

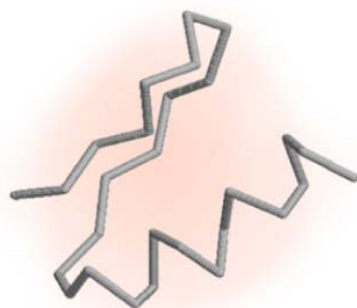
Section III - Designing Models for 3D Printing

In this section of the Jmol Training Guide, you will become familiar with the commands needed to design a model that will be built on a 3D Printer. As you become more comfortable using Jmol, this section will enable you to take the next step and design physical protein models.

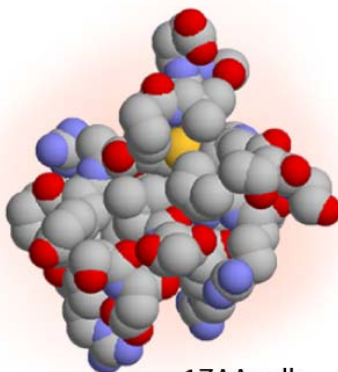
At the end of this section, you should be familiar with how to use Jmol to create a physical protein model on a rapid prototyping machine, including:

Section 3: Designing Models for 3D Printing

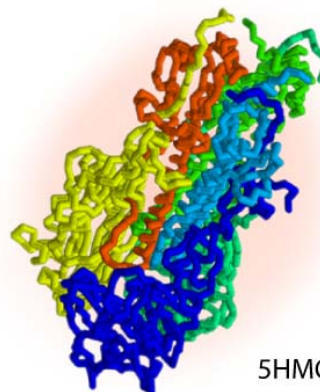
- 3-1 - What is 3D Printing?
- 3-2 - Selecting Appropriate Display Formats and Colors
- 3-3 - Adding and Removing Hydrogen Bonds and Disulfide Bonds
- 3-4 - Adding and Removing Support Struts
- 3-5 - Coloring Hydrogen Bonds, SS Bonds, and Struts
- 3-6 - Adding Sidechains with a "Clean Backbone"
- 3-7 - Additional Miscellaneous Jmol Commands



1ZAA.pdb



1ZAA.pdb



5HMG.pdb

3-1 - What is 3D Printing?

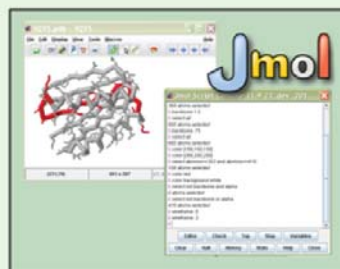
- 3D Printing is a cutting-edge technology that can use stacked serial planes to build physical 3-dimensional models from any computer generated 3D file. Every physical protein model design goes through four key steps:

1



X,Y,Z coordinates in the form of a PDB file are the starting point of every protein model.

2



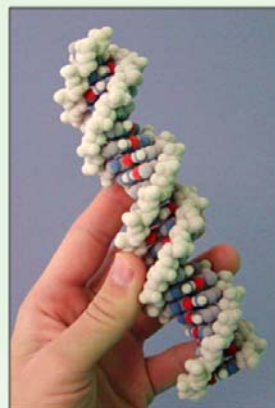
Design software is then used to edit the features and format of the protein model

3



The model is then exported as a file that is recognized by a 3D printer

4

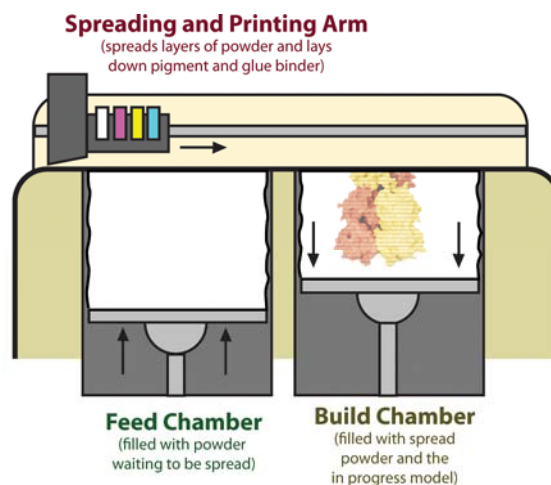
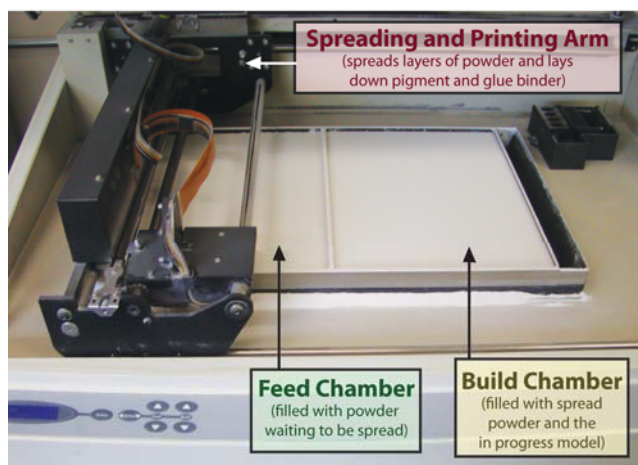


Finally, the physical model is removed from the printer and completed in a post production process.

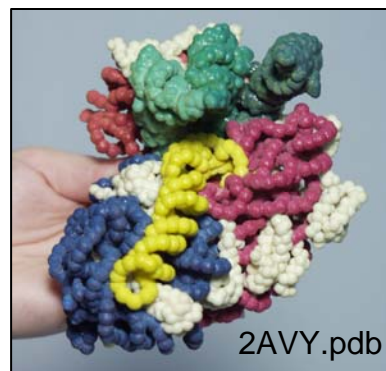
- Z-Corporation 510 and 650** - Although many varieties of 3D Printing and Rapid Prototyping machines exist, the MSOE Center for BioMolecular Modeling relies mainly on two Z-Corporation machines - the Z510 and the Z650.



- **How a 3D Printer Works** - The inside of a 3D Printer has three main parts: the feed chamber that stores the plaster powder to be used, the build chamber where the printed model will be built, and the spreading and printing arm which applies a glue and pigment solution to the growing model in the build chamber. While printing, the spreading and printing arm moves from left to right pushing a thin layer of plaster powder into the build chamber. The arm then makes a second pass printing a glue pigment solution onto the new layer in the build chamber. The build chamber is then lowered as the feed chamber is raised so a new layer of plaster can be spread. This process continues until all layers of the model have been completed.



- **The End Result** - A designed and printed protein model will be an exact replica of the model created in the 3D visualization program.

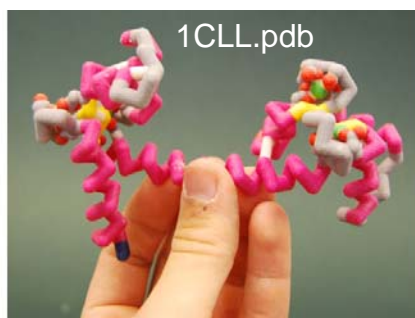


3-2 - Selecting Appropriate Display Formats and Colors

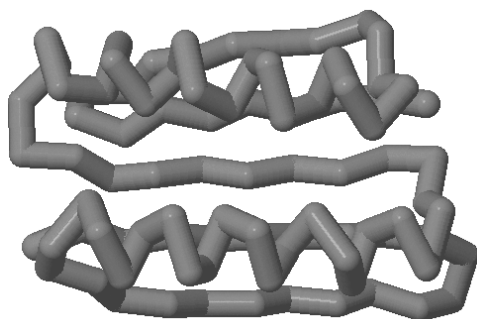
- **What Display Should I Use?** - This is a common question asked by model designers. And the answer to this question is another question: "What is the story that you are telling with your model?"
 - If your story is focused on a particular active site within the molecule, then perhaps the Alpha Carbon Backbone Format displaying key active site amino acid sidechains in Ball and Stick (Wireframe and Spacefill) Format would be best. If your story is focused

on how two subunits interface at the surface, then perhaps the Spacefill Format is the best choice. Ultimately the choice is yours. In Section I, there is a table highlighting the advantages and disadvantages of each display format. This will assist you in deciding which format is the best for telling your story.

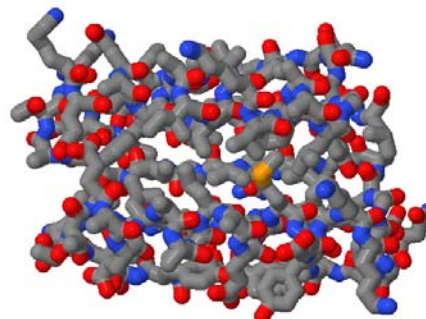
- The important point to remember is that no one model will tell every aspect of the story. Using Jmol in combination with a physical model will assist you in telling multiple aspects of your story.
- The CBM will build models in the Spacefill Format and the Alpha Carbon Backbone Format. Models in the Alpha Carbon Backbone Format can have sidechains and heterologous groups displayed. We currently do not build models in Ribbons or Strands Format.



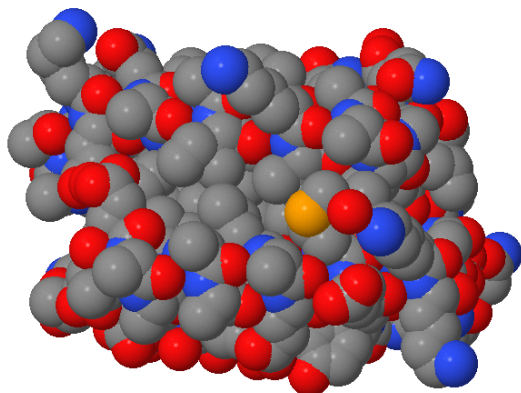
- **Format Sizes** - Every display format in Jmol is able to be displayed with various thicknesses (see section 1.4 for more information on Display Formats). If a display format is too thin, a protein model will be more likely to break during the 3D printing process. If a format is too thick, the model will use excessive amounts of material and details will be less visible. Below is a list of suggested sizes for the different formats:



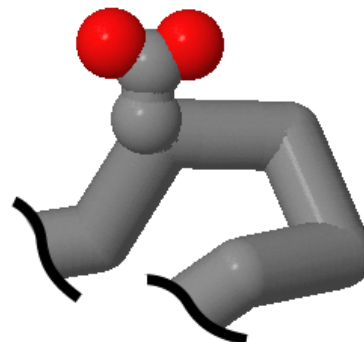
Backbone 1.5



Wireframe 1.25



Spacefill



(for sidechains only)

★ Note: The default spacefill does not include a number. This is known as Full Spacefill.

Wireframe 1.25

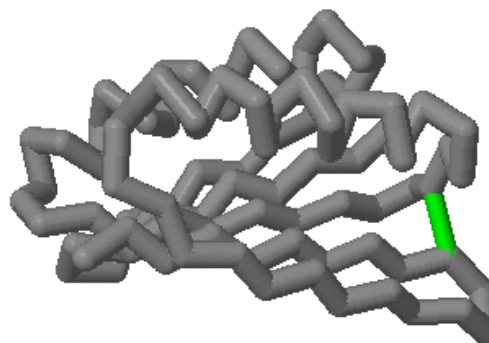
Spacefill 1.5

For all bonds and struts

hbonds 1.0

ssbonds 1.0

Struts 1.0



- **Which Colors Should I Use?** - Which colors you use, like which Display Format you choose, is largely influenced by the story you would like your model to show.
 - If the overall shape and fold of your protein is important to your story, you may want to color the secondary structures (alpha-helices and beta-sheets) a bright eye-catching color.
 - If there is a specific sidechain or active site area that is important to the function of your protein, you may want to highlight that area with a unique or bright color.
 - You can color a protein model in an almost unlimited variety of ways so it is very important to consider what you want to show and what colors would be best to highlight the key points to your protein's story.

- **Colors Not to Use** - There are a few colors or color combinations that you should avoid while designing a protein model to be build using 3D printing technologies.

- Do not use any black or extremely dark colors. The 3D printers do not print well with dark colors and often leave stripes or unevenness in the final physical model.



- Do not use colors that are too similar to highlight different features. These will often not be distinguishable in the final physical model.



- Do not use too many extremely bright colors together on the same model. When used rarely, very bright colors are extremely good at drawing the eye to important areas of the protein model. Overuse of bright colors can distract from the model.

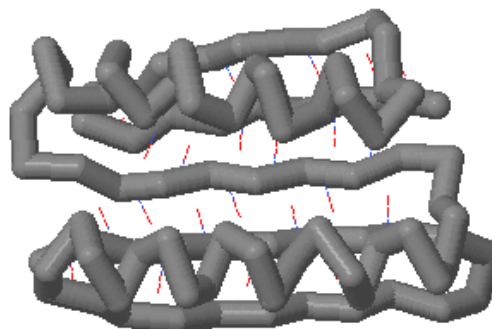


3.3 - Adding and Removing Hydrogen Bonds and Disulfide Bonds

- **Hydrogen Bonds** - Hydrogen bonds (referred to as "hbonds" in Jmol) are essential to the stability of secondary structures in a protein. They form between the backbone oxygen of one amino acid and the backbone nitrogen in a second amino acid. To add hydrogen bonds to secondary structures within your model, use the `calculate hbonds` command. We typically do not add hydrogen bonds to alpha-helices, since they do not add stability to the model and actually clutter the view of the structure. Adding hydrogen bonds to beta-sheets provides additional support for the final model and is recommended. Use the following commands:

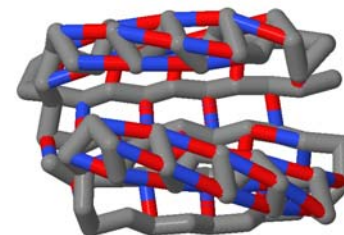
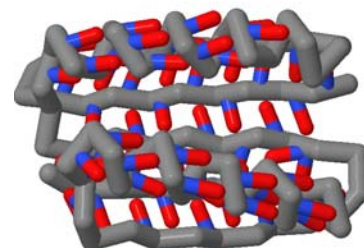
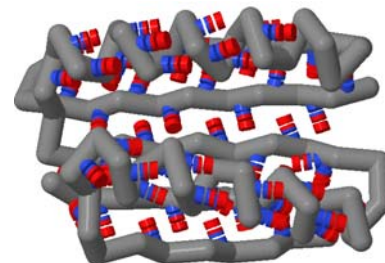
- `select sheet`
- `calculate hbonds`

- After you have entered this command, you will notice that there are dotted lines that have appeared.



These are the hydrogen bonds.

- The default size when you first add hydrogen bonds is the same as for wireframe which is very thin.
- **Thickness** - Hydrogen bonds, like wireframe, backbone, and spacefill, can be thickened by placing a number after the `hbonds` command.
 - `hbonds 1.0`
- **Solid hbonds** - The default display for hydrogen bonds is a dashed line. You will need to change this into a solid cylinder for building a physical protein model using 3D printing. You can do this using the command:
 - `set hbonds solid`
- **Setting hbonds to the Backbone** - Notice that the hydrogen bonds are now thicker and solid, but that they appear to be floating in air. This appearance results from the fact that hydrogen bonds form between the atoms that make up the backbone of the amino acid (the nitrogen and the oxygen atoms), but since we have displayed only the alpha carbon atoms in Backbone Format, it appears as if the hydrogen bonds are floating in space. Therefore, we must set the hydrogen bonds to the backbone using the command:
 - `set hbonds backbone`
- **Removing all hbonds** - To turn off all the hydrogen bonds in a selected area, use the `hbonds off` command.
 - `hbonds off`
- **Summary of Commands to Add hbonds:**
 - `select sheet`
 - `calculate hbonds`
 - `hbonds 1.0`
 - `set hbonds solid`
 - `set hbonds backbone`
- **Adding or Removing Individual hbonds** - At times you may want to add or remove a single hydrogen bond. First, you need to know the two **amino acid residue numbers** that the hydrogen bond connects. To do this, click on the two residues in the Display Window (see section 1.5 for more information on identifying atom numbers). Once you know the two



residue numbers, you must select only these two amino acids. Finally, use the `hbonds 1.0` command to add an hbond or the `hbonds off` command to remove the bond between the two selected amino acids.

- For example: `select 167 or 73`
`hbonds 1.0`

For example: `select 321 or 334`
`hbonds off`

- **Disulfide Bonds** - Some molecules will have disulfide bonds (referred to as "ssbonds" in Jmol) present within the structure. These bonds form between two cysteine amino acids that lie close to each other in 3D space. Disulfide bonds are added using the command `ssbonds`.

- `ssbonds`

- **Thickness** - As we saw with the hydrogen bonds, simply typing `ssbonds` by itself will only produce thin lines. To give these bonds dimension, we must add a thickness (number) to the ssbonds.

- `ssbonds 1.0`

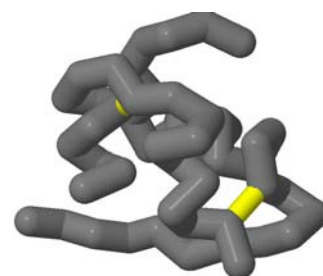
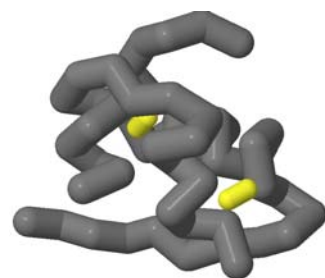
- **Setting ssbonds to the Backbone** - Notice that this command gives the disulfide bonds a thicker dimension, but as we saw with the hydrogen bond, the bonds are "floating" in space. This is because the disulfide bond is actually between the sulfur groups of the cysteine sidechains, and not the alpha carbons. To make the disulfide bond connect between the backbone units, we need to set the bonds to the backbone. Note that the disulfide bond is orange (the CPK color for sulfur).

- `set ssbonds backbone`

★ Note: You may wish to display the sidechains of the cysteines involved in the disulfide bond. If that is the case, then you will not need to set ssbonds to the backbone.

★ Note: Not every PDB file has ssbonds. The Top 7 protein PDB file used in Section 1 does not have a disulfide bond. To practice adding disulfide bonds, try the PDB file 2hiu.pdb which contains the coordinates for one molecule of human insulin.

- **Adding or Removing Individual ssbonds** - At times you may want to add or remove a single disulfide bond. To do this, you first need to know the two **amino acid residue numbers** that the bond connects. To do this, click on the two residues in the Display Window (see section 1.5 for more information on identifying atom numbers). Once you know the two



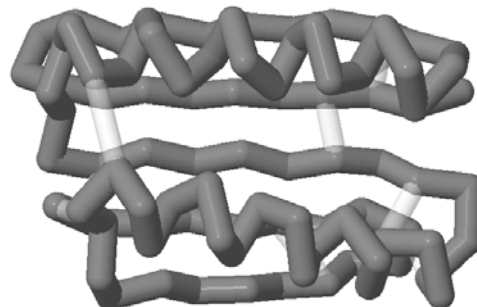
residue numbers, you must select only these two amino acids. Finally, use the `ssbonds 1.0` command to add an ssbond or the `ssbonds off` command to remove the bond between the two selected amino acids.

- For example: `select 167 or 73`
`ssbonds 1.0`

For example: `select 321 or 334`
`ssbonds off`

3.4 - Adding and Removing Support Struts

- **Support Struts** - When a model is built on a 3D printer, it needs to be supported in order to withstand the additional pressure as powder builds up and as the model is removed and cured. Hence, support struts are added within the model for support. To do this, select your entire model and use the calculate struts command.



- `Calculate struts`

- **Thickness** - Struts, like hydrogen bonds and ssbonds, can be set to different thicknesses by adding a number after the strut command.

- `Strut 1.0`

- **Adding or Removing Individual Struts** - At times you may want to remove a single strut (for example, if it goes through a sidechain you have represented). To do this, you first need to know the **atom numbers** of the two atoms the strut connects. To do this, click on the two atoms in the Display Window (see section 1.5 for more information on identifying atom numbers). Once you know the two atom numbers, you must select only those two atoms using the `select atomno=` command. Finally, use the `struts 1.0` command to add a strut or the `strut off` command to remove the bond between the two selected amino acids.

- For example: `select atomno=167 or atomno=73`
`strut 1.0`

- For example: `select atomno=321 or atomno=334`
`strut off`

★ Note: The calculate strut command may not always provide enough structural support. Additional struts may be necessary. Struts will need to be added manually to attach any ligands (substrate, inhibitor, cofactors) to the protein.

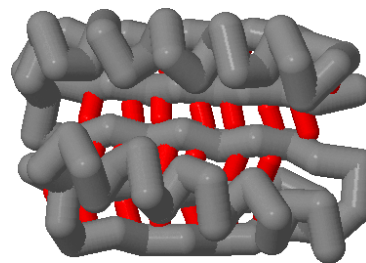
3.5 - Coloring Hydrogen Bonds, SS Bonds, and Struts

- **Default Bond Colors** - When bonds or struts initially appear, they will be the default Jmol colors (a translucent white for struts and the colors of the atoms that they connect for hbonds and ssbonds).
 - If the two atoms the bond is connecting are the same color, then the hbond or ssbond will be the same color throughout the entire length of the monitor line.
 - If the two atoms are different colors, then the hbond or ssbond will be half one color and half the other color.
- **Changing All Bonds** - To color all of a certain type of bond or strut, select all and then use the color command followed by the type of bond or strut you want to color and then the color you would like to color them.
 - For example:

- `select all`
- `color hbond red`

or:

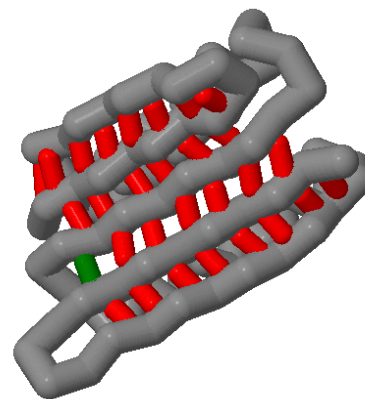
- `select all`
- `color strut red`



★ Note: We recommend that you choose a light color for your struts, such as white or light gray. Struts are support structures and should not be the emphasis of your model. A vibrant or dark color will draw the user's eye to the struts, and that should not be the focus of your model.

- **Changing A Single Bond Color** - To change the color of an individual bond or strut, follow the same steps for removing an individual bond or strut. Identify the two atom numbers that the bond or strut connects. Then, select only those two atoms. Finally, use your color command to apply the new color.
 - For example:

- `select atomno=333 or atomno=65`
- `color hbond green`



3.6 - Adding Sidechains to Create a "Clean Backbone"

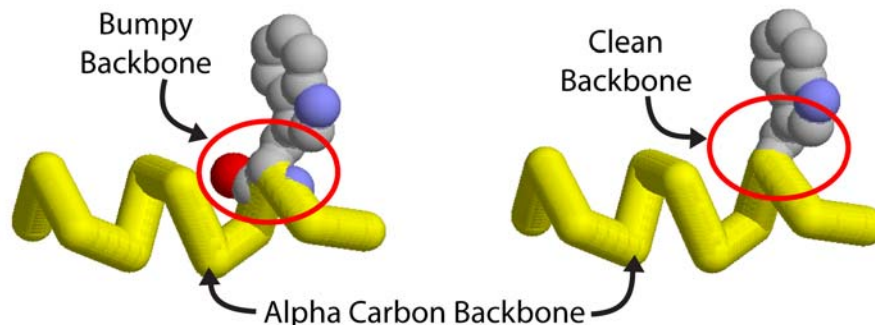
- If your molecular story requires that you display specific amino acid sidechains in the model, we recommend that you do only display the sidechain atoms, rather than all of the atoms of the amino acid. This gives the model a more aesthetically pleasing appearance. In previous sections, we have simply selected the amino acid by its residue number and displayed all of the atoms in that amino acid. In this section, we are going to use Boolean operators to select just the atoms in the sidechain and display only these atoms.
- To select and display only the atoms of the sidechain of a specific amino acid, you want to use the `select` command followed by the amino acid name/number and end with the `and (sidechain or alpha)` text.
 - `select cys30 and (sidechain or alpha)`
 - This command selects the amino acid at residue 30, but limits the selected atoms to the sidechain atoms and the alpha carbon of that amino acid.

★ Note: It is important to select the alpha carbon atom in addition to the sidechain atoms because this will attach the sidechain atoms to the alpha carbon. If you do not select the alpha carbon, the sidechain will build as a separate unit from the rest of the molecule. Once your sidechain is connected, give it wireframe and spacefill dimensions:

- `wireframe 1.0`
- `spacefill 1.25`

★ Note: These two commands together will generate a ball and stick appearance. You can enter just the wireframe command and create a "sticks" appearance, but you cannot enter just the spacefill command. If you enter just the spacefill command, the atoms will be displayed as little spheres and the spheres may not be securely connected to one another. The image MAY look fine on the screen, but the model will come out of the 3D printer in pieces that can't be put back together! It is therefore imperative to add the wireframe command to connect the spheres together.

- If you do not use the `and (sidechain or alpha)` command when displaying sidechains, you will generate a "bumpy backbone." Unless the backbone atoms of the amino acid (the amino nitrogen or the carbonyl oxygen) play a specific role in the story you are telling, we do not recommend displaying these atoms on the model. It is typically (although not always) the sidechain that has the specific chemical role within the molecule and plays the key role within your story. Therefore, this should be the part that is displayed within your molecule.



3.7 - Additional Miscellaneous Jmol Commands

- In Section I of the Jmol Training Guide, you were introduced to the basic commands of Jmol. We will now present additional commands to further develop your Jmol design skills.
 - **The “Center” Command** - This command allows you to center the rotation of the molecule on a certain portion of the molecule.
 - - **center**
 - Jmol will by default center the molecule at the center of the entire molecule. If you restrict your viewing to a certain subset, when you rotate the molecule around in space, the molecule will seem lopsided. This is because although you have restricted your viewing to just a part of the molecule, all of the atoms are still present and being used to determine the center of rotation; you just do not see them.
 - Therefore, you need to use the **center** command to center the molecule on the restricted region.
 - For example: **center *c**

★ Note: Once again, we are using the ***** command to dictate to Jmol that we are centering the molecule on the all the atoms in Chain C.

- **Identifying the Amino and Carboxy Termini** - An important concept in protein structure is that each protein has an amino terminus and a carboxy terminus. Through Jmol, you can readily identify each of these termini.
 - **Amino Terminus** - The Amino Terminus is the first amino acid in the protein. When a protein is synthesized, it begins with the 5' end of the mRNA and synthesizes in a 5' to 3' fashion. Therefore, the first amino acid in the protein will be the amino acid that is encoded at the 5' end of the mRNA.

- To determine the amino terminus of the protein in the PDB file, click on the atom at each end of the protein. The atom with the lowest amino acid number will be the amino terminus.
- Alternatively, you may search the PDB sequence information to identify the amino terminus amino acid and use the Jmol command line window to select the specific amino acid.
- **Carboxy Terminus** - The Carboxy Terminus, on the other hand, will be the last amino acid in the protein.
 - To determine the identity of the carboxy terminus, click on the atom at each end of the protein. The amino acid with the largest number will be the last amino acid in the protein.

★ Note: Occasionally a PDB file will have gaps or missing sections in a chain because of incomplete experimental data in the file. This can easily be confused for the Carboxy Terminus. Be sure that you are really looking at the last amino acid in the chain and not just the beginning of a gap.

- **Repeating a command** - By pressing the up arrow key on the keyboard, the previous commands that you have entered into the command line window will be displayed, in reverse order. You can edit these commands before hitting enter.
- **Undo Button** - Near the bottom right of the Console Window is an undo button. This button can be used to undo recent commands and take your protein design back to a previous state. Be sure to save your work frequently! Use judiciously; sometimes you need to undo multiple steps to get to the command you want to undo.
- **Selecting specific types of atoms** - If you wish to select a subset of atoms, such as all of the carbon atoms, you can do so by using the `select` command followed by the type of atom you would like to select.
 - For example: `select carbon`

Quick Reference Standard Sizes for SMART Team Models

Standard Display Format Sizes

```
backbone 1.5  
wireframe 1.25  
spacefill
```

Standard Bond Sizes

```
hbond 1.0  
strut 1.0  
ssbond 1.0
```

Standard Clean Sidechain Sizes

```
spacefill 1.25  
wireframe 1.0
```