

PyMOL Tutorial Notes

What is a PDB file?

Go through the Ubiquitin pdb file: 1UBQ.pdb

<https://bit.ly/2o5uqqa> (link to slides showing 1UBQ.pdb)

“The primary information stored in the PDB archive consists of [coordinate files](#) for biological molecules. These files list the atoms in each protein, and their 3D location in space. These files are available in several formats (PDB, mmCIF, XML). A typical PDB formatted file includes a large "header" section of text that summarizes the protein, citation information, and the [details of the structure solution](#), followed by the sequence and a long list of the atoms and their [coordinates](#). The archive also contains the [experimental observations](#) that are used to determine these atomic coordinates.”

(<https://pdb101.rcsb.org/learn/guide-to-understanding-pdb-data/introduction>)

Basic PyMOL Controls

https://pymol.org/dokuwiki/doku.php?id=mouse:one_button -- good link on mouse controls

Especially relevant for laptop users:

Mouse Controls: One Button Mode

PyMOL is not designed to be driven with a one button mouse. However, there are times when that is all you have, for example, when using a laptop in a cramped airline seat. In such cases, the following limited manipulations are available if you select "1 Button Viewing Mode" from the "Mouse" menu:

Action	Effect
click-and-release	toggle selection
click-and-drag	rotate the camera
double-click	activate context-sensitive menu
SHIFT-click-and-drag	drag selection box
CTRL-click-and-drag	zoom (move camera along Z axis)
CTRL-shift-click-and-drag	adjust clipping planes
ALT-click-and-drag	move camera in XY plane

Most other manipulations are not available when running in one-button mode.

Examining basic PyMOL controls with 3BTA.pdb

We will be examining the following protein -- a bit about it:

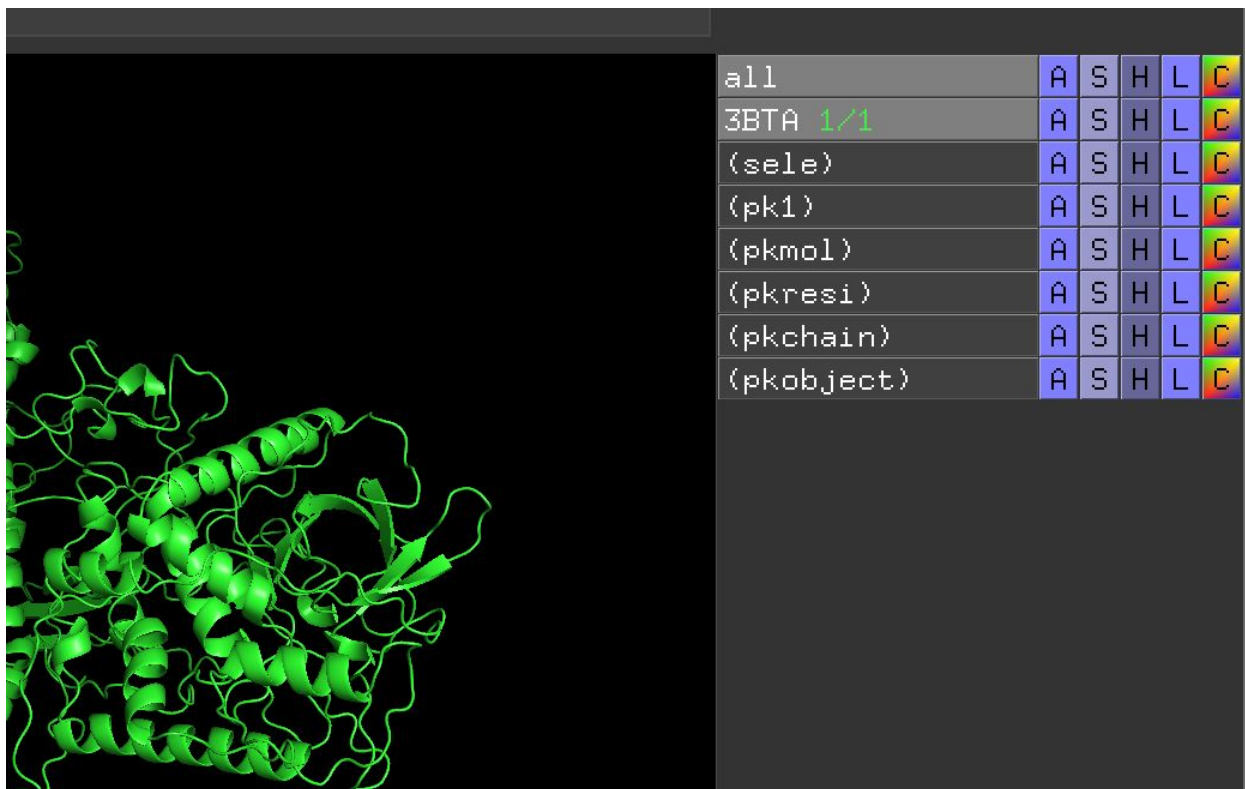
“Botulinum neurotoxin type A (BoNT/A) is the potent disease agent in botulism, a potential biological weapon and an effective therapeutic drug for involuntary muscle disorders. The crystal structure of the entire 1,285 amino acid di-chain neurotoxin was determined at 3.3 Å resolution. The structure reveals that the translocation domain contains a central pair of alpha-helices 105 Å long and an approximately 50 residue loop or belt that wraps around the catalytic domain. This belt partially occludes a large channel leading to a buried, negative active site--a feature that calls for radically different inhibitor design strategies from those currently used. The fold of the translocation domain suggests a mechanism of pore formation different from other toxins. Lastly, the toxin appears as a hybrid of varied structural motifs and suggests a modular assembly of functional subunits to yield pathogenesis” (from RSCB PDB)

We will begin by examining how to select atoms & different visual coloring schemes.

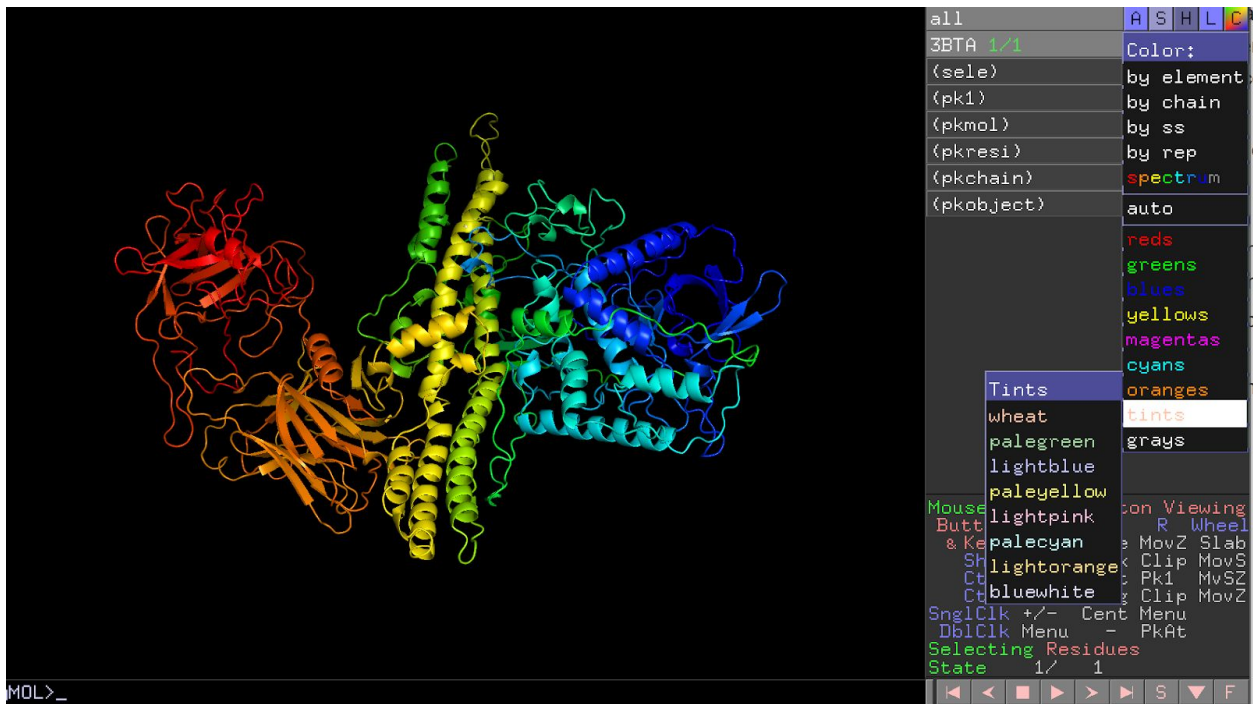
→ “fetch 3BTA” in the PyMOL command terminal to get the PDB

→ Ctrl + right click to select individual atom

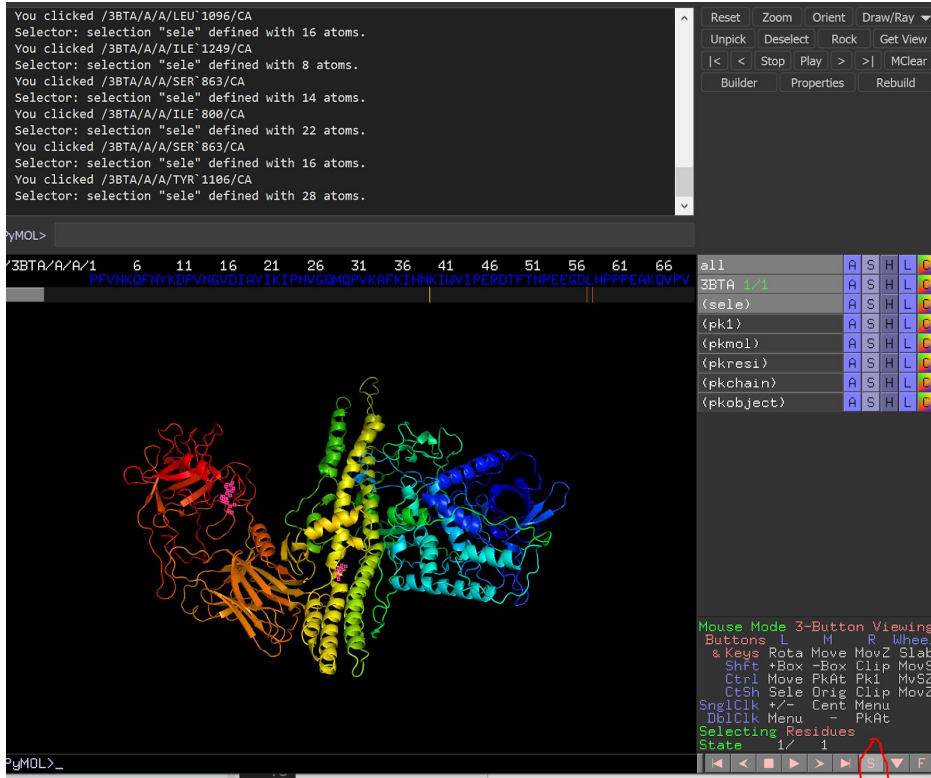
→ Click the side “S” to show different coloring forms e.g. ribbon, cartoon, stick, etc.



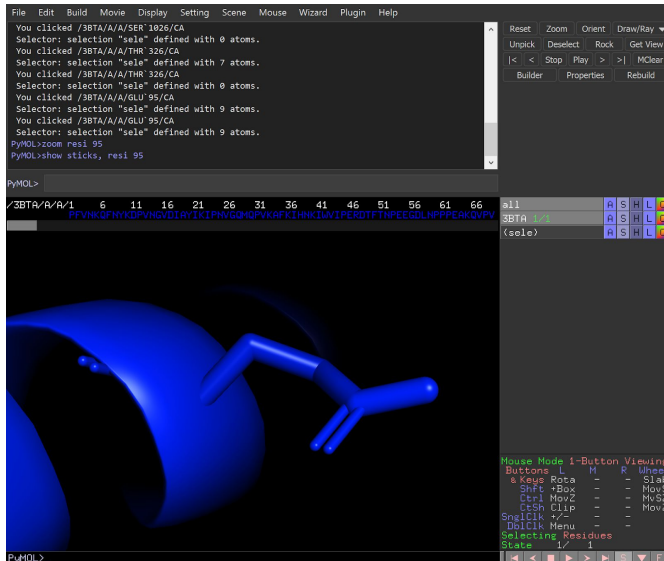
→ Show coloring by chains -- chainbows



→ Selecting specific residues -- can click on the circled "S" in the bottom right hand corner. Note that the notation "TYR`1106" refers to a TYR amino acid that is residue number 1106 in this specific protein.



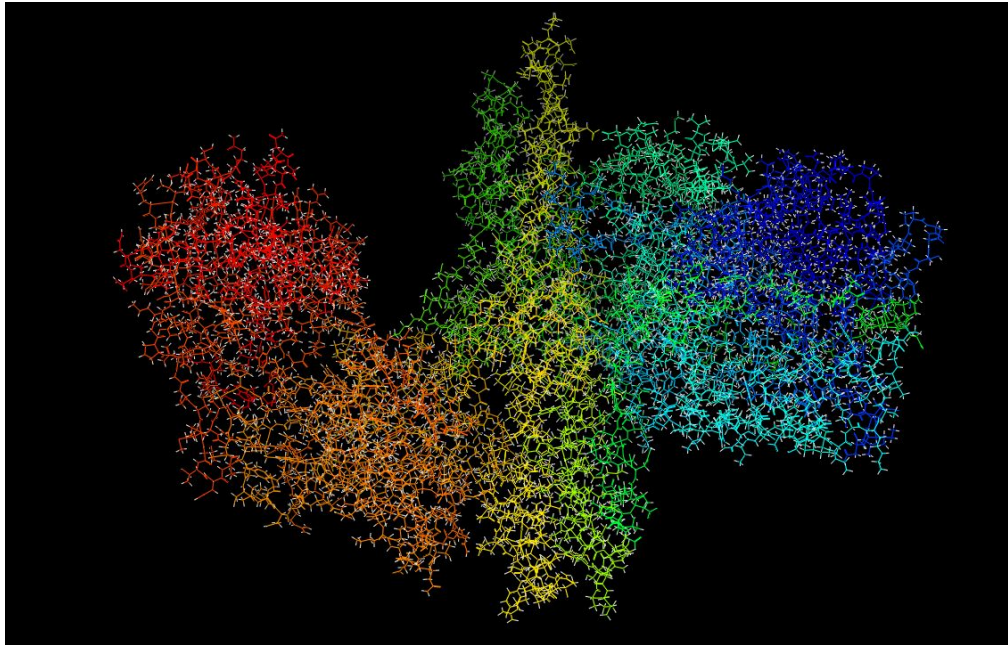
→ Show zooming/other mouse control functionalities (see previous section)
 → Show how we can use zoom from the command prompt in PyMOL, e.g. "zoom resi 95" or whatever residue is selected/of interest and then type in "show sticks, resi 95" to better see the residue



→ Click the "reset" button on the side to go back to the original full screen view.

Visualizing hydrogens can be helpful to better understand a protein's structure. We will go through doing that now.

→ Show wires, hide cartoon (from side menu representation elaborated on earlier), then, type in `h_add` to show hydrogen bonds. After adding hydrogen bonds should look like:



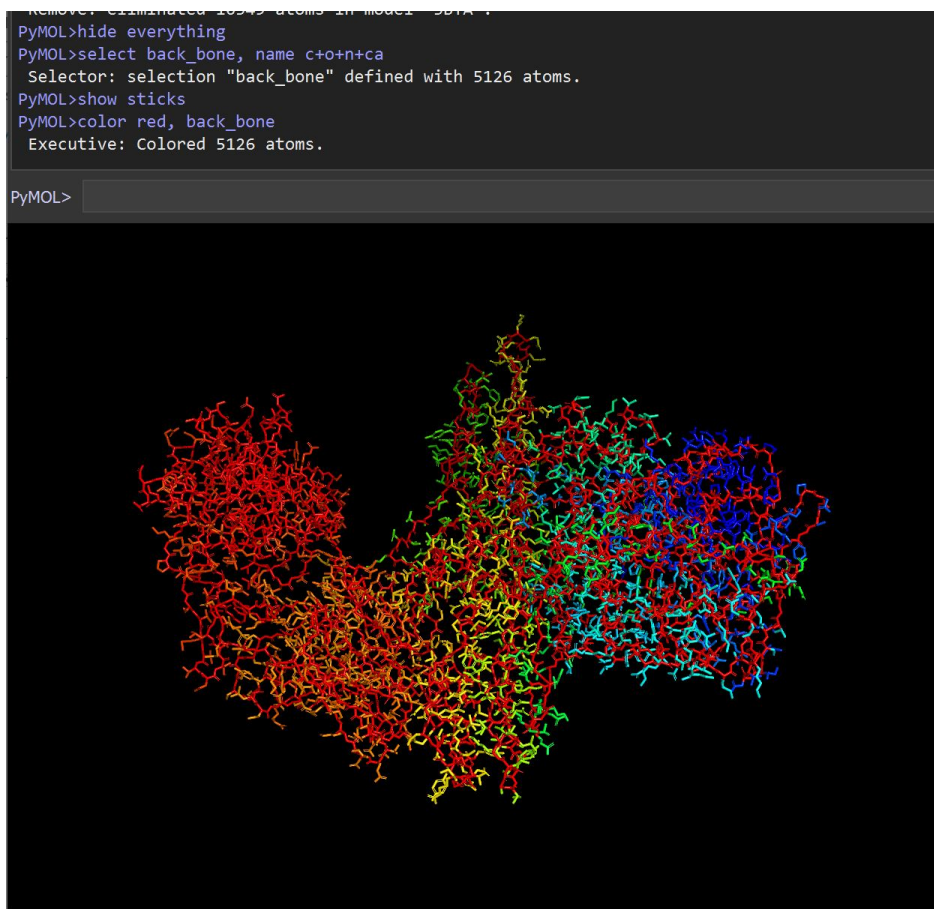
→ Then type in `remove hydrogens` to remove hydrogens.

We will now visualize the backbone of this structure.

Enter following commands:

→ `hide everything`
→ `select back_bone, name c+o+n+ca`
→ `show sticks`
→ `color red, back_bone`

Name `c+o+n+ca` indicates the atoms you are selecting and these constitute the backbone. Point out that this now shows the protein backbone.



Good links:

http://pymol.sourceforge.net/newman/user/S0210start_cmds.html

https://pymolwiki.org/index.php/Practical_Pymol_for_Beginners

Examining the mutagenesis wizard & keratin

Note: enter "reinit" to reinitialize.

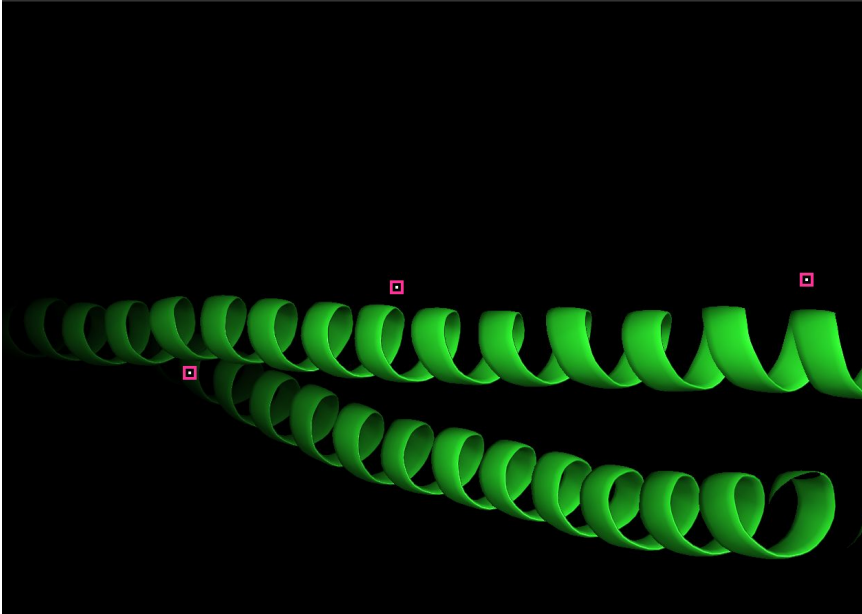
We will now examine keratin, a critical structural protein found in hair and nails, you may be familiar with. The PDB identifier for keratin is 3TNU.

Enter the following commands to better examine the structure:

```
fetch 3TNU
remove resn hoh -- to remove water, can also use "remove solvent"
select sulfur_atoms, name sg
zoom sulfur_atoms
```

```
PyMOL>fetch 3TNU
TITLE Heterocomplex of coil 2B domains of human intermediate filament proteins, keratin
5 (KRT5) and keratin 14 (KRT14)
ExecutiveLoad-Detail: Detected mmCIF
CmdLoad: loaded as "3TNU".
PyMOL>remove resn hoh
Remove: eliminated 6 atoms in model "3TNU".
PyMOL>select sulfur_atoms, name sg
Selector: selection "sulfur_atoms" defined with 3 atoms.
PyMOL>zoom sulfur_atoms

PyMOL> |
```



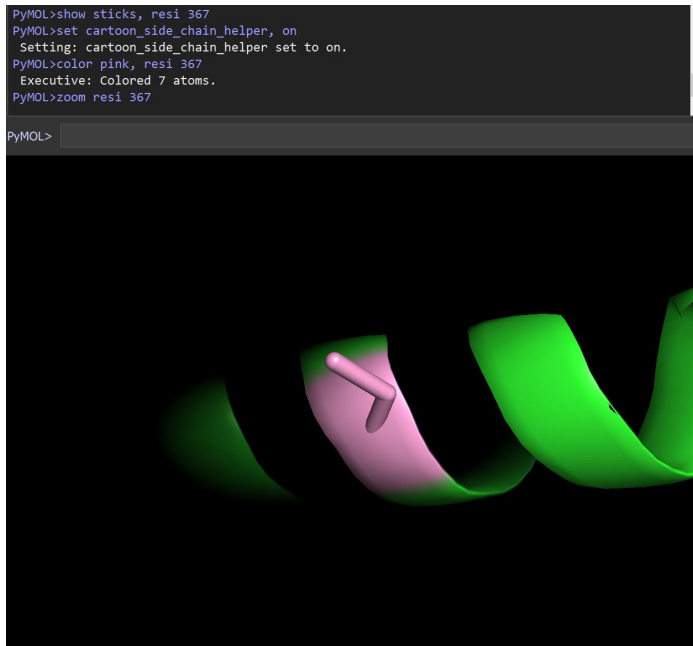
The above commands select sulfur atoms. You can now see with the next few commands, that they are part of the cysteine residues here. Cysteine performs an important stabilizing role; Cysteine residues are part of the disulfide bridges that stabilize keratin. Cysteines are polar. Now, we select all cysteine residues.

```
PyMOL>sele resi cys
Selector: selection "sele" defined with 0 atoms.
PyMOL>sel cysteines, resn cys
Selector: selection "cysteines" defined with 18 atoms.
PyMOL>color slate, cysteines
Executive: Colored 18 atoms.
You clicked /3TNU/B/B/CYS`407/CA
Selector: selection "sele" defined with 6 atoms.
```



Now, we examine one cysteine residue in particular.

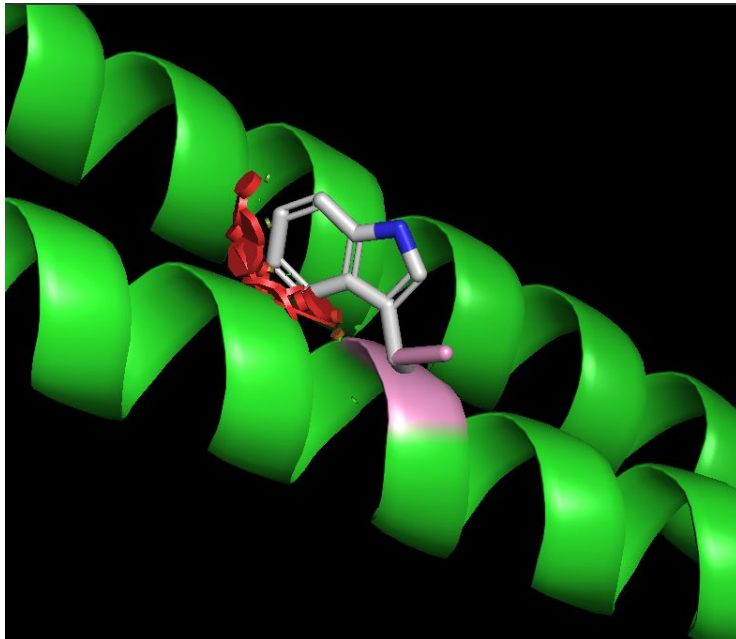
```
PyMOL>show sticks, resi 367
You clicked /3TNU/A/A/CYS`367/CA -> (pk1)
PyMOL>set cartoon_side_chain_helper, on
Setting: cartoon_side_chain_helper set to on.
PyMOL>color pink, resi 367
Executive: Colored 7 atoms.
PyMOL>zoom resi 367
```

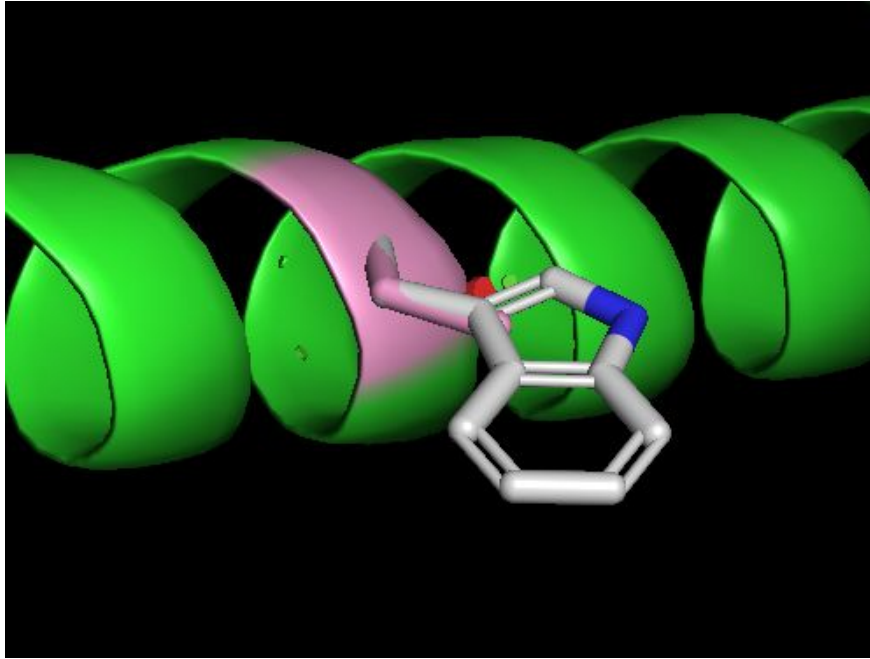
We will now mutate this cysteine.

Let's mutate the cysteine to TRP which is non polar so should result in a structural change -- using mutagenesis wizard which you can find in the browser bar under Wizard → Mutagenesis → Protein. Highest strain rotamer is 7, collides with helix, lowest strain is 4, is parallel. Note the red circles denote clashing.

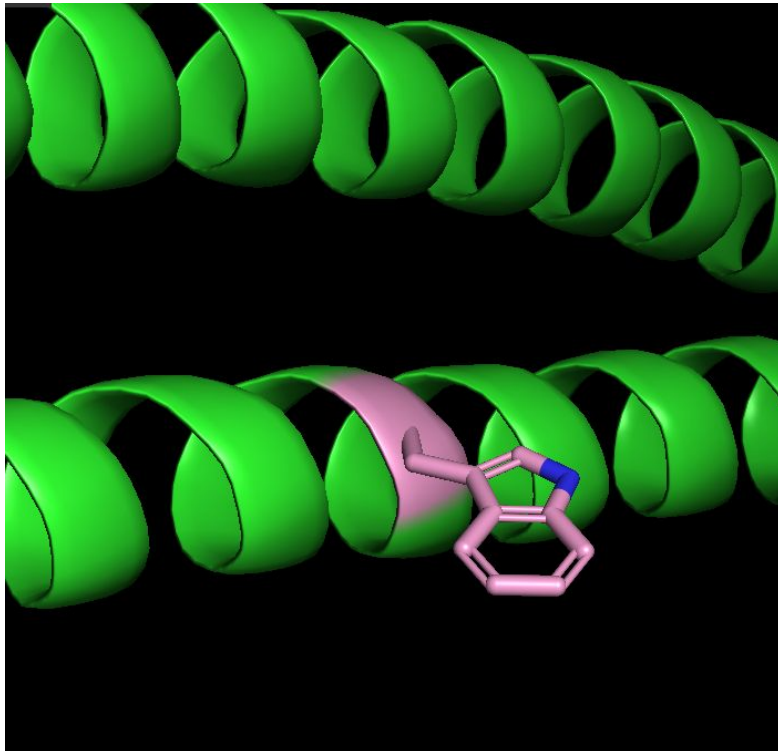
Highest strain rotamer:



Lowest strain rotamer:



Click apply -- you now see the mutated keratin.



Examining ricin, a toxin found in castor beans

<http://pdb101.rcsb.org/motm/161> -- we will run through this. You can:

→ fetch 2aai

→ remove solvent

→ note parts "A" and "B" as identified in the article -- how would you use PyMOL to visualize them differently using what we have learned?

< can use the shift + click + drag to select A and the color command to color >

→ visualize the disulfides as spheres with the GUI