

Bioinformatics 2 -- Lecture 8

Homology modeling:

Insertions.

Unalignment.

8.1 Insertions

How to insert 'by hand' in MOE

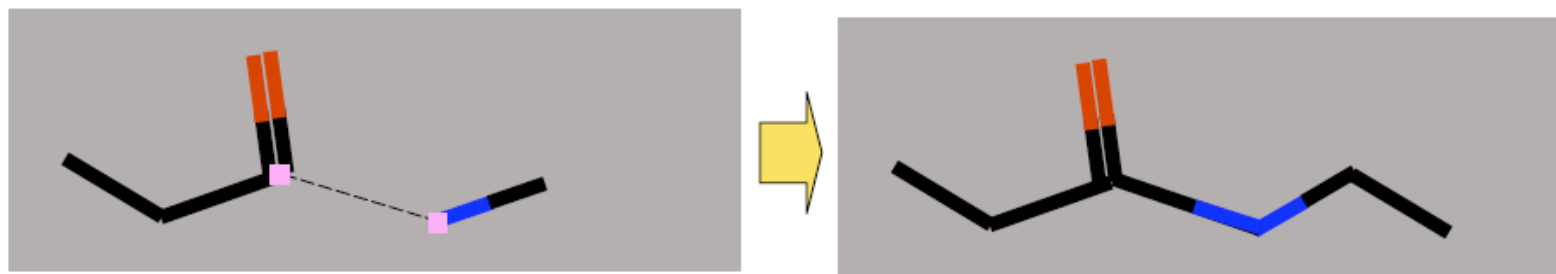
- Starting with a single chain structure, cut the chain where you want to insert the loop.
SEQ: select residue, **Edit | split chain**
- Create a loop or extract a loop from a PDB file (For example, by deleting all residues except the loop)
- Move the loop chain to be between the N-terminal and C-terminal chains. It should look like this:

1	ACDEFGHIKLMN
2 INSRTDSEQ
3 PQRSTUVWXYZ

- SEQ: select chains, **Edit | Join chains**
- Create new peptide bonds. Energy minimize.

Supplementary: How to make a new peptide bond

Zoom in on splice points.

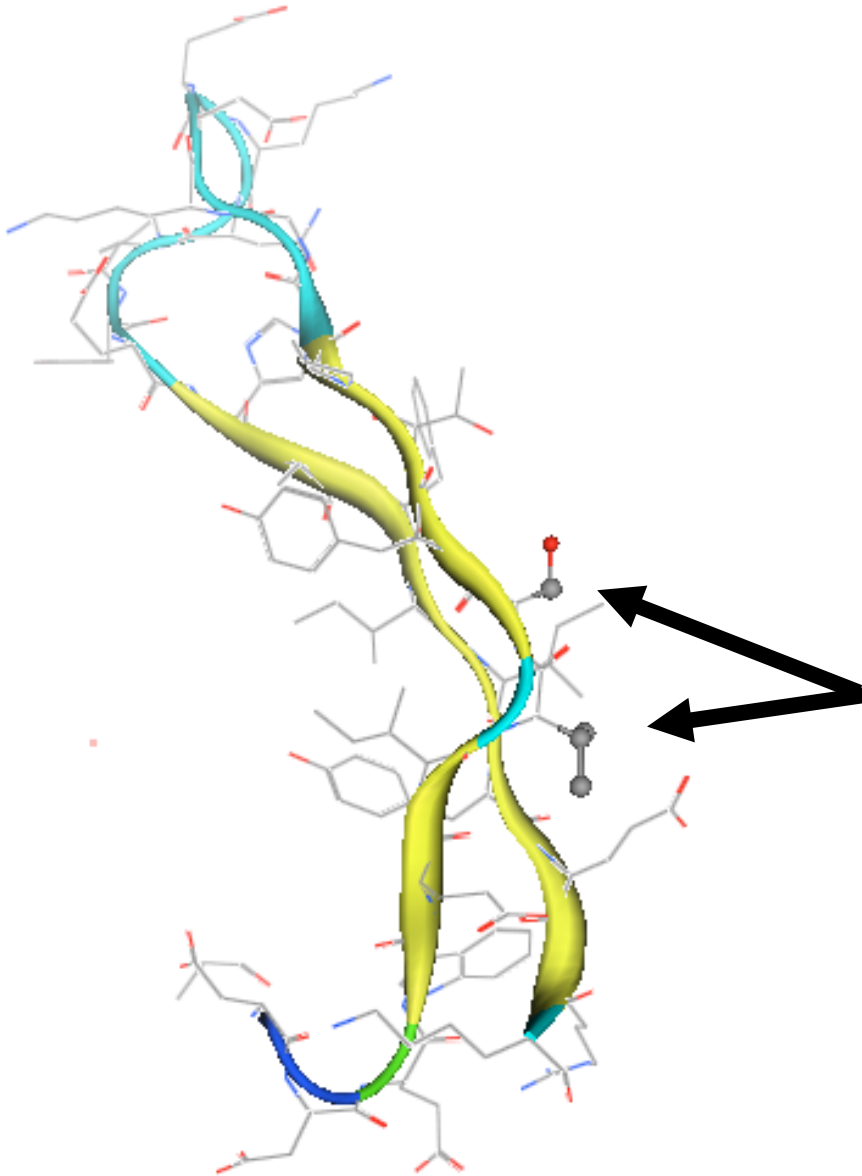


Delete extra oxygen, if present.
Select C, N. **Build | single bond**.
Select N. **Window | Atom manager** (or cntrl-a)
Select the atom. Set geometry to sp². Apply.

Rules for insertions, deletions

- **Rule 1.** Don't allow a deletion in the middle of a helix, even if the highest scoring alignment puts it at that position.
- **Rule 2.** Don't allow a deletion in the middle of a beta strand, even if the highest scoring alignment puts it at that position.
- **Rule 3.** Don't allow an insertion in the middle of a beta strand, unless there is room for a bulge or blow-out.
- **Rule 4.** Don't allow an insertion in the middle of a helix, unless you know it belongs there.

An exception to the *no-insertions-in-strand* rule



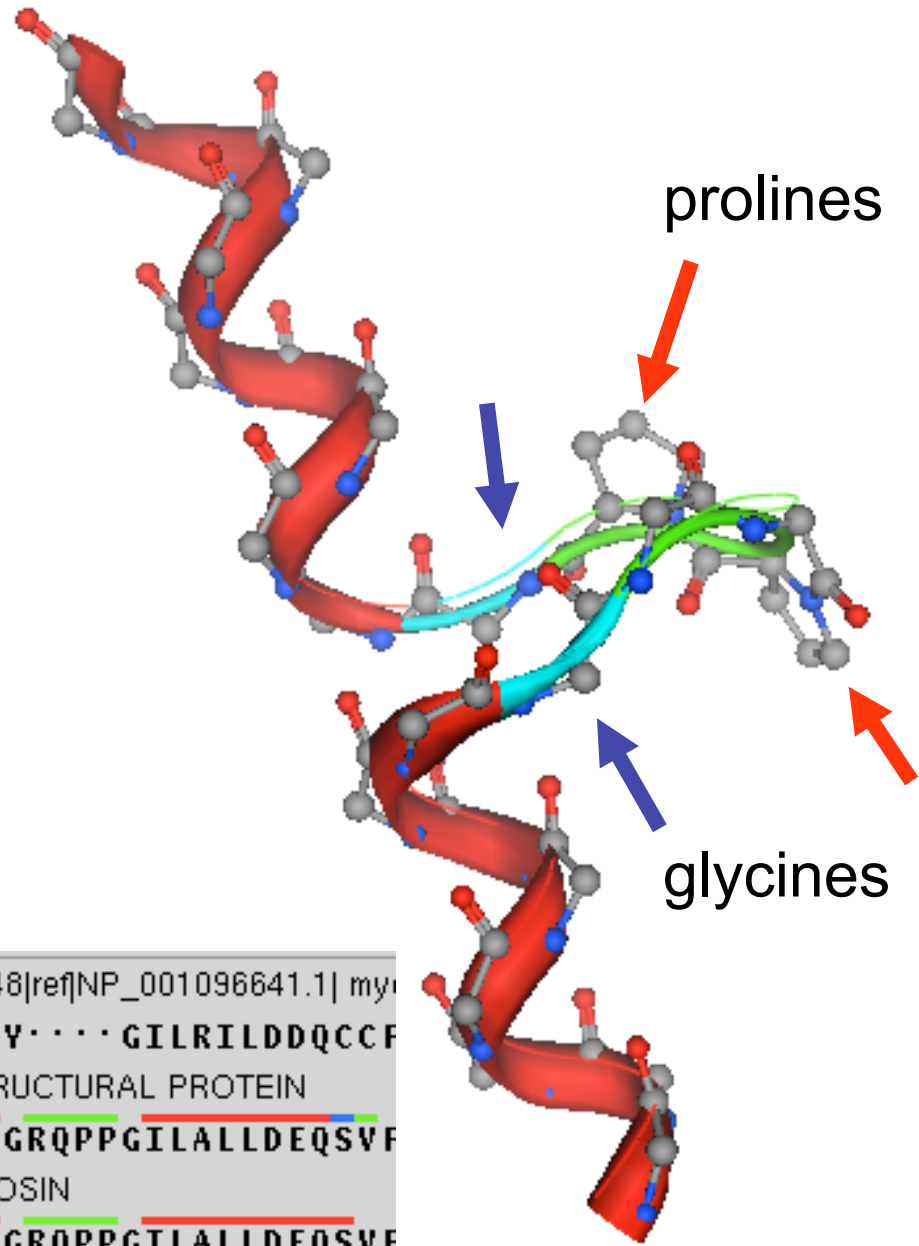
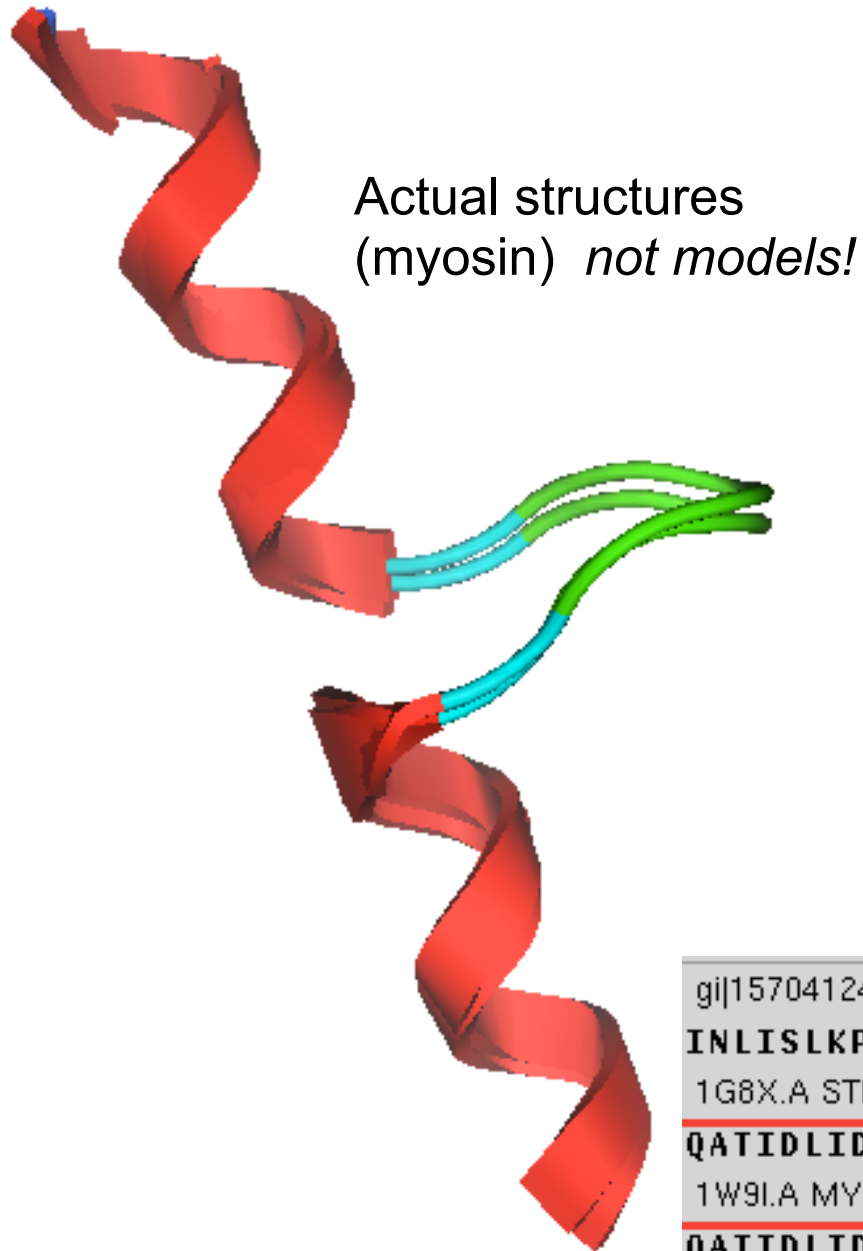
Beta-bulge :

Two sidechains (usually polar) point to one side of the sheet, instead of just one. This causes a kink in an otherwise continuous pairing.

A beta-bulge is almost always a 1-residue insertion, here, usually a polar amino acid, most frequently D.

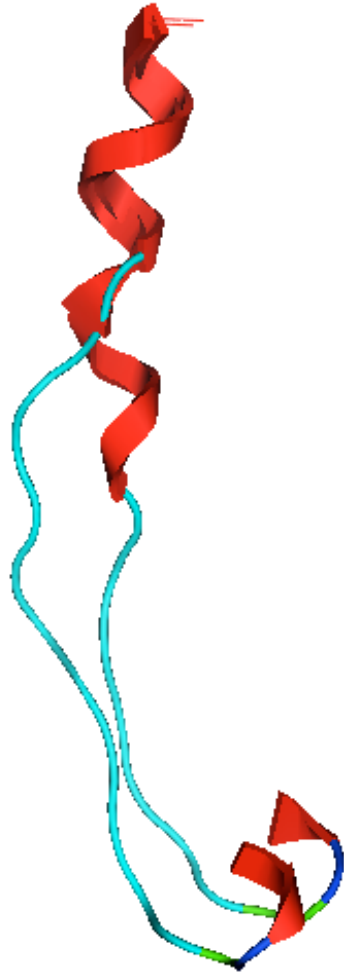
Occasionally, a many residue insertion occurs here. It is called a “beta-blowout.”

An exception to the *no-insertions-in-helix* rule



```
gi|157041248|ref|NP_001096641.1| myo  
INLISLKPY····GILRILDDQCCF  
1G8X.A STRUCTURAL PROTEIN  
QATIDLIDGRQPPGILALLDEQSVF  
1W9I.A MYOSIN  
QATIDLIDGRQPPGILALLDEQSVF
```

Not an exception to the *no-deletions-in-helix* rule



Deletion in the sequence leads to shorter helix, not a helix with a "*deletion*" in it.

This shows two actual structures. The one with the deletion is more extended, to span the distance.

Alignment

```
AFADNQ·PCINLIS  
TFIDFGLDSQATID
```

8.2 Unalignment

Telling MOE how to anchor a loop search: the wrong way



MOE will attempt to
connect template G to H !!

MOE will attempt to insert
NP between M and Q, which
are bonded!!

Instead, carefully choose the “anchors” of the loop search. The anchors (or, pre-flex and post-flex) will define the loop search and will be restrained (tethered) to the template during energy minimization.

Telling MOE how to anchor a loop search: the right way

MOE will swap 2 residues
into the space of 6.
2 for 6.

MOE will insert NPQ into the
space of Q.
3 for 1.



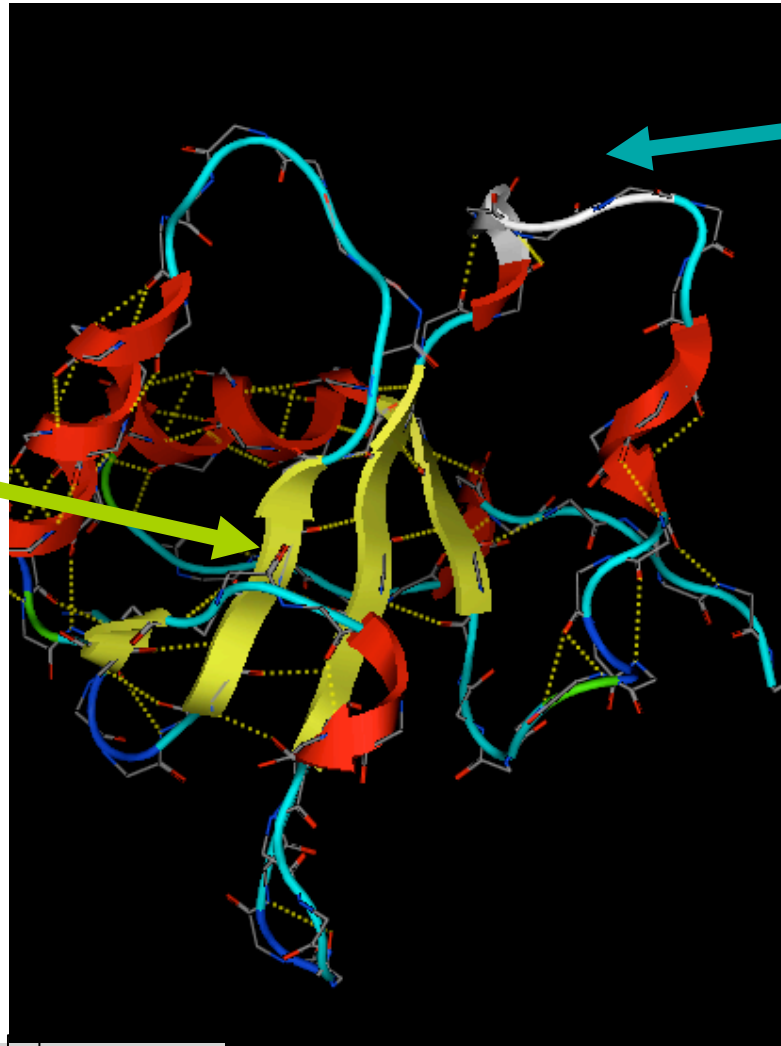
The anchors for
this loop search
are DF and IY

The anchors for
this loop search
are IM and QS

Remplate positions to be treated as

Setup of Loop building: the wrong way

Deleted cys is here, in the middle of a strand.



This part of the template will be deleted. A long bond will connect the ends. Energy minimization will cause commotion.

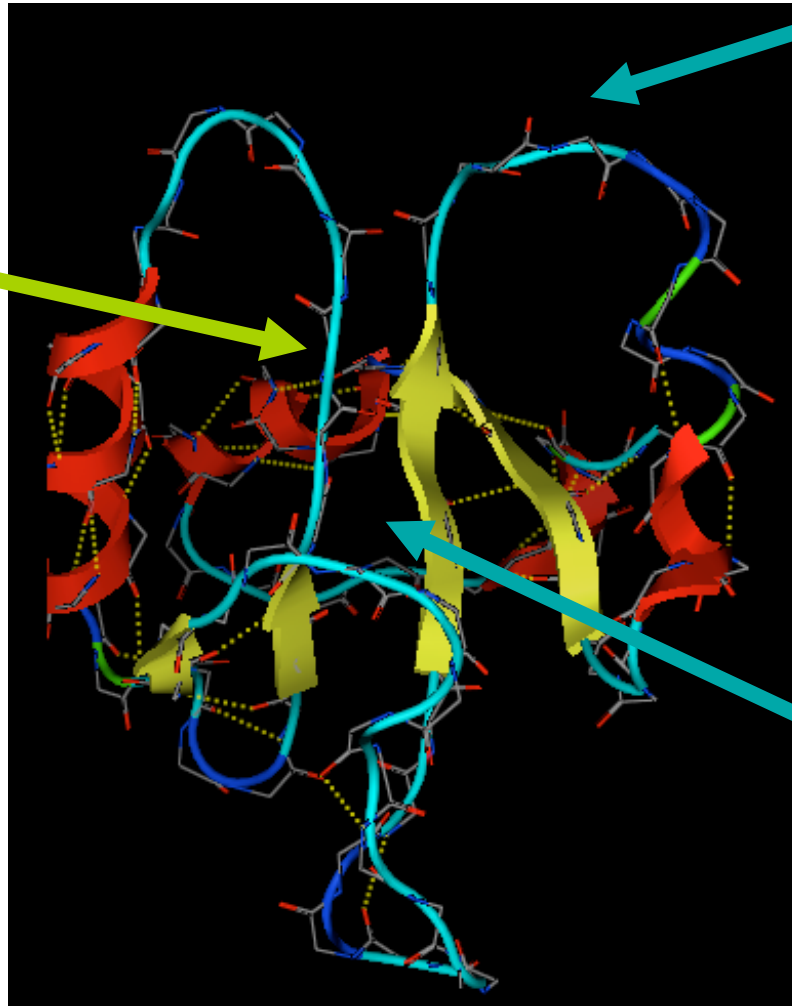
target
template

AGFYI · YPTPHVQP
RGL EICCYGPFTNM

QNYVSHSIVLGGE · · · EDKSEVENAAI
GTGV · HPIVVVQPPDAW TEDNGFHAIGI

Results of Loop building: the wrong way

Deletion here results in straightening and shifting of the strand.



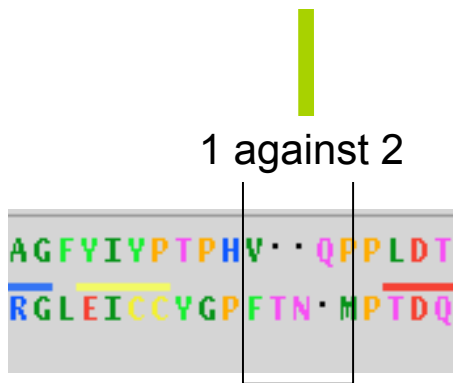
The resulting model has a short loop here.

Energy minimization has *pulled the sheet apart*.

No H-bonds in middle of sheet!

Setup of Loop building: the right way

Location of indel moved to here.



1 target residue unaligned,
 2 template residues
 deleted



Old deletion
 0 against 3



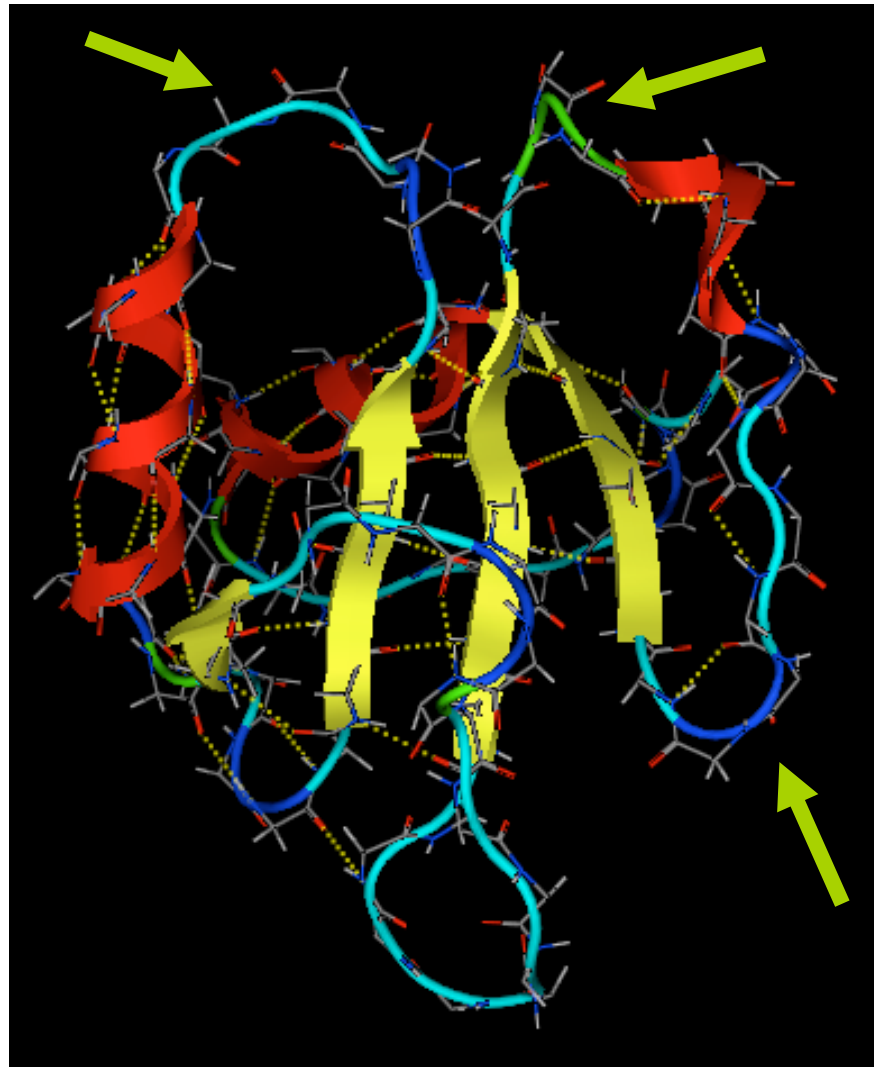
New deletions

2 against 5

2 against 3



Results of Loop building: the right way



New loops
are at the
ends of
SSEs.

Sheet has
retained H-
bonds.

Take-home lesson: Unaligning makes loop search work better.

Work on Homework 4

- Fixing a bad model by fixing the alignment.
- http://www.bioinfo.rpi.edu/bystrc/courses/biol4550/homework4_refine.pdf