Graphical visualization of ligand-receptor complexes

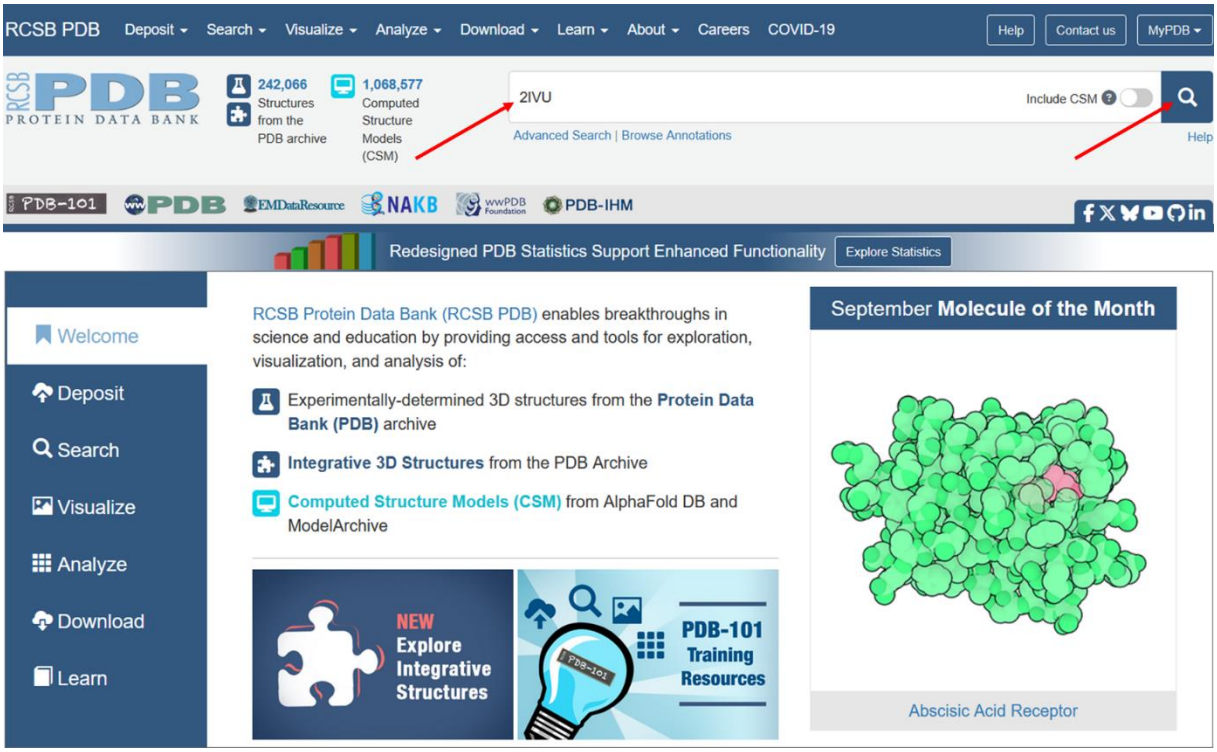
1. Introduction

The graphical visualization of ligand-receptor complexes will be carried out using the Chimera program. UCSF Chimera is a highly extensible program for interactive visualization and analysis of molecular structures and related data, including density maps, supramolecular assemblies, sequence alignments, docking results, trajectories, and conformational ensembles. High-quality images and animations can be generated. This program can be downloaded free of charge for academic, government, non-profit, and personal use (https://www.cgl.ucsf.edu/chimera/download.html). Chimera is developed by the Resource for Biocomputing, Visualization, and Informatics and funded by the National Institutes of Health.

2. Getting started

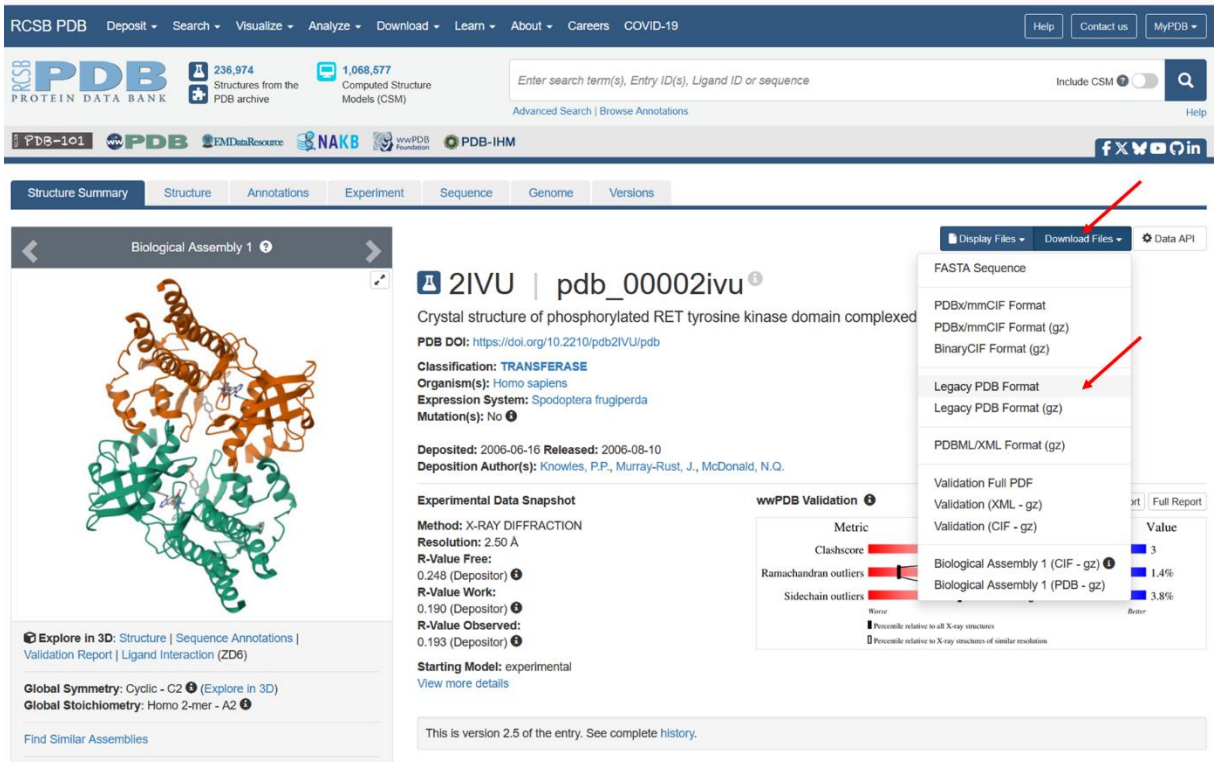
Prior to use Chimera, we need to download an X-ray structure from the Protein Data Bank (PDB):

- Open Google Chrome (from the "Applications" menu) and go to the PDB homepage (https://www.rcsb.org/)
- In the search panel, digit the code 2IVU and press the Search button

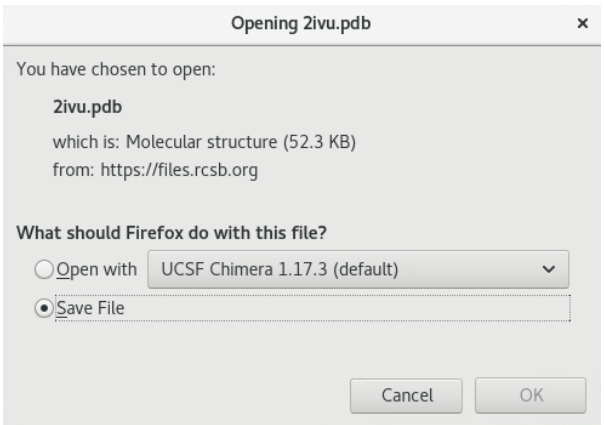


The page related to the desired X-ray structure (PDB code 2IVU) will be loaded.

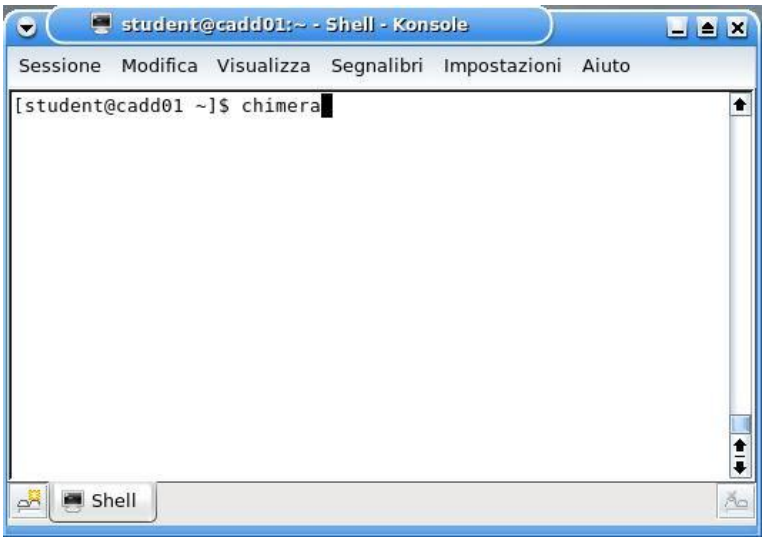
- Now click on the "Download Files" button and select "Legacy PDB format" from the drop-down menu



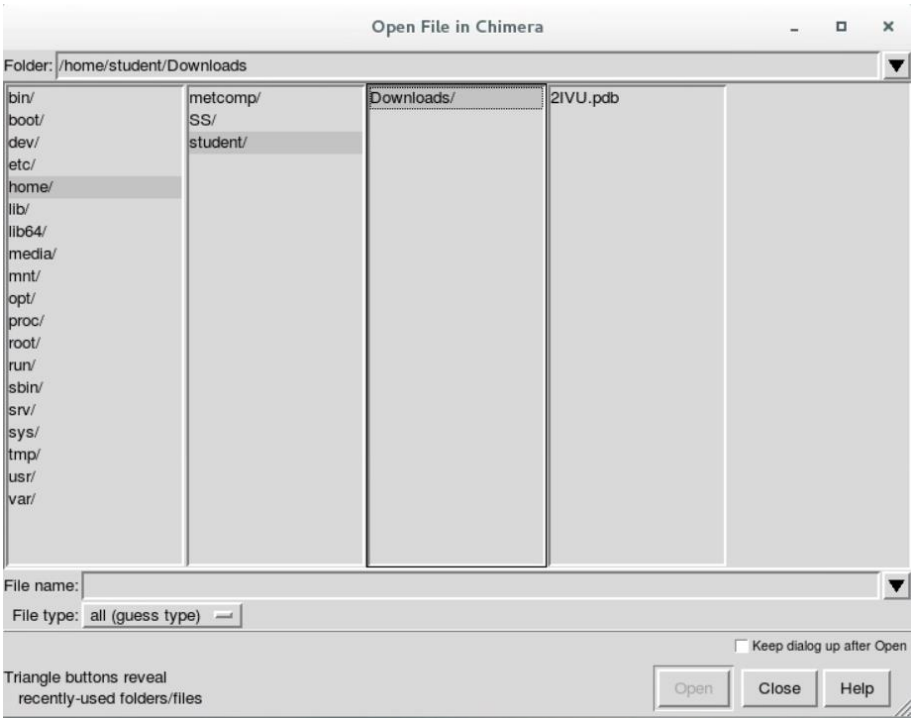
The browser should automatically save the file in the "Downloads" folder of your home directory. Otherwise, if the following popup window appears, select "Save File" and click "OK". The file will be saved in the "Downloads" folder of your home directory.



To open the software Chimera, open a linux shell (Applications/Terminal), type “chimera” within the shell and press Enter.



Now we have to open the X-ray structure that we just downloaded: click on (with left mouse button) File/Open, select the file 2IVU.pdb from the “Open File in Chimera” window, and click on “Open”.



3. Mouse Manipulation of Models

Many operations within Chimera can be accomplished in multiple ways. For example, colors and molecular representations can be changed by using the Actions menu or by typing commands into the Command Line. In general, using the menus involves more steps, but does not require one to remember commands and their syntax.

By ***default***, using a three-button mouse, models are:

- ✓ **rotated** with the left mouse button in the graphics window. Rotation is about the **X** and/or **Y** axis when the cursor is in the central region of the graphics window (the cursor becomes a small circle) and about the **Z** axis when the cursor is in the periphery of the graphics window (the cursor becomes two curved arrows in yin-yang configuration).
- ✓ **XY-translated** with the middle mouse button (the cursor will look like a cross formed by two double-headed arrows).
- ✓ **scaled** with the right mouse button (the cursor will look like a diagonal double-headed arrow enclosing a small square); movements downward and/or to the right increase the scale, whereas movements upward and/or to the left decrease it.

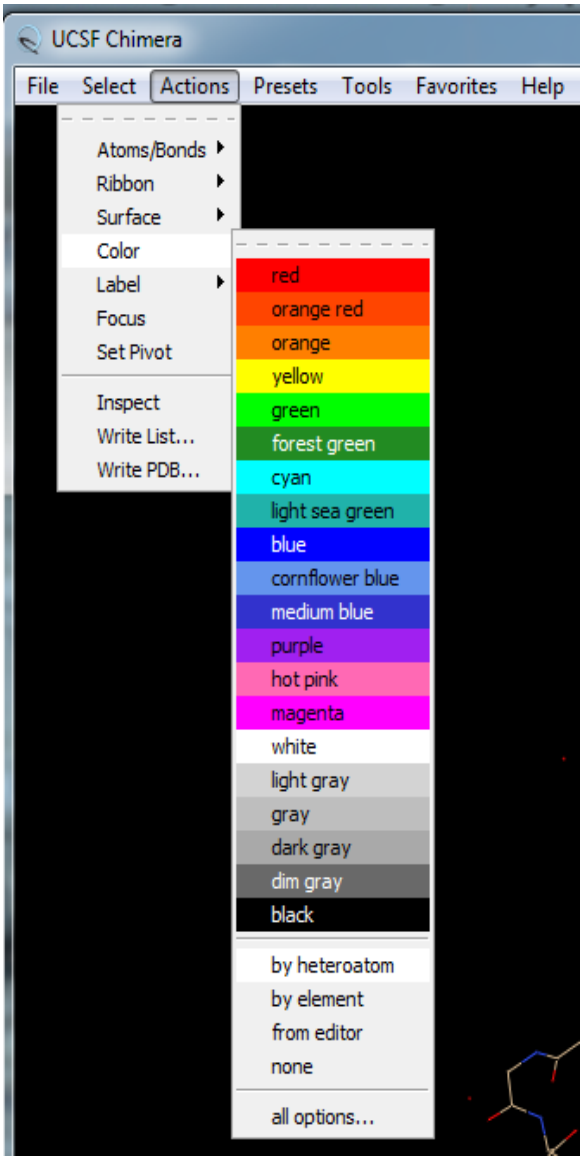
Also by ***default***,

- ✓ **Ctrl**-left mouse button performs picking (selection from the graphics window); the cursor will look like a pointing hand.
- ✓ Placing the mouse cursor close to an atom or bond (without clicking any button) will cause the corresponding label information to appear.
- ✓ Additionally, holding down the **Shift** key reduces the speed (mouse sensitivity) of manipulations in the main window by a factor of 10.

4. Molecular Selection

First of all, we will colour the RET kinase X-ray structure by atom type.

- Click on Action/Color

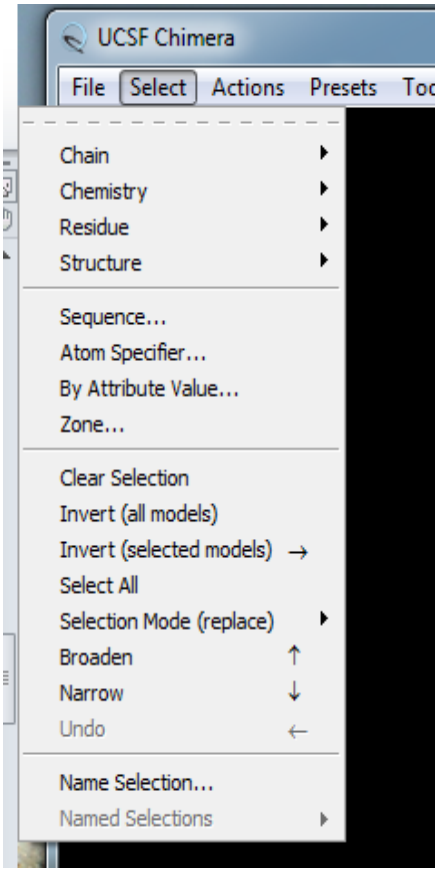


As shown in the Figure you have many possibilities for colouring the structure, you can choose a particular colour or you can choose to colour the atoms on the basis of their atom type ("by element") or colour only the heteroatoms ("by heteroatom").

- Click on Action/Color/by element

4.1 The Selection Menu

- Click on Select



The Selection menu will appear.

Clicking on Chain you can select one single chain, clicking on Chemistry you can select a single element, a particular functional group or an atom type. This last option is very useful, for example, for selecting all the nonpolar hydrogens in the refined proteins (this is possible clicking on Select/Chemistry/IDATM type/HC). Now they are absent, as in all the crystallographic structures.

Clicking on Residue you can select a particular aminoacid type or, very important, a non-standard residue such as the ligand.

A very important panel is the Zone panel, it allows to select atoms and bonds within (or beyond) a specified distance from the currently selected atoms, on a per-atom or whole-residue basis. We will better describe this panel below.

The other important commands are:

Clear Selection: clear (deselect) the current selection

Invert (all models): select all currently unselected atoms and deselect all currently selected atoms.

Invert (selected models): in models containing selections only, select all currently unselected atoms and deselect all currently selected atoms.

Select All: select all open models and their constituent parts.

Selection Mode (*current_mode*): control whether a new selection *via* the menu will be added to (append), subtracted from (subtract), intersected with (intersect), or used to replace the existing selection (replace).

Undo: undo the most recent selection operation.

4.2 The manual selection

You can also select atoms using the mouse, for selecting one atom press “Ctrl” and then click with the left button on one atom. For adding a selection hold down the “Ctrl” and “Shift” key and click with the left mouse button on the atom. If you already have one atom selected, you can press the up arrow key (on the keyboard) to expand the selection to the whole residue, then again for the whole chain, and again for the whole molecule model. To go back down, you could use the down arrow key.

For deselecting everything you can a) press “Ctrl” and click with the left button on a blank area or click on Select/Clear Selection for the menu.

4.3 Selection by the Command Line

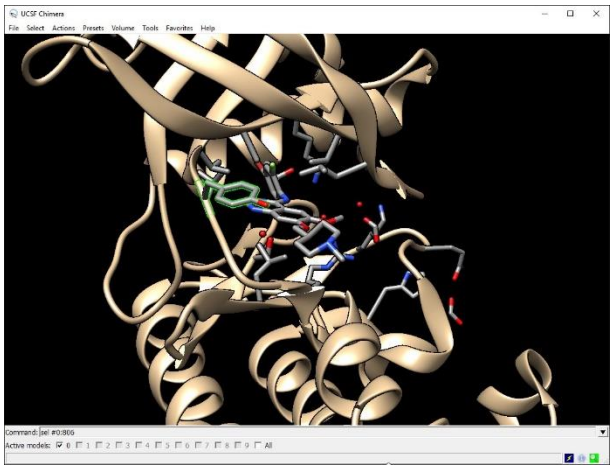
- Click on Favorites/Command Line

The Command Line will appear in the low part of the program. A Chimera command may include arguments and/or an atom specification.

Symbol	**Meaning**	**Usage**
#	model	#*model* (model ID number)
:	residue	:*residue* (residue name or number)
@	atom	@*atom* (atom name)

For selecting a particular residue (for example Tyr806) type in the Command Line:

- sel #0:806



5. Molecular Representation

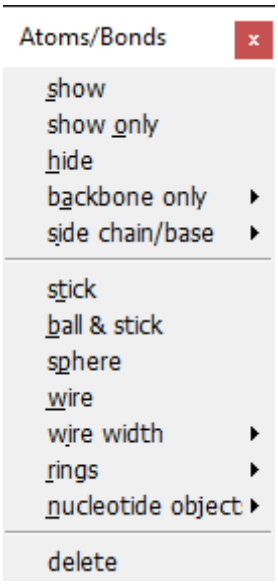
Clear the selection (Ctrl-left mouse button on a blank area or Select/Clear Selection from the Menu)

5.1 The Actions Panel

Which items are affected by Actions (the targets) depends on the current selection. When nothing is selected, the various operations act on all open molecule models and their associated molecular surfaces. Differently, if something is selected, all the action commands will modify only the selected portion.

- Click on Actions/Atoms/Bonds

A new panel will appear, showing the actions executable on the Atoms and Bonds.



show

show only

hide (only from display)

backbone only: visualize only the backbone of the residues

side chain/base: visualize the side chain of the residues

stick: stick representation

ball & stick: ball-and-stick representation

sphere: sphere or CPK representation

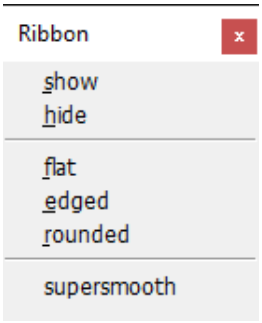
wire: wire representation

wire width: select the thickness of the wire representation

ring: modify the representation of the aromatic rings

delete: remove atoms/bonds (caution: irreversible)

- Click on Actions/Ribbon



Ribbons are only drawn for proteins and nucleic acids. By default, showing ribbon hides the corresponding main chain atoms.

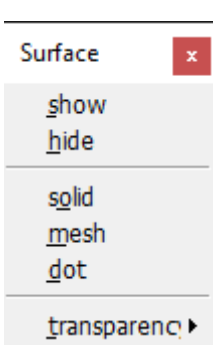
show

hide

There are different 3D ribbon visualizations, the most common are flat, edged and rounded

- Click on Actions/Surface/Show

Chimera shows ***solvent-excluded*** molecular surfaces, composed of probe contact, toroidal, and reentrant surface. The surface representation options (solid, mesh, and dot) always apply to entire molecular surface models even though the targets may be narrower.



show

hide

solid

mesh

dot

transparency: atom-level surface transparency

- Click on Actions/Focus

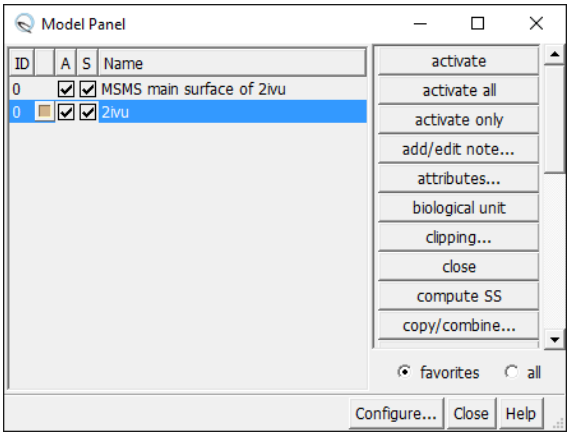
This command focuses the view on the target atoms, bonds, and/or ribbon segments that are selected, if nothing is selected then it will focus the view on the displayed atoms.

6. The Model Panel

Each file of coordinates opened in Chimera becomes a model with an associated model ID number. Non molecular models are also given ID numbers.

The Model Panel lists the models in Chimera and conveniently enables many operations upon them.

- Click on Favorites/Model Panel



In this panel are stored many functions:

activate: activate for motion (it allows to move single models) (CAUTION: also the "A" tagged has the same function; if you de-tag the "A" column, you de-activate the motion of the molecule)

activate all: activate all models even if not chosen in the left side of the panel

activate only: activate the chosen model(s) and deactivate all the others

add hydrogens: add hydrogens to the chosen model(s)

attributes...: list model attributes and allow changes

close: close (remove) the model(s)

deactivate: deactivate for motion; the opposite of activate

show all atoms: show the chosen model(s) *and* display all atoms within them

show only: show the chosen model(s) and hide all of the others

surface main: for the chosen model(s), display the molecular surface of any atoms categorized as main

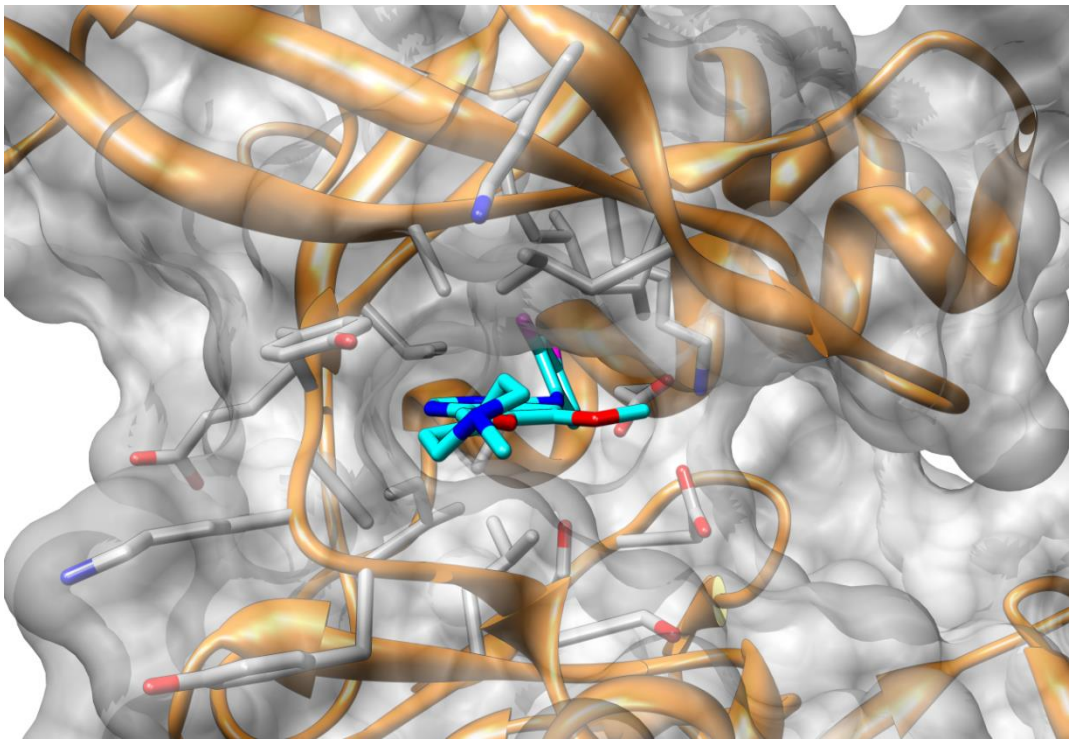
7. Presets

A preset is a predefined combination of display settings. Choosing an entry in the Presets menu applies its settings and is much easier than adjusting the many settings individually. Presets are provided for a handful of usage scenarios; of course, many more combinations of settings are possible.

- Interactive presets are meant for interactive manipulation and analysis. They may change what items (atoms, ribbons, surfaces) are displayed and how they are colored. The ribbons setup shows atomic detail for all nucleic acid residues, and for any amino acid sidechains and other residues within 3.5 Å of a ligand residue or metal ion. A hydrophobicity surface shows amino acid hydrophobicity in the Kyte-Doolittle scale with colors ranging from dodger blue for the most hydrophilic to white at 0.0 to orange red for the most hydrophobic. Surfaces of nonpeptides (which do not have Kyte-Doolittle hydrophobicity values) will be colored to match the underlying atoms instead.
- Publication presets are intended for generating images for presentation and publication. They do not change what items are displayed or their colors, but may change their styles and the color of the background. For example, choosing a preset with rounded ribbon or licorice will not turn on ribbon display, but will adjust any currently displayed ribbon segments; similarly, only the currently displayed atoms and bonds will be shown as sticks. Publication presets may decrease interactive performance because finer divisions are used to depict curved objects (molecular surfaces, ribbons, etc.). Individual display parameters are discussed in more detail in the tips on preparing images.

8. Example

In this section we will developed a picture of the inhibitor ZD6474 complexed with the RET kinase.



1) Close the current section

- Click on File/Close Session

2) Open the X-ray structure

- Click on File/Open, select the file 2ivu.pdb (which is in your Downloads directory) from the Open File window, and click on “Open”.

3) Colour the system by atom type

- Click on Actions/Color/by element

4) Remove the water molecules and the formic acid

- Click on Select/Residue/HOH

- Click on Actions/Atom/Bonds/Delete
- Click on Select/Residue/FMT
- Click on Actions/Atom/Bonds/Delete

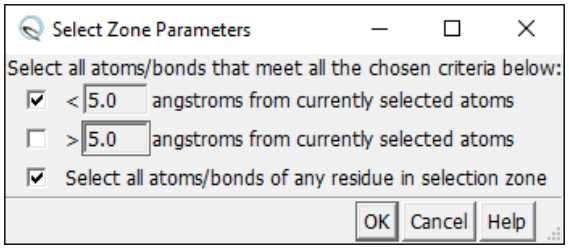
5) Ribbon of the protein

- Click on Actions/Color/all options…
- Select ribbons in the Color Action menu
- Click on the orange color
- Close the Color Action menu

6) Rendering of the inhibitor

- Click on Select/Residue/ZD6
- Click on Actions/Color/cyan
- Click on Actions/Color/by heteroatom

7) Visualization of the binding site



- Click on Select/Residue/ZD6
- Click on Select/Invert (all models)
- Click on Actions/Atoms/Bonds/hide
- Click on Select/Residue/ZD6
- Click on Select/Zone and change the parameters like the Figure; in this way the program will select all residues within a radius of 5 Å from the ligands. The activation of the third button ("Select all atoms/bonds of any residue in

selection zone") allows the visualization of entire residues and not only the residue fragments within a radius of 5 Å from the ligands.

- Press the OK button
- Click on Actions/Atoms/Bonds/Show
- Unselect (Click on Select/Clear Selection)

8) Create the surface

- Click on Favorites/Model Panel
- In Model Panel click on "surface main" (if it is hidden, you can tag the "all" option at right bottom of Model Panel and tag it as favorite)
- Click on Actions/Color/all options…
- Select surfaces in the Color Action menu
- Click on the light gray color
- Close the Color Action menu
- Click on Actions/Surfaces/trasparency/50%

9) Final rendering

- Rotate and translate the system for a good visualization
- Click on Presets/Publication 1

10) Save the session

- Click on File/Save Session as
- Enter the file name and click on save

11) Save the image

- Click on File/Save Image…
- Enter the image name, choose the Tiff format and click on save

9. Exercise

Do the same picture for the PDB code 3BGZ.