BIOC351: Proteins

PyMOL Laboratory #2

Objects, Distances & Images

Version 2.1

Valid commands in PyMOL are shown in green.

Let's fetch 1fzc, a crystallographic dimer of a fragment of human fibrinogen
How many chains? _____ (you can answer this with PyMOL or at the PDB)

Selecting and Objects

The power of PyMOL is the easy in which you can select/control objects and the attributes (color, visual representation, etc) assigned to them. To harness this power it is important we learn the vocabulary and operators that can be used.

The format of a select command is:

select NAME, chain a+b+g

where

NAME is the name you want to appear in the Objects Menu (it will appear in the list as (NAME) signifying it is a selection of an existing object)

Chain is from the Selector column from the list below you want to use

a+b+g are the 3 chains you want to select from the object

Handy PyMOL Selectors

Selector	Short Form	Macro Form	Description / Example
chain	c.	A+B//	select heavy, chain a select heavy, c. a select heavy, A//
resname or resn	r.	asp+glu/	select aa, resn tyr+trp select bases, a+g
resi, resid, residue, or resident	i.	1-10/	select nterm, resi 1-10 select activesite, resi 53+75+122
name	n.	*/CA+CB	Selects individual atoms by their name. select mainchain_atoms, name n+ca+c+o
hetatm	het		Selects everything labeled HETATM in the PDB file. select other, het

To use the select command we need to use the command line in the Upper Controls menu:

PyMOL>

Exercise B: Let's experiment!

Color each chain differently of 1fzc (**C** → by chain)

Figure out which chains are in which molecule (**L** → Chains)

So now lets just select a single molecule from 1fzc:

select molecule, chain __+__+__+__+__+__ (you fill in the blanks!)

Note when you use the select command, you should observe the following (see figure below):

1. On the left side of the Upper Controls window you should see your command as well as the result (how many atoms where selected)
2. In the Display area you should see all of the atoms selected above as pink diamonds
3. In the Object menu you should see (molecule) appear in the list

Note the changes in the three fields when using the select command. The command used to generate this image was: select molecule, chain A+B+C+I+G

With something selected you can change its color or its representation (i.e. make it a surface or sticks). The problem with a selection made in this manner is that you can't really separate it from the main object 1fzc. For example, click on 1fzc in the Object menu and (molecule) also disappears. Therefore I encourage people to <u>create</u> objects rather than select from an object. Objects copy the atoms from the original object to a new one, and therefore can have many attributes associated (color, type of representation, etc).

<u>Exercise C</u>:

Let's remove (molecule). To do that, A → delete selection
Now lets create an object:

create molecule, chain __+__+__+__+__+__ (you fill in the blanks)

What's different? Click on 1fzc to see some of the differences. Make another object:

create peptides, chain g+i

Change their representation to sticks and color them orange. Hide the cartoon of molecule and then put a white surface on it. Make the surface 80% transparent and turn back on 1fzc.

Now create some new objects using other selectors from the table above.

Building Complex Selections

The power of selecting comes from the ability to make refined selections so that you can isolate just what you want.
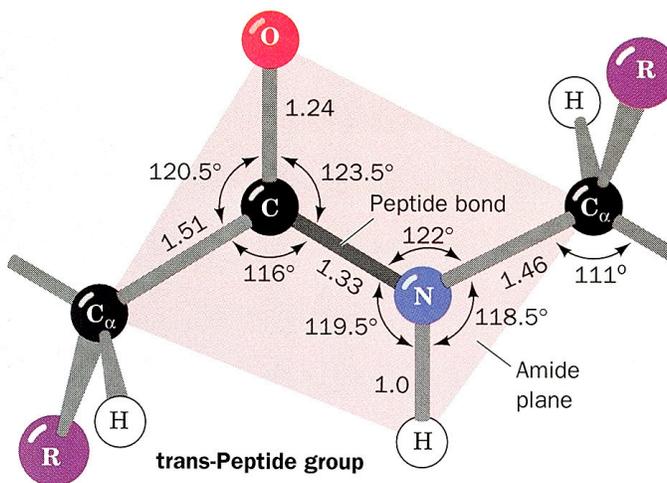
Nomenclature of the peptide backbone (Cα would be ca or CA when using the name selector).

Logical Selection Operators [†]

Operator	Short Form	Description Example
not s1	! s1	Selects atoms that are not in s1
		select activesite, (resi 123+233+256) not (name n+o+c)
s1 and s2	s1 & s2	Selects atoms in both s1 and s2
		select ligands, chain a and resi 23+67+89+123

[†] Create may (should) be substituted with these commands.

Use parentheses to ensure operations are preformed in the correct order. For example,
    create cleft, (chain a or (chain b and (not resi 31-55)))
which creates an object of all atoms not in residues 31-55 but in chain b to which it
adds all atoms in chain a.

Proximity Operators [†]

Operator	Short Form	Description Example
s1 around x	s1 a. x	Selects atoms with x Å of s1 select with5A, resi 336 around 5
s1 expand x	s1 e. x	Selects all atoms with x Å of any atom within s1 select cleft, within5 expand 5
s1 extend x		Selects all atoms with x bonds of s1 select region, resi 34 extend 4
s1 with x of s2	s1 w. x of s2	Selects atoms in s1 within x Å of the centers of atoms in s2 select bb10, (name n+o+c) within 10 of resi 234

[†] Create may (should) be substituted with these commands.

Selection Macros
        Sometimes a selection like this is cumbersome:
            create 142, 1uwh and chain b and resi 142 and name ca
        So this is also acceptable,
            create 142, /1uwh//b/142/ca
            Which signifies:/entity/segment/chain/residue/atom

Exercise D:
        Open the session you saved on Monday. Does it look like you remembered?
        Create an object representing a random set of amino acids.
        Display the structure as cartoon and visualize your random amino acids on it.
        What do you observe? Now lets create a new object, using the same AA but this
        time include (and not name n+o+c). What did you do and why? _____
        _____
        How does that look with your cartoon structure?_____
        Either create separate objects for different chains or split your chain in half.
        Color each differently and display a surface.

New Menu Buttons

No visualization program is useful unless you can measure the distances or angles between things.

Wizard

Measurement

This brings up a menu (seen to the right). By default it measures distances between each two clicks and creates an object. Click on Distances to measure something else!

| Measurement |
| Distances |
| Create New Object |
| Delete Last Object |
| Delete All Measurements |
| Done |

Measurement window.

Exercise E:

Let's check some standard values:

C – C bond _____	C = O bond _____
C – N bond _____	S – S bond _____

What about a hydrogen bond—how long are they? _____ (remember how to display H-bonds? (Action → Find → polar contacts → involving side chains)

More New Menu Buttons

Display

Sequence On

Displays the single letter code for the AA sequences for each object.

Alignments

Included in PyMOL is an algorithm for structural alignments. We mentioned last time that you can use the Action → Align from the Object Menu panel to align multiple objects. You can also use the command line in the Upper Controls menu with:

align obj1, obj2

will align obj1 on top of obj2. However sometimes you need to specify the chain such as:

align (chain b and obj1), (chain d and obj2)

or

align obj1//b//CA, obj2//d//CA

To fit specific amino acid regions:

align (chain b and obj1 and resi 100:200), (chain d and obj2 and resi 300:400)

or

align obj1//b/100-200/CA, obj2//d/300-400/CA

<u>Exercise F:</u>

Lets reinitialize and fetch the following two structures: 1UWH and 1Z5M
The Reset button will place both structures in the screen. Are they similar?
What is the best way to get a sense with overlaying them?
Now overlay the structures. What are the RMS deviations? _____ (look at the left side of the Upper Control menu). Were you right? Do they completely overlay?
By default, when you overlay structures in PyMOL it always does chain A on chain A. Can you see if chain B from 1UWH overlays with 1Z5M?
Turn on the sequence menu—how do things look there?

Even More New Menu Buttons

File

Save Image As
> Allows you to save an image in a PNG format for use in other programs (i.e. Powerpoint).

Making Images

The most creative part of PyMOL is designing visually striking images that get your point across without being cluttered. This means the view has to be right, the colors meaningful, and the

By default PyMOL gives all of your images a transparent background (personally I find this to be GREAT—it easily inserts into any presentation). If you want to change this, you need to uncheck Opaque Background under **Display** → Background

The true beauty of your image will appear when you ray trace it. To accomplish that, click on the Ray button in the Upper Control panel (most upper right). It shows a progress bar just under the buttons while it works. If you click on anything before selecting **File** → Save Image you will drop out of the high-resolution mode.

For this exercise I want you to work with a protein (or two) of your interest and design some visually striking images that show off: 1) the overall structure; 2) place a surface on your structure; 3) highlight a unique feature (binding site, active site, DNA/RNA, sugar, metal, etc); and 4) find a homolog and indicate shared homology.

Please save a session for each image so that we might quickly return and try to improve upon each image.