3D Printing Protein Structures Worksheet

Safety First!: Get an adult's permission before downloading any software or files onto your computer. Caution – be careful with the print, while it is somewhat tough, it is mostly intended to be looked at and could break if squished or dropped.

Introduction: While you may not be able to do the activity associated with the 3D-printed protein from home, we hope that this worksheet will help you be prepared to print your own protein at the Corona Public Library Maker Space soon! In the meantime, you can have fun learning about different proteins and looking at structures from the Protein Data Bank!

Martin Lab at UCI YouTube Video Link: https://youtu.be/m4EUzpv7lPw

Procedure *(modified from Reference #3)*

Set-Up:

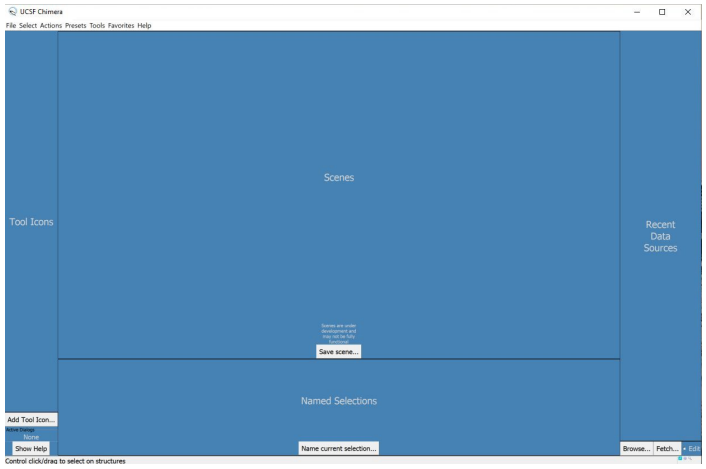
1. Download UCSF Chimera https://www.cgl.ucsf.edu/chimera/olddownload.html

Finding a Protein:

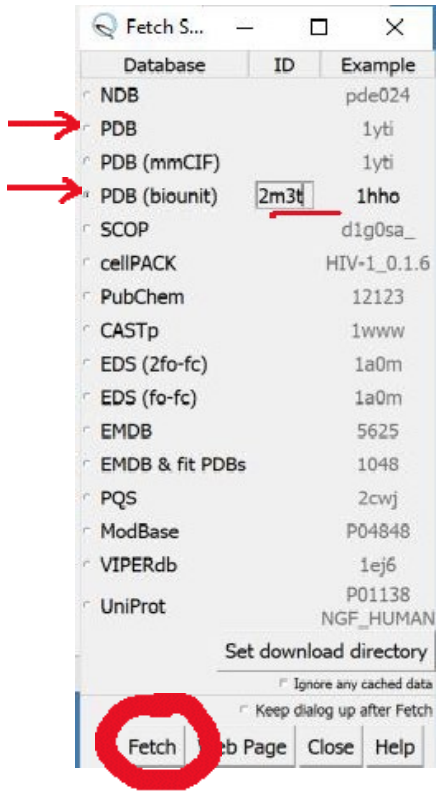
2. Go to https://www.rcsb.org/
    a. Use the search option to find a protein you are interested in.
    b. If you aren't sure, try searching "2M3T" which will bring you to the solution-state NMR structure of human gamma(S)-crystallin solved by the Martin lab at UC Irvine. This is a protein in your eye lens that helps you see by refracting light onto your retina!
3. Note the ID (four-digit combination of letters and numbers) or go to Download Files dropdown on the upper right and click the PDB Format.
    a. Note for proteins solved by NMR you may see more than one structure solved. This is called an ensemble above the 3D View on the left part of the page (this is because proteins can have parts that move around a lot; the ensemble represents a few of the lowest-energy conformations). If this is the case, when you open the Download Files dropdown, select Biological Assembly 1 in order to get only one structure.

Opening a Protein Structure in Chimera:

4. Open the UCSF Chimera program on your computer. The program should appear as shown below:



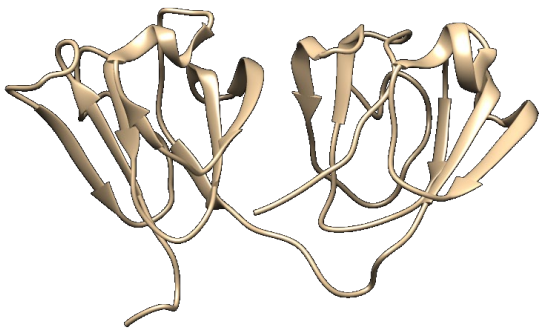
5. To open your protein structure file, do one of the following:
    a. Go to File → Fetch by ID. Then type in the ID you noted in the blank text box next to PDB(only structure) or PDB (biounit) (NMR ensemble) in the pop-up that appears. Then clickFetch on the bottom left.



OR

    b. Go to File → Open and select the .pdb (only structure) or .pdb1.gz (NMR ensemble) file for your protein. This may require scrolling to the left to find the folder where your file is saved, possibly in Downloads

You should now see a protein structure similar to that in the 3D View on the RCSB page. This representation is called a ribbon diagram. This makes it easy to see structural components of the protein and we think they look pretty cool this way too! Right click and hold to rotate your protein to see it fromdifferent orientations. Left click and hold to zoom in or out. Example shown is 2M3T.

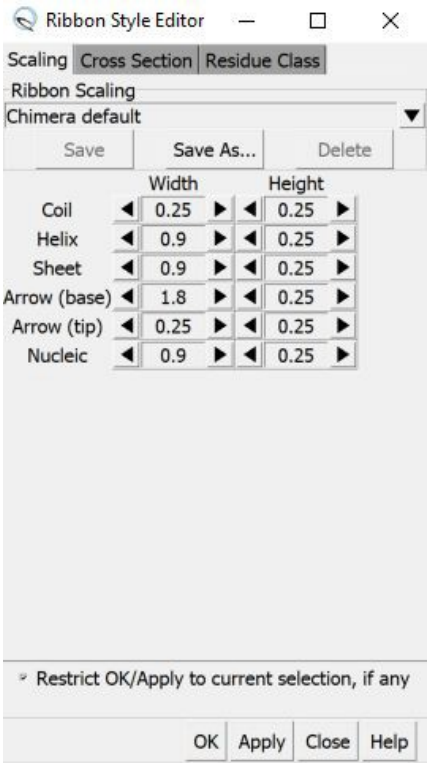


Preparing the Protein File for 3D Printing:

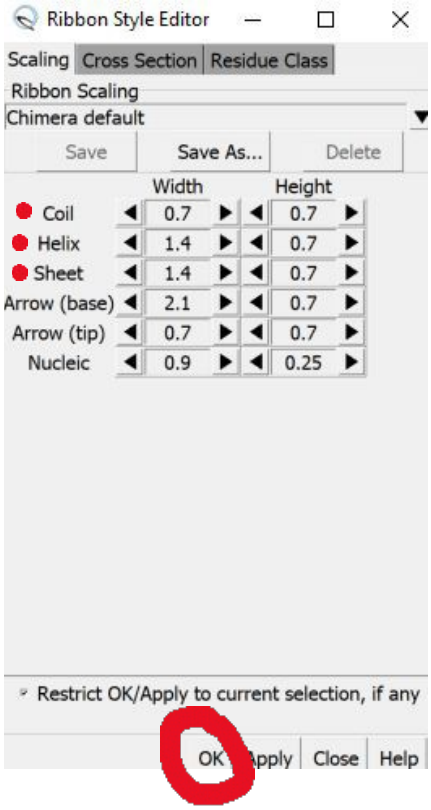
Note: The following steps will make the overall structure thicker, making it more rigid, which will likely help produce a successful print.

6. Go to Tools → Depiction → Ribbon Style Editor. A pop-up should appear.
7. Adjust the following parameters from the start values:
    a. Change the Height of every item to at least 0.7
    b. Change the Width of Coil to 0.7; Helix to 1.4; Sheet to 1.4; Arrow (base) to 2.1; Arrow (tip) to 0.7. Then click apply/OK.
    c. Below is a representation of the starting and ending parameters in the pop-up.

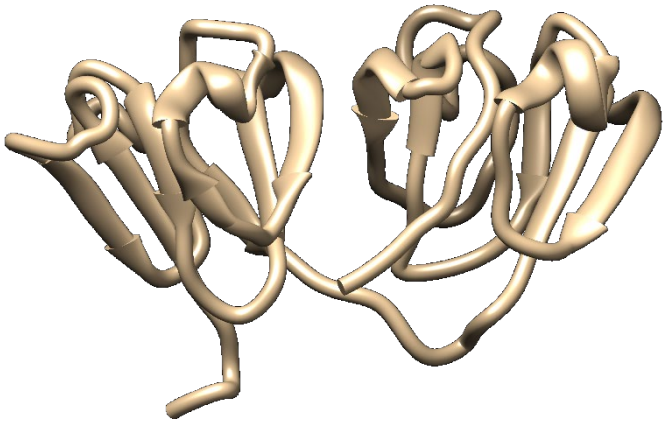
START



END

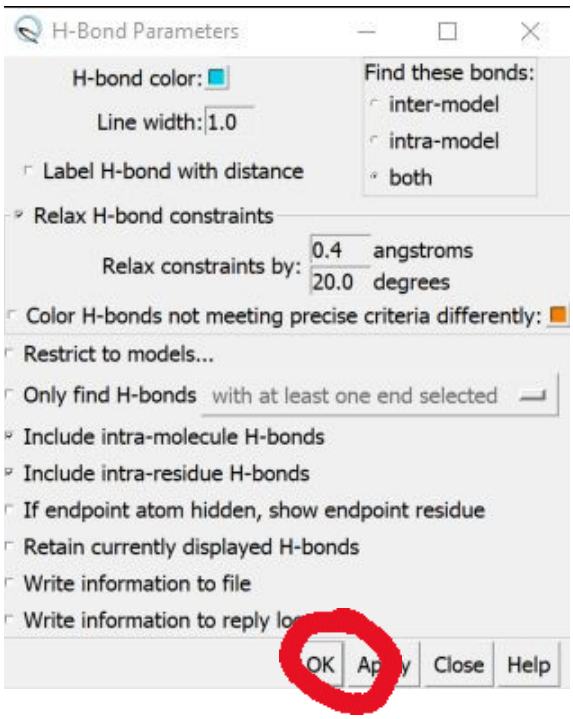


Your protein should now look thicker overall as shown below.

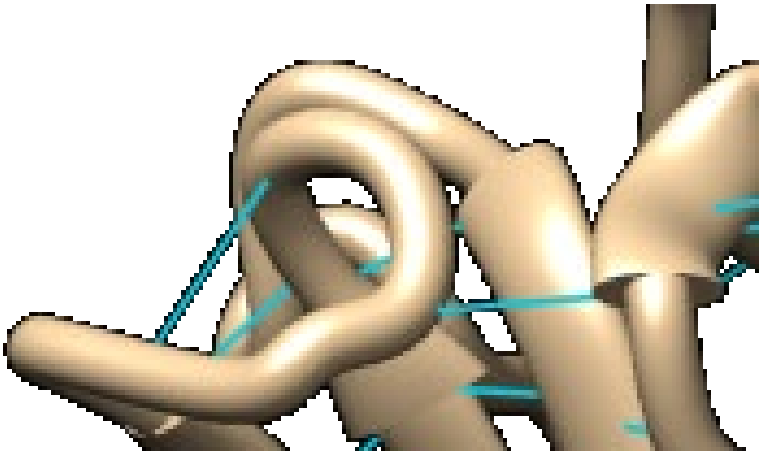


Next, we will add additional structure to help the printing.

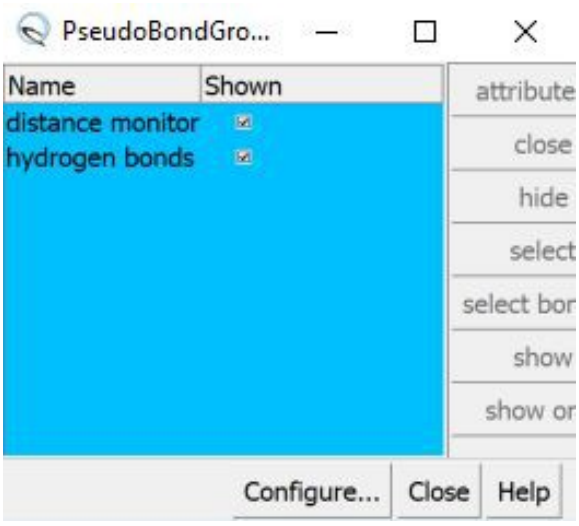
8. Go to Tools □ Structure Analysis → FindHBond. You don't need to change anything on thispop-up, just click OK.

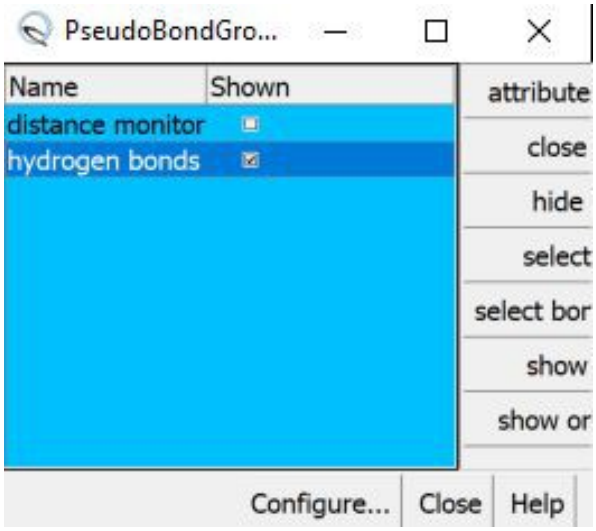


You should now see hydrogen bonds as blue lines on parts of the protein as shown below (zoomed in).



9. Go to Tools → General Controls → PseudoBond Panel. Another pop-up will appear as shown below. Ensure only hydrogen bonds is checked/selected and then click “attributes”.

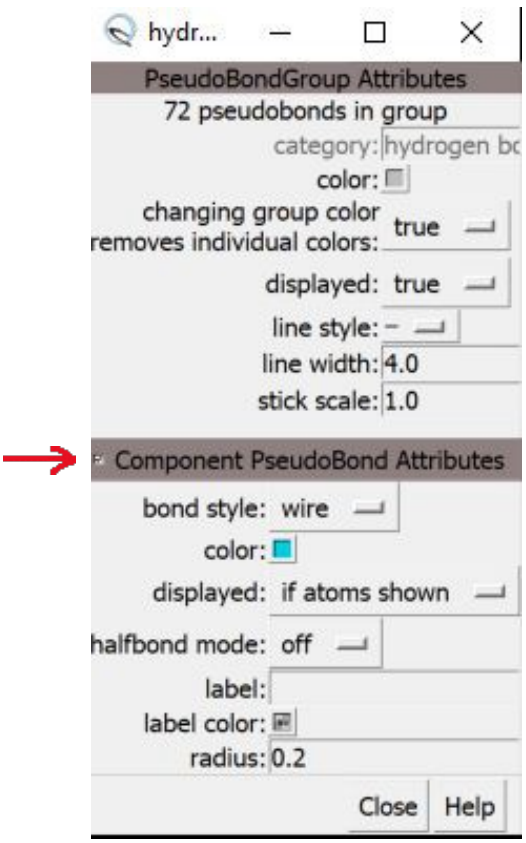




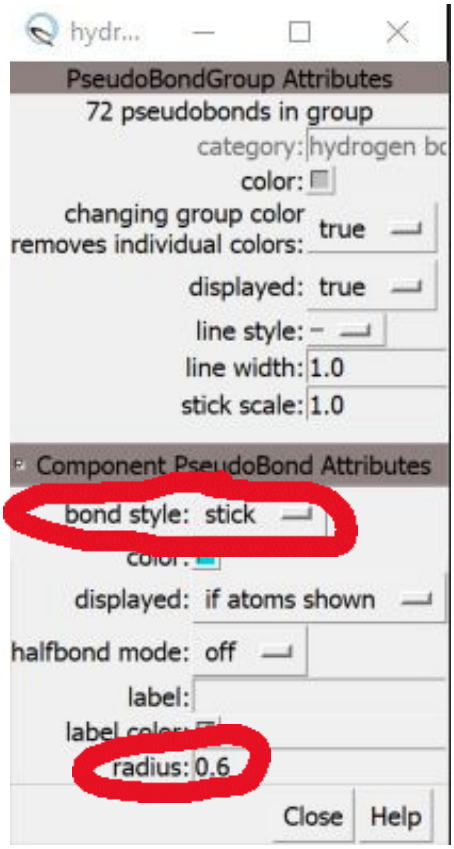
10. On the next display you will see the option Component PseudoBond Attributes. Check this to open it, shown with the red arrow in step 11.

11. Once open, change the bond style from “wire” to “stick” using the dropdown, and change the radius from 0.2 to 0.6.

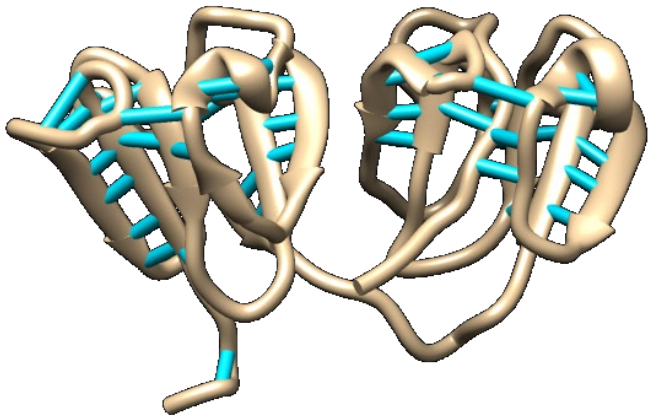
START



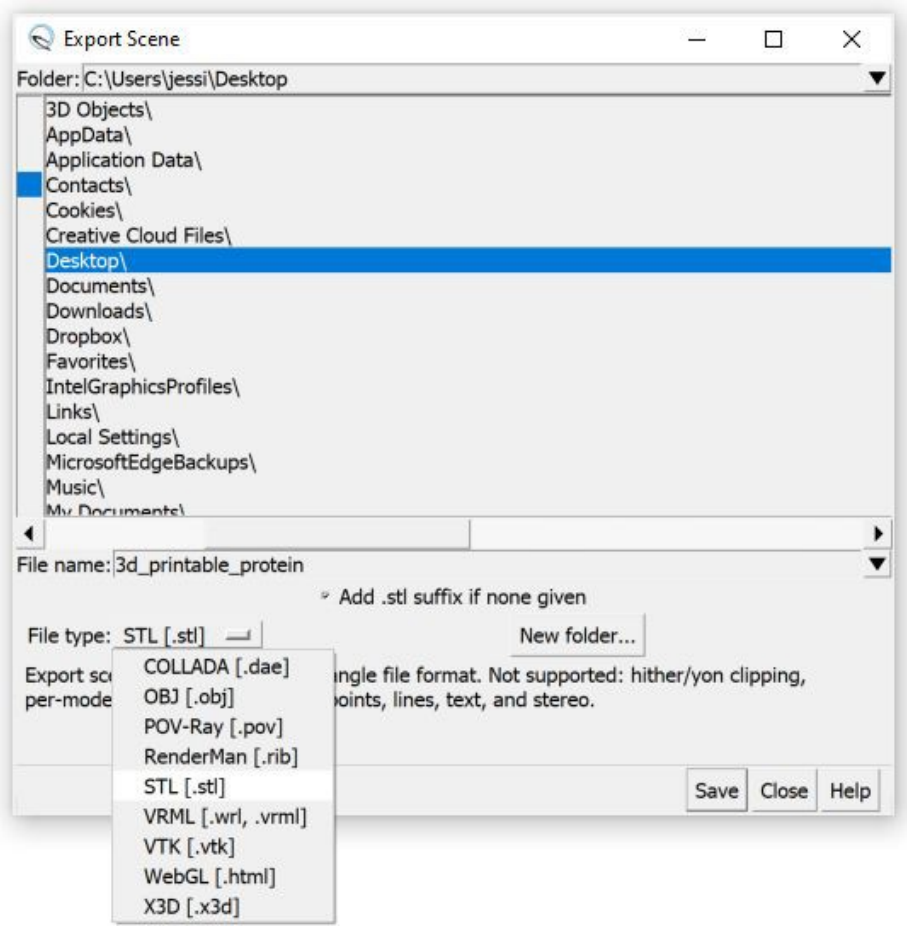
END



Your protein should now look something like what is shown below and it is almost ready to 3D print!



12. To save the file in a format that will be recognizable to most 3D printer software, go to File → Export Scene. Choose where to save your file, the name and in the File type dropdown choose STL.



Now you can take this file with you to the library to print your protein! Beyond this, the details of how to set up prints will vary some based on your printer and the material you are printing with so it is best to follow the instructions developed on site.

If your print fails start by discussing this with the expert staff at the Maker Space, because it might need adjustments to supports or other aspects of the print set-up. If that is not the case, consider revisiting step 7 and adjust parameters to be proportionally larger, or look up Reference #3 for other more detailed considerations and options. You can also try this with other proteins from the PDB – look up human insulin, or check out the molecule of the day.

Observations / Discussion:

1. What secondary structure elements do you see in the plant-specific insert (PSI) 3D print that came in your kit?

2. What do you notice about the secondary structure on the protein that you have modeled in Chimera? (Hint: it is something that you selected to show as part of the process).

3. How does the structure of the protein you chose from the databank compare to the PSI? What is the same? What is different?

Please feel free to write us and tell us which protein you chose and why! We look forward to talking with you more about protein structures and 3D printing!

Further References:

1. Just How Small Is An Atom? https://youtu.be/yQP4UJhNn0I
2. More about *Drosera capensis*: https://en.wikipedia.org/wiki/Drosera_capensis
3. 3D Printing of Biomolecular Models for Research and Pedagogy (written) https://www.ncbi.nlm.nih.gov/pmc/articles/PMC5408980/
4. 3D Printing of Biomolecular Models for Research and Pedagogy (video) https://www.ncbi.nlm.nih.gov/pmc/articles/PMC5408980/bin/jove-121-55427-pmcvs_normal.mp4
5. National Institutes of Health (NIH) 3D Print Exchange https://3dprint.nih.gov/