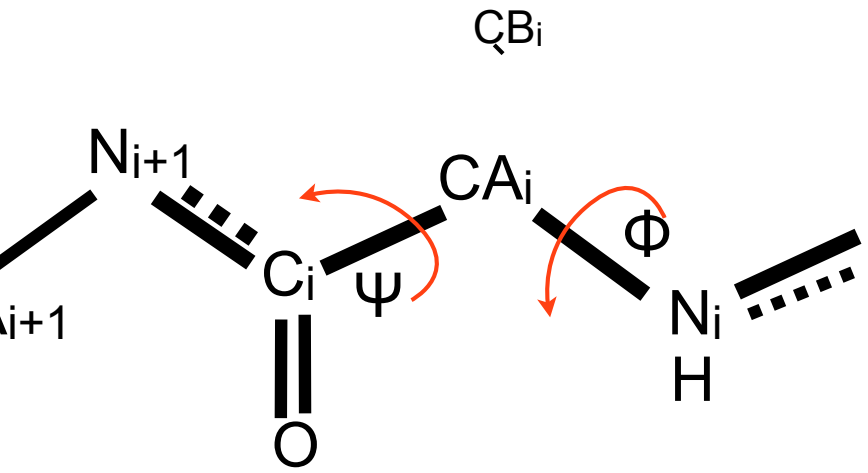
Ramachandran used a physical model of dipeptides to determine the allowed (dark) and disallowed (white)combinations of phi and psi backbone angles.
The observed frequencies roughly agree with R's allowed regions.

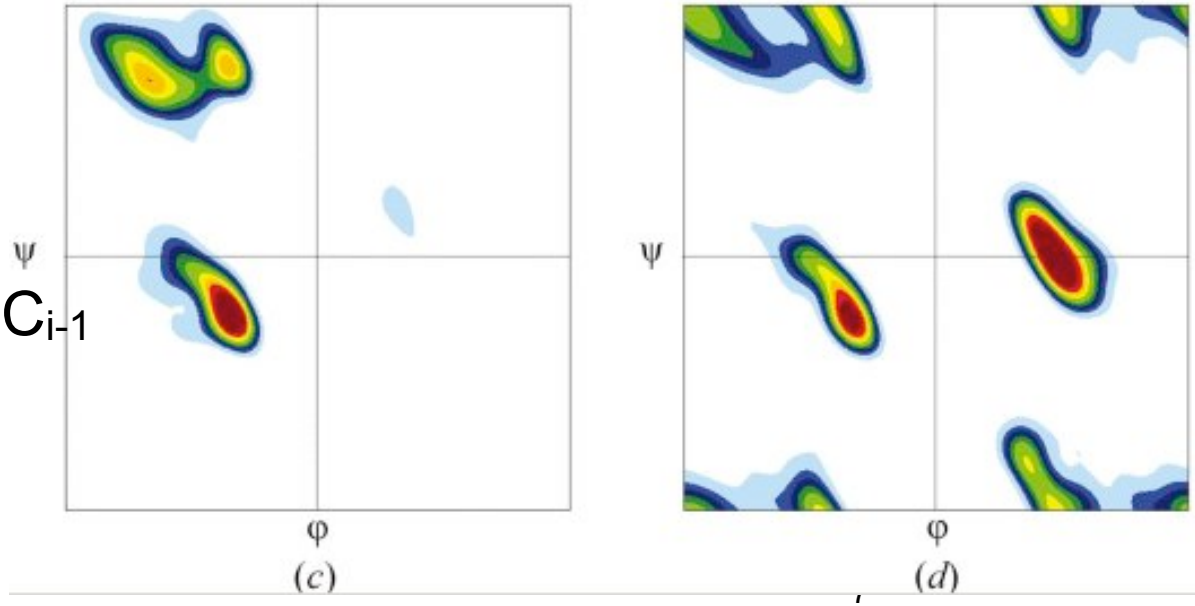
Ramachandran & Sasisekharan (1968)




non-glycine, non-proline

glycine

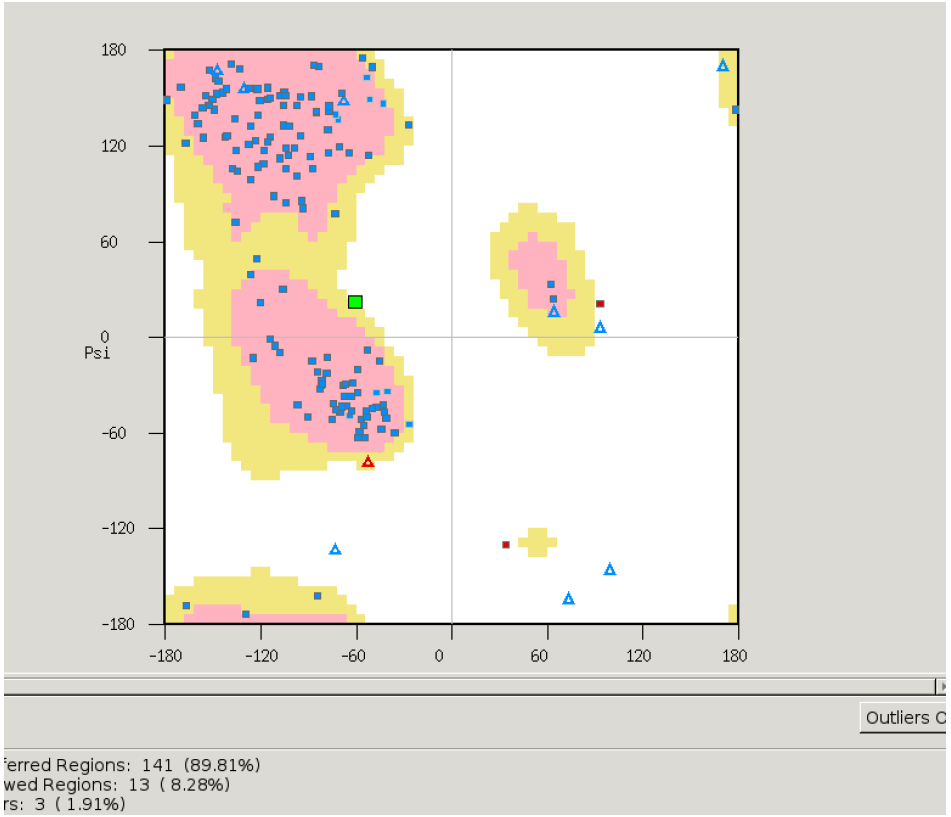


# Ramachandran plot in Coot

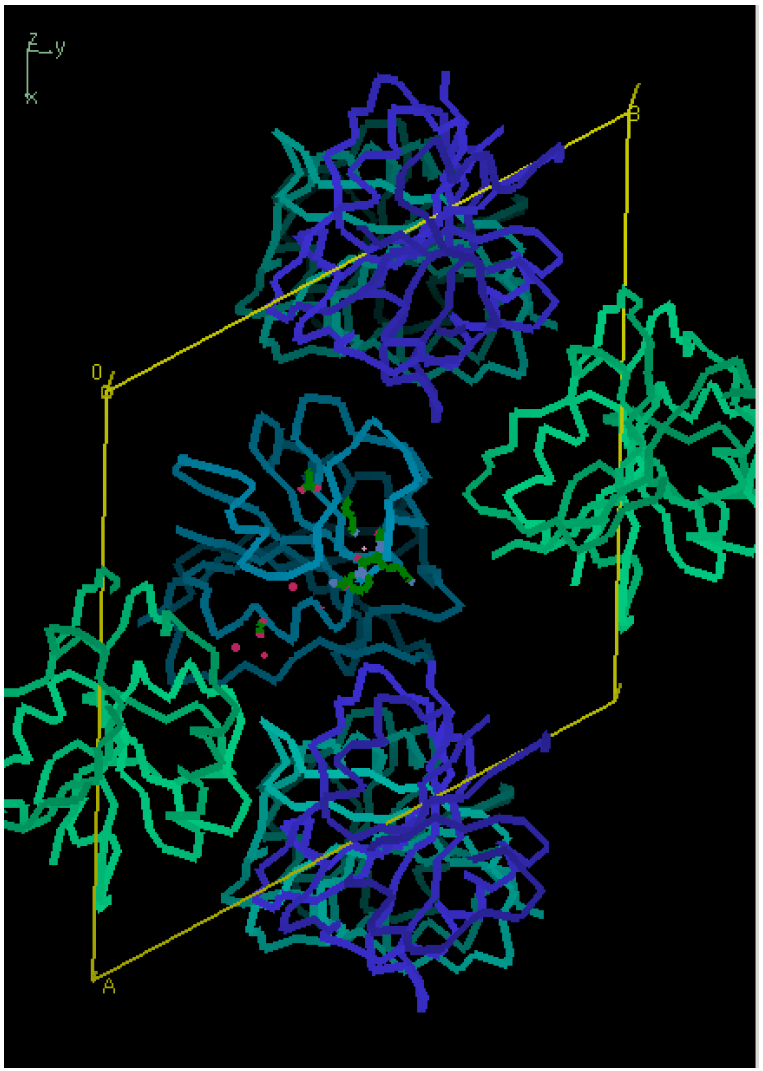
Read in **7dfr.pdb**
Validate > Ramachandran plot

- Click to identify residue
- Plot changes to match residue type
- Helps to identify outliers, poorly modeled residues
- Find a glycine.
- Find a proline.



# Showing symmetry in Coot

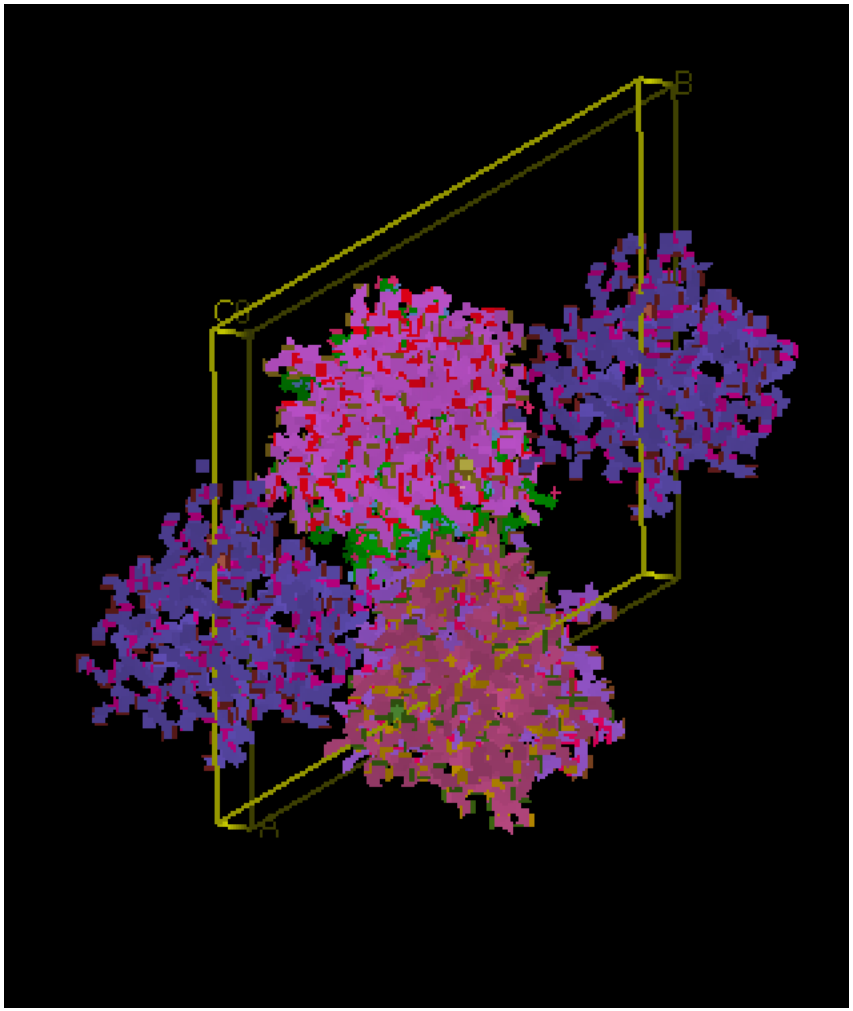
Draw > Cell & Symmetry
- Master switch: Yes
- Show unit cells: Yes
- Symmetry by molecule:
  - Display as CAs, Color by symop

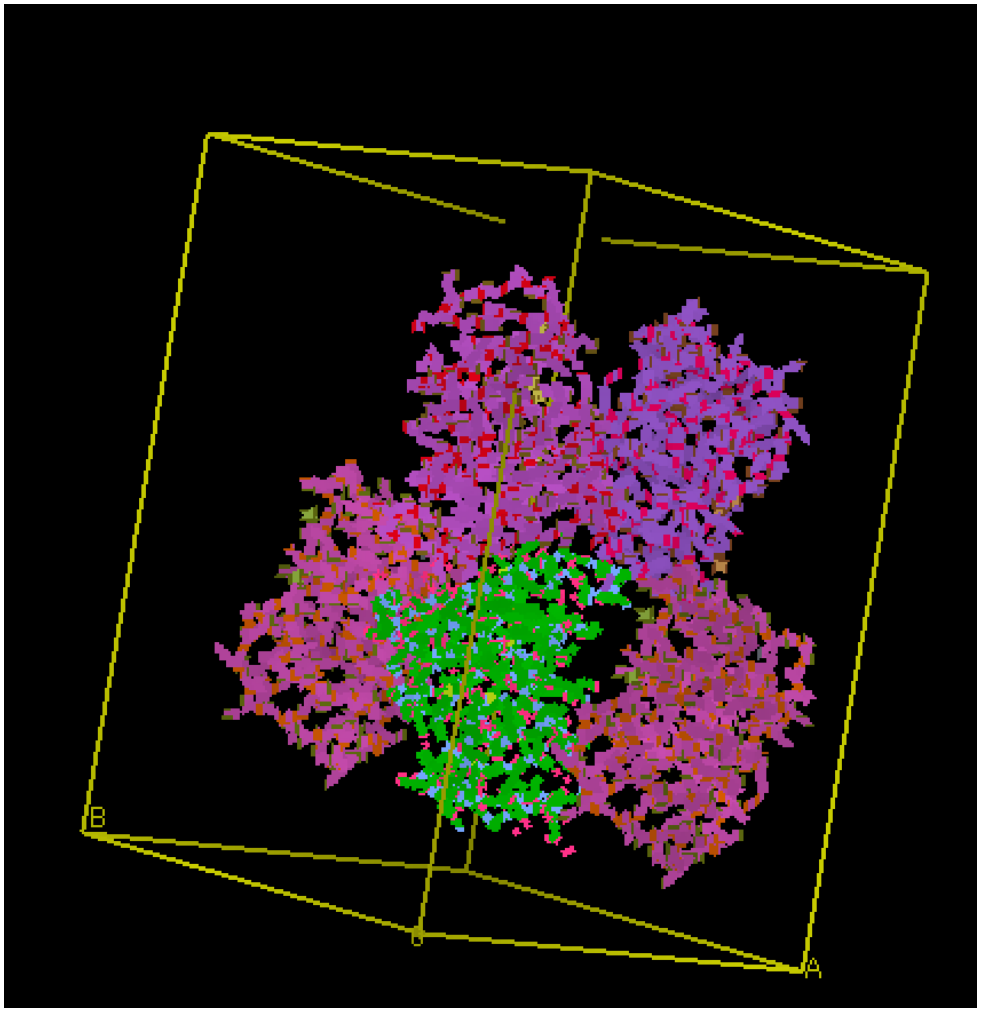


Draw > Cell & Symmetry

Symmetry by molecule:

Display near chain, Color by symop





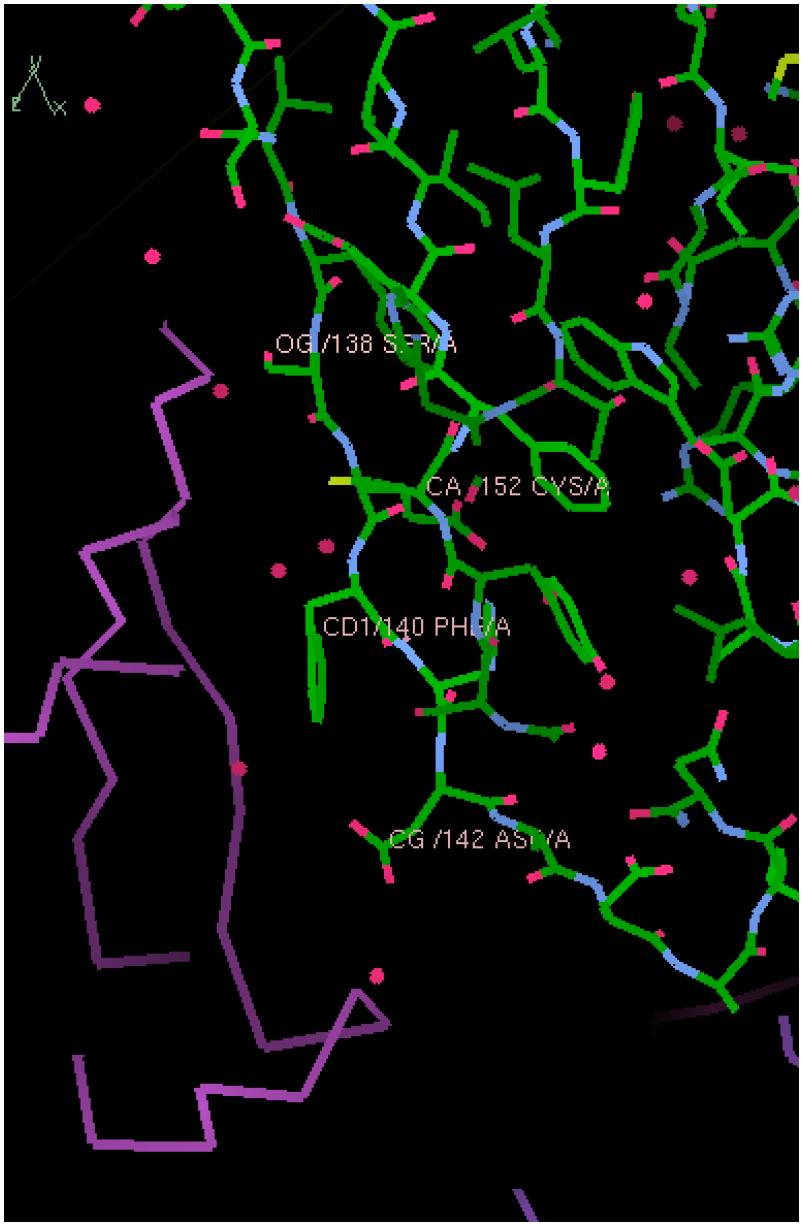
**trigonal space group!**

# Find crystal contacts

Crystal contacts are locations where atoms are close to atoms of a symmetry-related molecule. (Can be non-crystallographic symmetry)

Play a role in crystal formation.

May be distorted by the interaction energy.




Click to label atoms in crystal contact

S138, C152, F140, D142

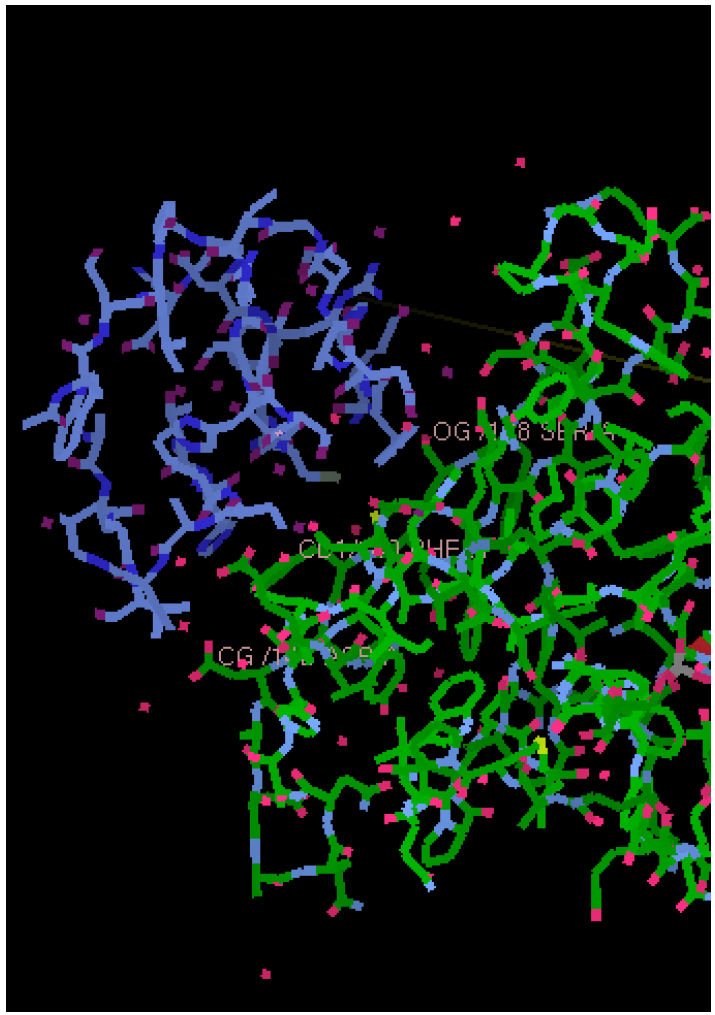
# Find crystal contacts

Draw > Cell & Symmetry

Symmetry by molecule:

Display sphere, standard colors

Use control-leftmouse-drag to pan.



# Superposing two molecules in Coot

- **Superposition**
  - ...requires a sequence alignment.
  - ...solves for rotation/translation by least-squares

**least-squares superposition minimizes...**

$$\sqrt{\frac{SUM(\underline{M}r^{a}_{i} - r^{b}_{i})^{2}}{N}}$$

**... to solve for $\underline{M}$**

$r^{a}_{i}$ and $r^{b}_{i}$ are the coordinates of the $i$th residue (more precisely, the two residues in $i$th column of a sequence alignment) of molecules **a** and **b** respectively.

# Superposing two molecules in Coot

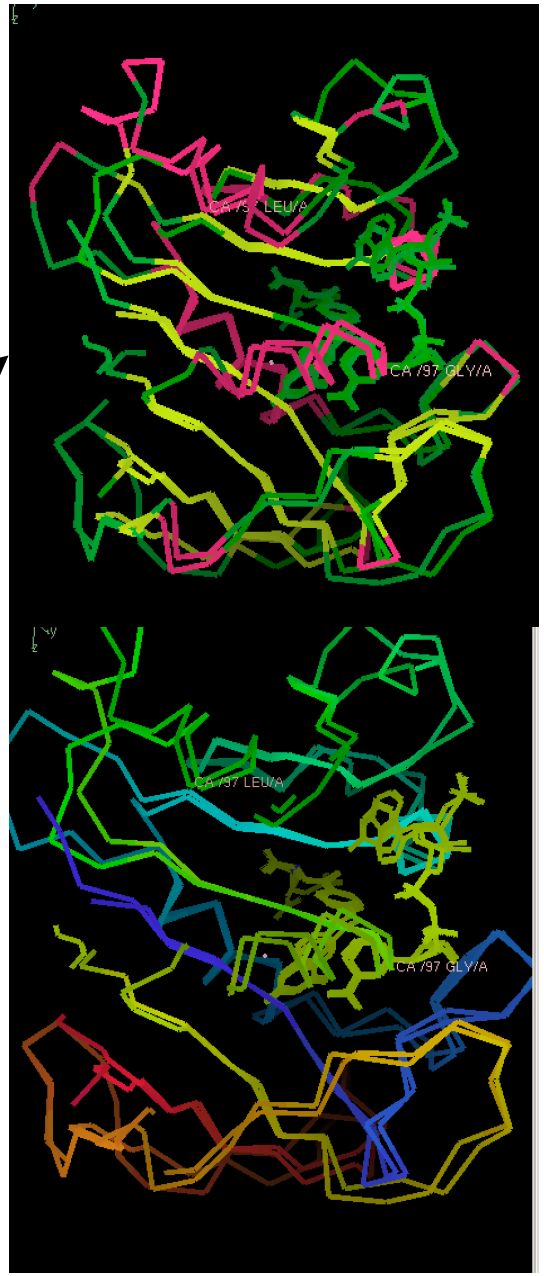
Read in **7dfr.pdb** (if not already)
Read in **4m6k.pdb**

**Calculate > SSM superpose**
select chain A for both

**Display manager**
**CAs+Ligs SecStr Col**

Later, try...

**Display manager**
**Jones Rainbow**



# Comparing models

- Questions you ask when comparing two models:
  - How similar?
  - What method was used? (Xray, NMR, cryoEM, homology)
  - Where are the differences?
  - How significant are the differences?
  - What may have caused the differences?
  - Any differences in function?

**metrics and factors**

- RMSD
- sequence differences, insertions/deletions
- ligands
- crystal contacts
- relative model quality (see previous slide)