

UCSF Chimera - GroEL Visualization

The purpose of this exercise is to get familiar with [Chimera](#) features for density map display, and display of Protein Data Bank (PDB) models.

Outline of what follows:

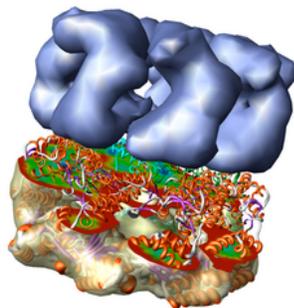
- [Density map basics](#)
- [Working with atomic models](#)
- [Hand aligning PDB models in density maps](#)
- [Image saving](#)

Followed by more advanced features:

- [Tricks for better images](#)
- [Density map slicing and coloring](#)
- [Using symmetry matrices](#)
- [Displaying b-factor as ribbon thickness](#)
- [Aligning protein domains](#)

And miscellaneous topics

- [Toolbar buttons](#)
- [Chimera manual - Help buttons](#)
- [Session saving](#)
- [More tutorials](#)



Starting Chimera

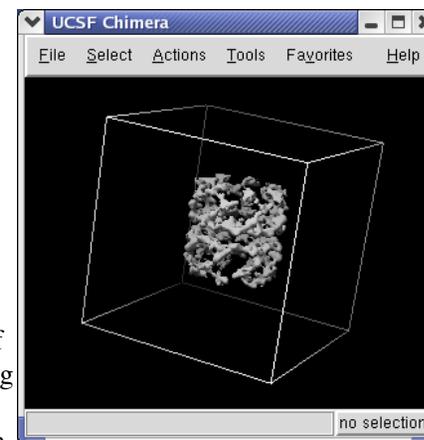
On **Linux** run the executable "chimera" in the bin directory of your Chimera installation. If Chimera is installed in /usr/local/chimera, then you run /usr/local/chimera/bin/chimera from a shell.

On **Windows** start Chimera by double clicking the Chimera icon in the directory called **bin** in your Chimera installation. If Chimera is installed in \Program Files then the executable will be found in directory \Program Files\Chimera\bin.

On **Mac** start the Apple X server found in /Applications/Utilities/X11. Then double click the Chimera application to start. The X server is not installed in Mac OS 10.3 by default and if you do not have it you can download it from [Apple](#). It is necessary for running Chimera on the Mac.

Open GroEL density map

Use the Chimera **File / Open** dialog to open **emd_1080.map**. In the **File Open** dialog you should change the **File Type** to **All (guess type)**. Initially it is set to only show Protein Data Bank files (*.pdb).



About this density map

This is a single particle reconstruction of GroEL determined by Steve Ludtke using EMAN software. GroEL is a cylindrical 14-mer that refolds misfolded proteins in its interior cavity. [This density map](#) and others are available from the [EM database](#) at the [Macromolecular Structure Database](#).

This GroEL map has 11.5 angstrom resolution. Another map determined by Steve having 6 angstrom resolution is also available. We'll use the lower resolution map because it is easier to see the main features.

Moving the map

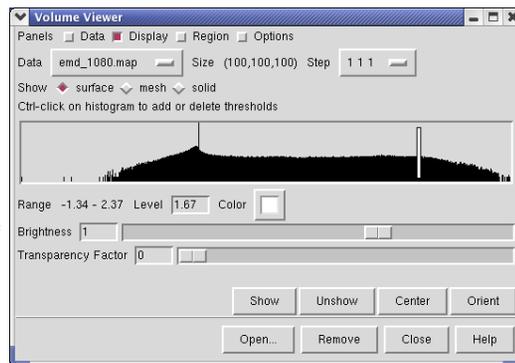
Dragging the mouse in the graphics window lets you rotate, translate, and zoom in on the map.

- **Left** mouse button - rotation
- **Middle** mouse button - translation
- **Right** mouse button - zooming

On a Mac with a one button mouse, hold down the Option key to get middle mouse button behaviour, or the Command key to get right mouse button behaviour.

Volume Viewer dialog

Controls for display of volume data (ie density maps) are in the **Volume Viewer** dialog that was displayed when you opened the file. If you close this dialog it can be shown again with menu entry **Tools / Volumes / Volume Viewer**.

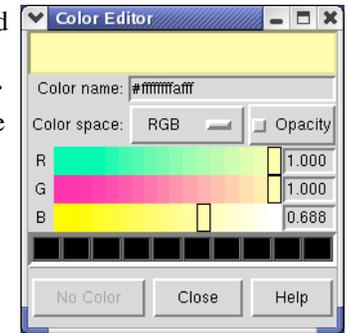


Contour level

The center histogram shows the distribution of data values in the density map. The range of data values (-1.34 to 2.37) is shown below the histogram. The white vertical bar on the histogram shows the contouring threshold. Drag this bar with the mouse to change the contour level. The numeric value of the contour level is shown below the histogram in the entry field. Contour levels can be typed in instead of being set with the mouse.

Color

Clicking on the white square with the wide raised border below the histogram shows a **Color Editor** dialog. The square button, called a **Color Well**, is used in many Chimera dialogs to choose colors and shows the current color in the center. Change the density map color by clicking on the color bars in the **Color Editor** dialog. You can **close** the dialog when you are done.



Mesh display style

Above the histogram is a radio button where you can choose mesh display style. This shows the surface as a mesh made up of triangles. You may need to zoom in to see the small triangles.

Data size and subsampling

Near the top of the dialog the data size, 100 by 100 by 100, is shown. The **Step** menu allows you to subsample the data. For example, a step of **2 2 2**, means to use every other density value along each of the 3 axes. Subsampling is useful with large maps to speed up moving and recontouring.

Outline box

The outline box shows the bounds of the data set. You can hide the outline box by clicking the **Options** checkbox on the right side of the top line of the **Volume Viewer** dialog. This displays a panel of options and the first one **Show outline box...** can be switched off. Clicking the **Options** checkbox again hides the long panel of options.

Brighter image

To make the density map display brighter use menu entry **Tools / Viewing Parameters / Effects** and switch off **Depth cueing** in the **Viewing** dialog. Depth cueing makes objects appear dimmer the further they are away from

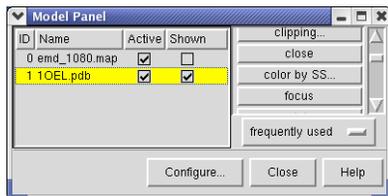
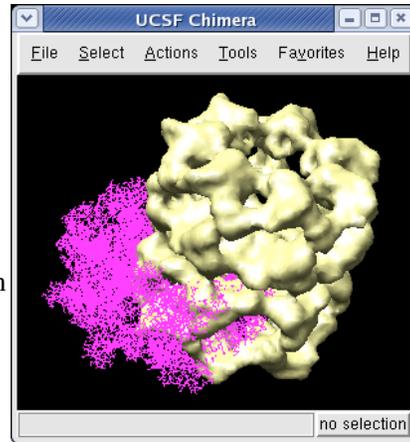
you. This will be discussed more in the section on [tricks for better images](#) It is a good idea to **close** Chimera dialogs when you are done so your whole screen does not become covered with dialogs.

Working with atomic models

We now look at an atomic model of GroEL determined by x-ray crystallography, and how this can be aligned with the density map.

Use **File / Open** to open Protein Data Bank model [1OEL.pdb](#). It is in the same directory as the density map. The atomic model of one half of GroEL will be shown shifted to one side of the density map.

We'll look at methods of atomic model display now. First hide the density map using menu entry **Favorites / Model Panel** and switching off the checkbox in the **Shown** column for **emd_1080.map**.

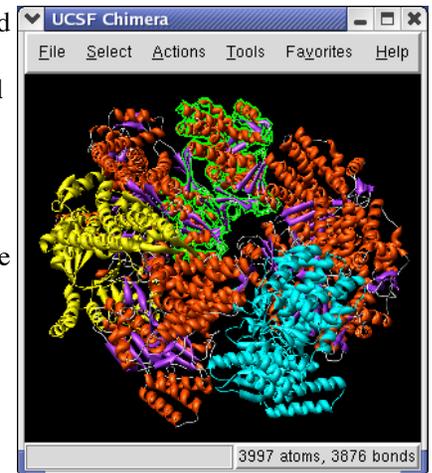


Showing a ribbon representation

To show a ribbon display of the model use menu entry **Actions / Ribbon / show**. To hide the original all-atom display use menu entry **Actions / Atom+Bonds / hide**. Zooming in shows a flat ribbon. To make the ribbon have an oval cross-section use **Actions / Ribbon / round**.

Coloring the ribbon secondary structure

To color the alpha helices, beta sheets and turns of the ribbon using separate colors, click on the **1OEL.pdb** line in the **Model Panel** dialog and then press the **Color by SS...** button in the righthand column of that dialog. The SS is short for secondary structure. Press the **OK** button on the color secondary structure dialog to use the default colors.



Coloring individual chains

To color one of the 7 proteins in this model differently, select a single residue using the mouse.

- **Ctrl left mouse button** - select objects

The selected residue will be highlighted with a **green outline**. It may be hard to see since it is small. Pressing the **up arrow key** on the keyboard (the one used for moving up one line in a text editor) will extend the selection to the entire chain containing that residue. Then use menu entry **Actions / Color / cyan** to color the selected chain.

Hiding a ribbon

To select a chain by its PDB identifier, use menu **Select / Chain** which contains entries **A, B, C, D, E, F, G** corresponding to the 7 chains in PDB model 1OEL. Choose any chain from the menu to select it.

To hide the selected chain use **Actions / Ribbon / hide**. You use **Ribbon / hide** instead of **Atoms+Bonds / hide** because the chain is displayed as a ribbon. The **hide** menu entries for the two styles are separate because you can have both ribbon and atoms/bonds showing at the same time.

Selecting more than one chain

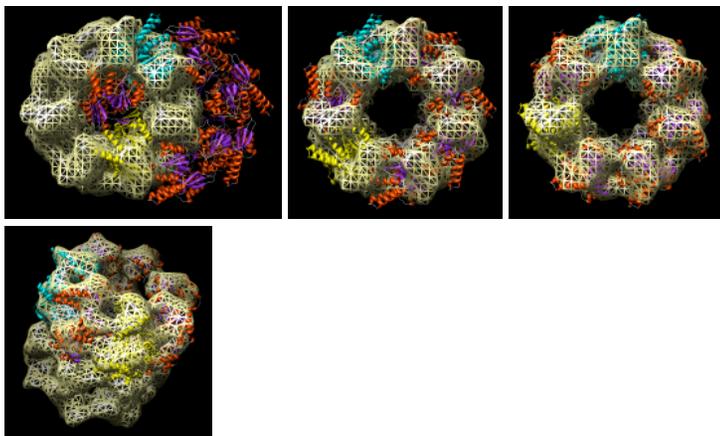
To select multiple chains, select a residue from one chain with **Ctrl left mouse button**. Then select a residue in another chain with **Shift Ctrl left mouse button**. Adding the **Shift** key extends the existing selection instead of replacing it. Then press the **up arrow key** to select both chains.

Slow rotation

When an object is selected the model may not rotate smoothly because drawing the green outline around the object requires a lot of computation. To clear the selection click **Ctrl left mouse** on empty space, or use menu entry **Select / Clear Selection**.

Hand align PDB model into density

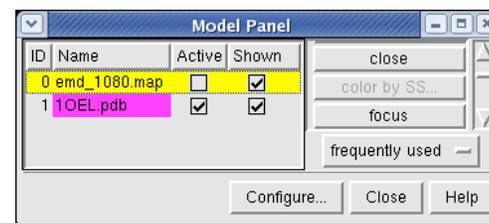
Now we will move the PDB model to align it with the density map.



Redisplay the density map using the **Model Panel** dialog (menu entry **Favorites / Model Panel**) by clicking the checkbox in the **Shown** column next to **emd_1080.map**. Display the density map as a **mesh** using the **Volume Viewer** dialog (menu entry **Tools / Volumes / Volume Viewer**).

Rotate with the mouse so you are looking directly down the axis of GroEL cylinder with the 1OEL model near you.

Now switch off the **Active** checkbox next to **emd_1080.map** in the **Model Panel** dialog. This is in the column before the **Shown** button and each model has a separate checkbox.



Active means that the model can be moved with the mouse. Now use the **middle mouse button** to drag the PDB model so that it is centered with the density map.

Rotating about the axis perpendicular to the screen

Once the PDB model is centered with the density map drag with the **left mouse button** near the edge of the graphics window to rotate the PDB model to match the shape of the density map. When you do a mouse rotation at the edge of the window, this rotates about the axis perpendicular to the screen. Using the mouse more towards the middle rotates the model about axes parallel to the screen as if you are grabbing the model. The boundary between these mouse rotation zones is not shown.

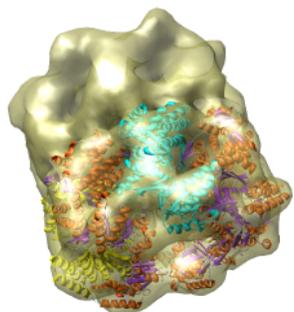
Turn on the **Active** button on the **Model Panel** for **emd_1080.map** so you can rotate both models at once and check the alignment from different view points.

Undoing accidental rotation

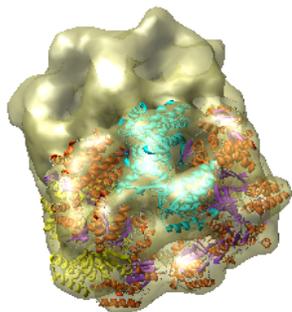
If you accidentally rotate the PDB model so its axis is not parallel to the density map cylinder axis, use menu entry **Tools / Utilities / Undo Move** to bring them back to having parallel cylinder axes.

Image saving

File / Save Image...



Screen shot (jagged edges)



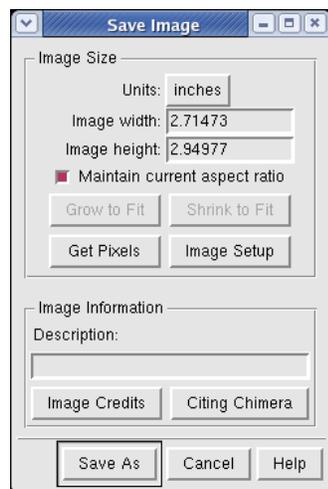
Use menu entry **File / Save Image...** and press the **Save As** button to save the graphics window image to a file. After the image is captured a dialog appears to choose the file name and format.

Image capture directly from screen

The Chimera dialogs will disappear and you will see magnified pieces of the image flash by in the graphics window. Chimera is capturing the image directly from the screen so no dialog should be placed on top of the Chimera window while the image is taken.

Supersampling

Using Chimera image saving will produce a higher quality image than taking a screen snapshot. The image is captured at a size 3 times larger than requested and then averaged down to the correct size. This is called **supersampling**. It reduces jagged edges.



Large image sizes

You can make images larger than the screen for publications and posters. Type a new value into the dialog width or height entry fields and press the **Enter** key. You only need to enter one of the dimensions. When you press **Enter** the other will be calculated to maintain the aspect ratio, and the image will be captured. To change the **pixels per inch** press the **Image Setup** button.

If you want an image exactly the same number of pixels as the Chimera window on the screen, press the **Get Pixels** button. This fills in the exact size in pixels of the Chimera graphics window.

White background

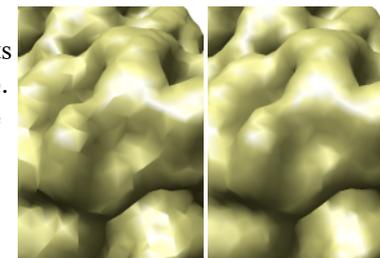
When making images sometimes you want the background to be white. Do this using menu entries **Actions / Color / background** followed by **Actions / Color / white**.

Tricks for better images

Smooth surfaces

Zooming in on the surface you can see facets from the triangles (use surface display style). To reduce these you can turn on the **surface smoothing**. Click the **Options** checkbutton at the top of the **Volume Viewer** dialog and check the **Surface smoothing iterations ...** checkbutton, the last entry in the options panel. The list of options is so long it may go off the bottom of your screen. You may need to hide the **Display** panel by unchecking the **Display** checkbutton at the top of the **Volume Viewer** dialog to see the full list of options.

Smoothing off. Smoothing on.



You can leave this option on -- its only serious drawback is that it can slow down recontouring the surface. To have make your changed **Volume Viewer**

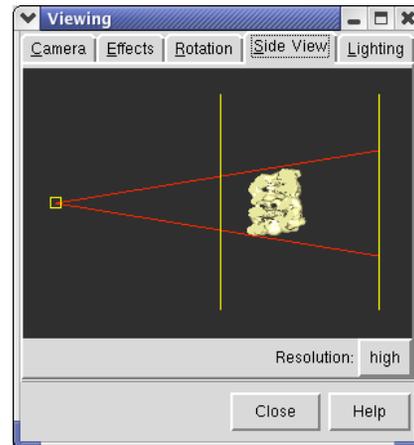
settings the defaults for future Chimera sessions press the **Save default dialog settings** button at the bottom of the **Options** panel.

Chimera settings are saved in a hidden directory called **.chimera** in your home directory in a file called **preferences**.

Depth cueing

Chimera dims parts of the scene that are further from you to give a sense of depth. This feature can be turned on and off using menu entry **Tools / Viewing Parameters / Effects**.

The dimming occurs between the **near and far clip planes**. The clip planes can be adjusted using menu entry **Tools / Viewing Parameters / Side View**. The **Side View** window displays the open models as seen from the side. Two vertical yellow lines indicate the front and back clip planes. The left line is the front clip plane. If you drag the front clip plane (yellow vertical line) towards the models with the mouse you will see the brightness in the main graphics window increase (provided depth cueing is turned on). If you move the front clip plane so it intersects the models, you will see the surface cut in the main graphics window.



The small yellow box at the left in the side view is your eye position. You can drag this left or right to zoom in and out.

Lighting and Shininess

The **Tools / Viewing Parameters / Lighting** menu entry lets you move the 2 Chimera lights. The **key light** provides the main illumination of your models while the **fill light** is intended to fill in dark areas.

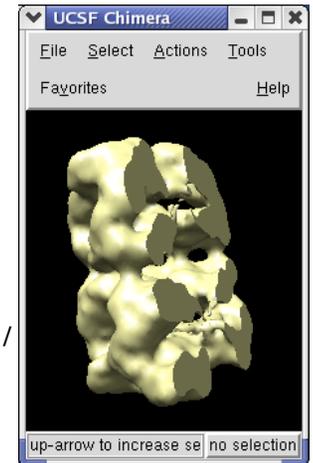
You can control how shiny the models appear using menu entry **Tools / Viewing Parameters / Shininess Control**. The shininess parameter effects

the size of **specular highlights**. Specular highlights are the white lighting spots on the model. The **brightness** parameter just effects the brightness of the highlights, not the whole surface.

Slicing surfaces

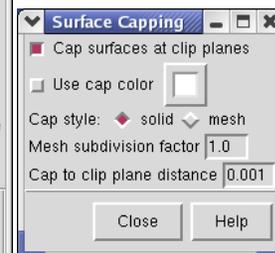
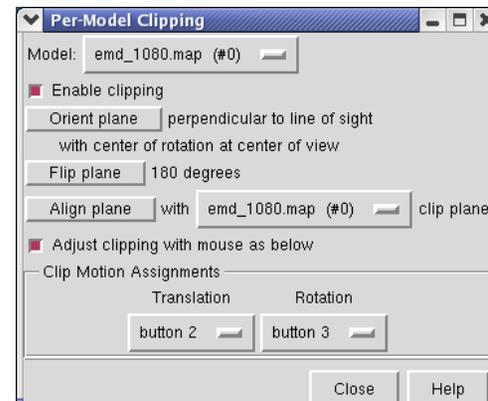
The **Tools / Graphics / Per-Model Clipping** menu entry lets you slice through surfaces (and protein models). Set the model to **emd_1080.map** and turn on the **Enable clipping** switch in the dialog that is shown. Then rotate the density map to see the cut.

The cut surface is visually confusing because you see inside of the surface. Use the **Tools / Graphics / Surface Capping** menu entry and switch on **Cap surfaces at clip planes**. This will cap the hole where the surface was cut.



Moving the clip plane

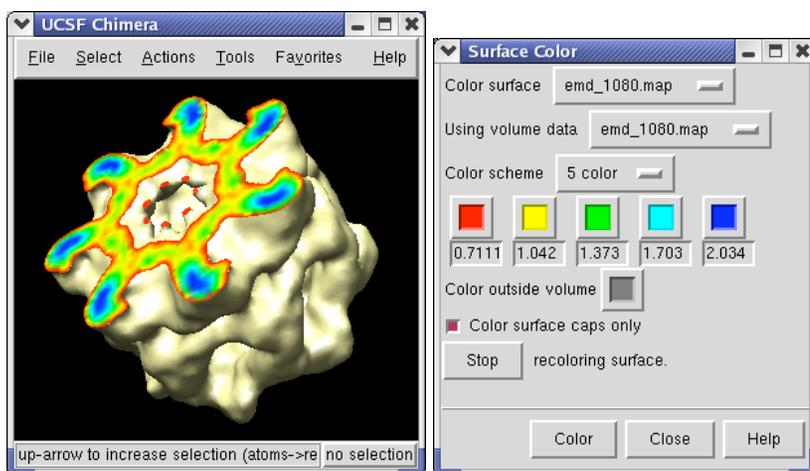
To move the cut plane, also called the clipping plane, switch on the **Adjust clipping with mouse...** switch in the **Per-Model Clipping** dialog. Then drag the clip plane with the middle mouse button or rotate it with the right mouse button. Turn off the **Adjust clipping with mouse...** switch to restore the mouse buttons their normal behaviour of translating models and zooming.



Surface coloring

The density map values vary across the clipped capped surface. The variation can be shown by coloring the cap using **Tools / Volumes / Surface Color**. Change the **Color scheme** in the **Surface Color** dialog to be **5 color**. (The 3 color color scheme is intended for display of electrostatic potentials.) To only color the cap switch on **Color surface caps only**. Then press the **Color** button at the bottom of the dialog.

When you move the clip plane, the coloring will be automatically updated.



Clip perpendicular to GroEL cylinder

To clip the GroEL density perpendicular to the cylindrical axis, first press the **Orient** button on at the bottom of the **Volume Viewer** dialog, then press the **Orient plane perpendicular to line of sight** button on the **Per-Model Clipping** dialog. The **Orient** button on the volume dialog rotates the density map back to the original orientation when the file was first opened (z-axis perpendicular to screen). After orienting the plane try moving it with the middle mouse button.

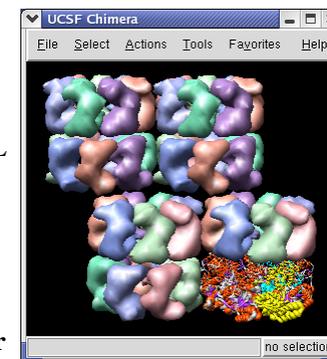
Accurate surface cap colors

If you change the contouring threshold in the **Volume Viewer** dialog by dragging the marker on the histogram, the colors on the capped surface will change by small amounts. Ideally they should not change and the problem is that the volume data is not being sampled at enough points on the cut surface. You can see the sampling by switching **Cap style** to **mesh** in the **Surface Capping** dialog. The **mesh** switch in the **Surface Capping** dialog effects just the cap, while the **mesh** switch in the **Volume Viewer** dialog applies to just the contour surface. Increasing the **Mesh subdivision factor** in the **Surface Capping** dialog to 2.0 (press **Enter** after changing the number for change to take effect) will cause cap vertices to be more closely spaced by a factor of 2. Moving the contour threshold then produces very little artifactual color variation, but updates are slower.

Crystal unit cell

The **1OEL** crystal structure has only one of the two 7 protein rings in its asymmetric unit. GroEL has two such rings stacked. The full GroEL can be seen by looking at the crystal unit cell.

Use menu entry **Tools / Multiscale / Multiscale Models** and near the bottom of the **Multiscale** dialog choose **Crystal unit cell** for the **Multimer type**, and press the **Make Models** button.



The **Multiscale** dialog makes copies of PDB chains using symmetry matrices. It displays the copies as low resolution surfaces. Each of the 7 chains of a ring is given a different color.

You will see 4 GroEL barrels. Each barrel consists of two rings of PDB model 1OEL stacked to form a cylinder.

Coloring monomers

You can select individual surfaces using **Ctrl left mouse button** and change their color with the **Color Well** button in the middle of the **Multiscale** dialog. If you select a surface and then press the **Copies** button near the top of the **Multiscale** dialog it will extend the selection to all copies of that chain.

Showing ribbons

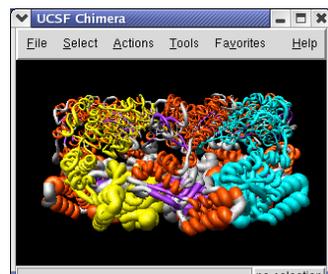
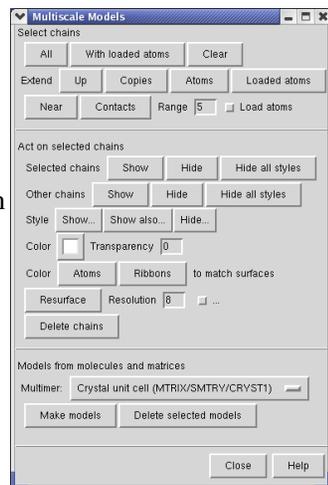
Press the **With loaded atoms** button in the selection pane at the top of the **Multiscale** dialog, and towards the middle of the dialog use the **Style Show...** button which pops up a menu and select **Ribbon**. This shows your original copy of 1OEL.

Deleting extra GroEL barrels

Delete the 3 extra copies of the GroEL barrel by selecting each barrel with the mouse and pressing the **Delete Chains** button in the **Multiscale** dialog. You can select a barrel with the mouse using **Ctrl left mouse button** and **dragging a box** around it. This selects everything in the rectangle you drag out (highlighted in green). To avoid selecting molecules you do not wish to delete it is best to drag a small box -- only a piece of each molecule need be in the box to be selected. Even if a molecule is hidden from view behind another one, it will be selected if it partially lies in the box you draw.

Displaying b-factor as ribbon thickness

The **1OEL PDB** model is a crystal structure that has a B-factor associated with each atom that indicates how well localized the atom is in the x-ray density. To display the B-factor using ribbon



thickness select the 1OEL.pdb line in the **Model Panel** and press the **Render by attribute** button in the right column of that dialog. It will take several seconds to scan the attributes of this large (27,000 atom) structure.

Choose **Attributes of residues** in the **Render by attribute** dialog, choose **Attribute** to be **average -> b-factor**, then switch to the **Worms** tab, and press **Apply**.

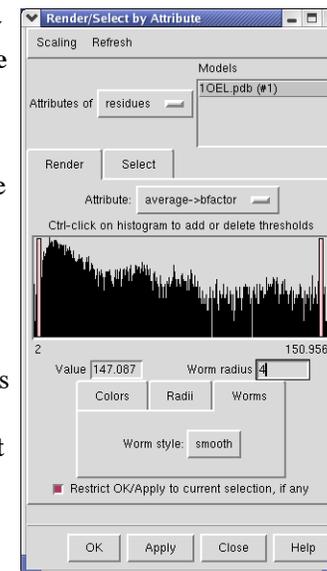
The ribbon thickness now corresponds to average residue b-factor value. The low b-factor face of 1OEL lie at the interface between the two 7-mer rings, and the high b-factor face is at the end of the barrel.

If you had a chain selected, only that chain shows the thickness variation. If you clear the selection (**Ctrl click** on empty space, or menu entry **Select / Clear Selection**) and press **Apply** again all chains will show b-factor.

Adjusting worm thickness

The **Render by attribute** dialog shows a **histogram** of the b-factor values. Vertical bars on the histogram mark B-factor values which have a specified thickness. For b-factor values between the bars the ribbon radius is linearly interpolated using the values defined for the bars.

Clicking on a histogram bar shows the corresponding ribbon thickness in the **Worm radius** entry field below. The bars can be moved and radii changed. Unlike the **Volume Viewer** dialog where moving the histogram bar causes an immediately updated display, here you need to press **Apply** to see the effect of your change.

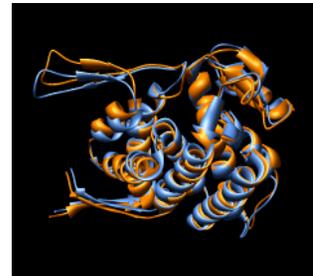
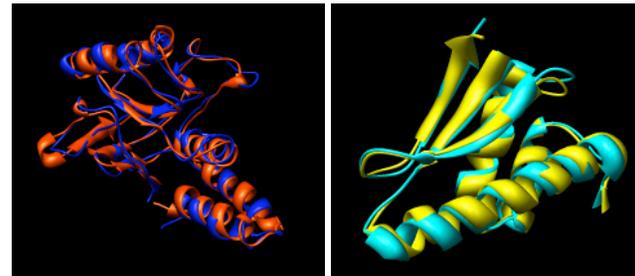
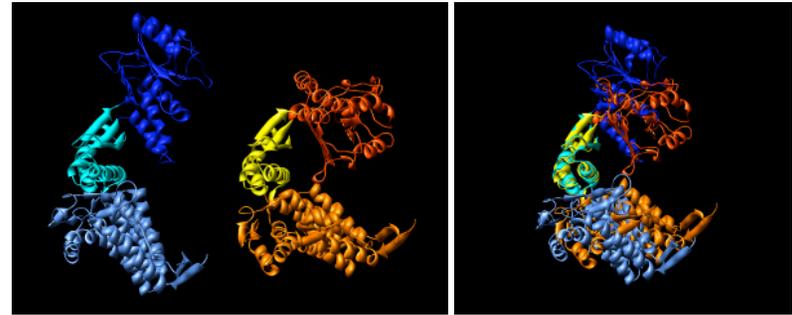
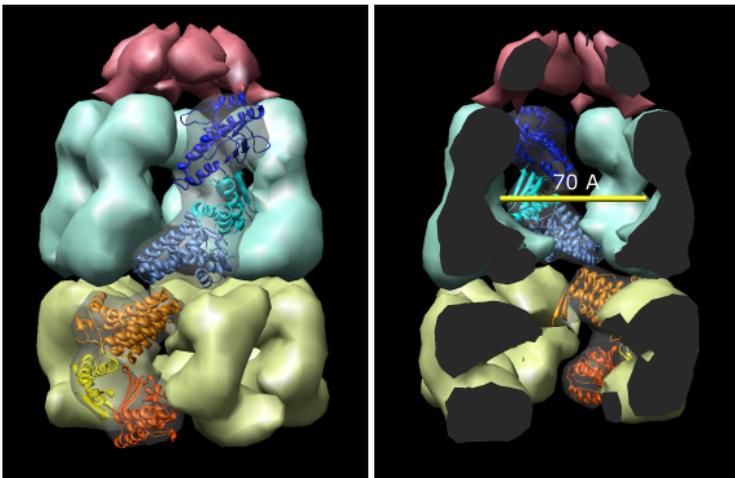


Aligning protein domains

GroEL adopts different conformations and our hand alignment of PDB model 1OEL and density map emd_1080 is only a first step in comparing this crystal and single particle EM data. The GroEL monomer has 3 domains that can assume different orientations relative to one another. A next step would be to computationally fit the individual protein domains into the EM map.

Chimera does not compute optimal positioning of PDB models or domains in density maps. You would need to use other software to do that.

Here are a series of Chimera images showing the 3 domains of the GroEL monomer. Two monomers in PDB model 1AON having identical amino acid sequence but different conformations are compared.



Several steps were used to produce these images that we will not describe here.

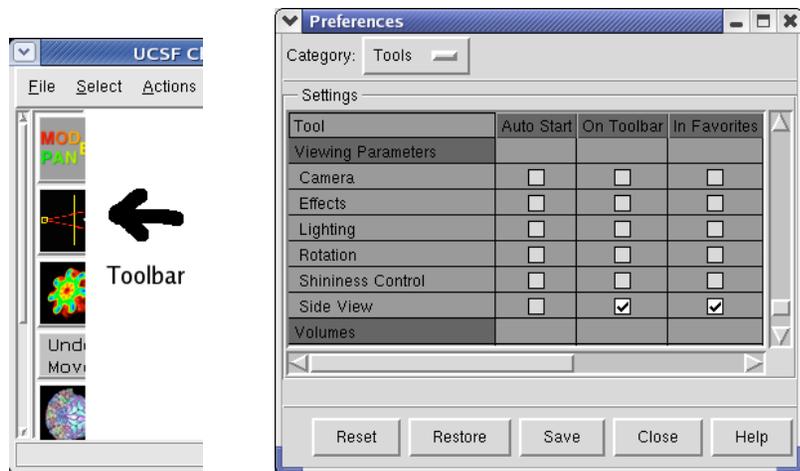
Some of the tools used to make these images are:

- **Multiscale Models** - menu Tools / Multiscale
- **Scale Bar** - menu Tools / Utilities
- **Match Maker** - menu Tools / Homology
- **Multalign Viewer** structure matching capability - menu Tools / Homology

- **Model Panel** write PDB - menu Tools / Inspectors or Favorites

Toolbar buttons

You can put buttons for frequently use Tools on the left side of the main Chimera window using **Favorites / Preferences....**



The **Preferences** dialog has several **categories** of preferences. Choose the **Tools** category. The entries in this list of tools are exactly those in the **Tools** menu, in the same order. Check buttons in the second column of the scrollable list of tools to put a button that invokes that menu entry on the **Toolbar** at left side of the main window. For example, check **Inspectors / Model Panel** and **Viewing Parameters / Side View**.

If you want these buttons to appear every time you start Chimera, press the **Save** button at the bottom of the **Preferences** dialog.

Chimera manual - Help buttons

Pressing the **Help** button at the bottom of any Chimera dialog will show the appropriate section of the Chimera manual (included with the Chimera distribution) in your web browser. Almost all Chimera dialogs have these help buttons. The user manual documentation is very thorough and worth looking at. Also look at the **Help** menu on the main window.

Session saving

You can save your Chimera session with **File / Save Session As....** Later you can restart Chimera and use **File / Restore Session** to redisplay your models. This will save the PDB models, Multiscale surfaces, density map display settings, colorings, background color, and many other attributes. **It does not save all aspects of the session.** The surface clipping, capping and coloring shown before are not saved in our current version, but may be in future releases.

The session is saved as a Python file (.py file suffix). It saves a copy of the PDB data but does not copy the density map data. For density maps it saves the path to the original file. For large PDB models such as 1OEL (27,000 atoms) saving and restoring the session can take a few minutes.

More tutorials

Chimera has many tools not explored in this tutorial. Most are for analyzing and comparing protein structures and sequences.

More tutorials can be found in the Chimera User's Guide using the Chimera **Help / Tutorials** menu entry, or possibly more recent ones on the Chimera web site <http://www.cgl.ucsf.edu/chimera> in the **User's Guide**.