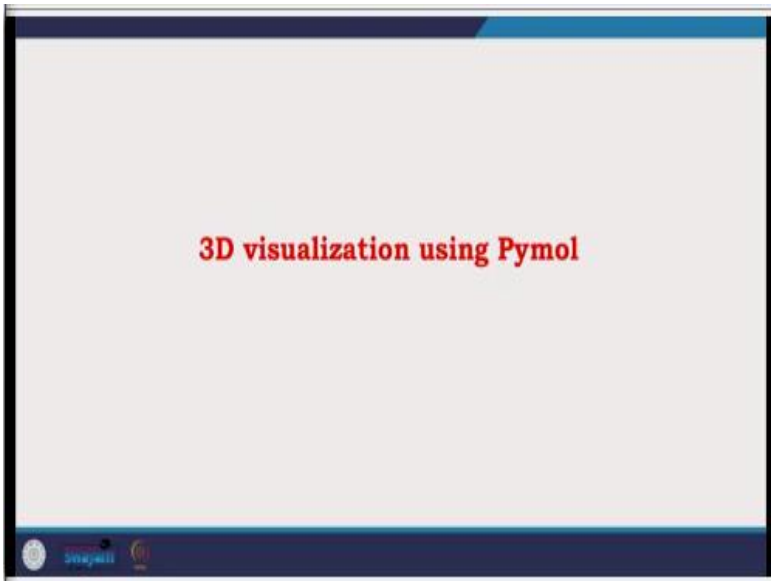
Structural Biology
Prof. Saugata Hazra
Department of Biotechnology
Indian Institute of Technology-Roorkee

Lecture-45
Demonstration of Pymol

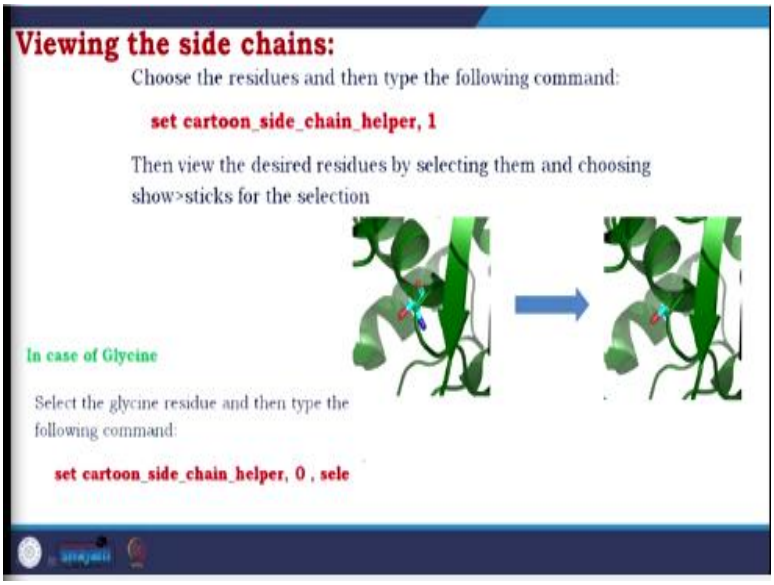
Hi everyone, welcome again to the course of structural biology. We are at the end of the visualization module, and I talked about visualization platforms in the last class. These platforms are used to examine the macromolecules, analyze them, and make publication quality figures. I continued with one of the example visualization platforms, which is Pymol. I have demonstrated the initial properties and what you could do, from uploading the pdb molecule to analyzing it. And today, I will continue with the part I have finished.

(Refer Slide Time: 01:18)



So, let us continue with the demonstration of the 3D visualization using Pymol. One of the critical parts of analyzing a macromolecule is to view the side chains. Why are side chains important? If you remember the protein structure, built up from amino acids, there are 20 different types of amino acids and 20 other side chains; the side chains give you the properties. So, we need to look at them for our analysis.

(Refer Slide Time: 01:56)



So, when choosing the residues, you must type the following command set cartoon underscore side underscore chain underscores helper, 1. Then you could view the desired residues by selecting them and choosing so rather than sticks for the selections. I have written the comments so that you can follow them here. So, you see, you could have looked at one side chain from here and then hidden the main chain for better viewing and only show the side chain.

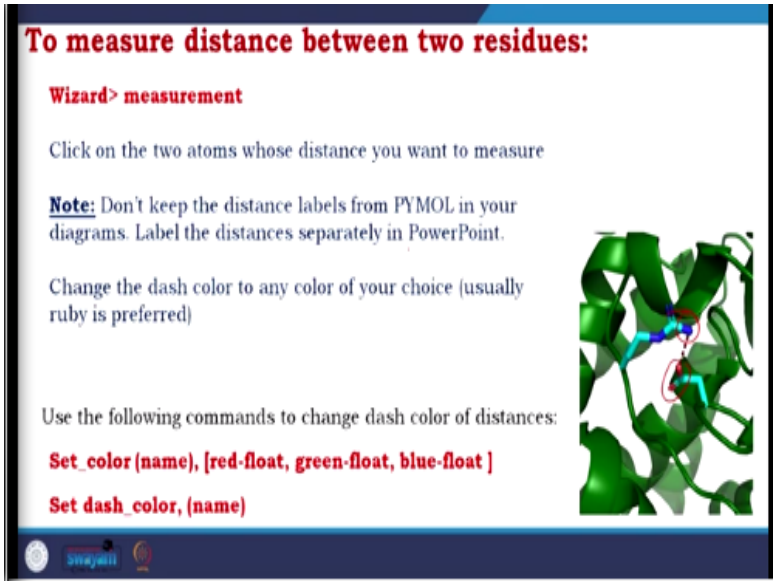
If you remember the side chain is H, you have to put the following command for glycine. **(Video Starts: 02:52)** I want to show you the demonstration. So, now you see the screen; whenever the screen comes and you start, you have to load the pdb file here. I have loaded the pdb file of the enzyme called beta-lactamase from mycobacterium tuberculosis, which I discussed in the **(())(03:11)** session. And now, you have to select the amino acid you have and want to show the side chain.

Here we have chosen the serine, which is serine 84; this is the catalytic residue of the enzyme. Through this enzyme, the enzyme performs its catalytic activity, and you see that the serine is represented as the whole protein; the entire enzyme is represented in cartoon format. Whereas we have given a different stick format to the amino acid, this will help you to identify the amino acid.

And next, this will also help you identify this catalytic amino acid's interaction with others. Now, if you remember, I talked about 20 amino acids; all of these amino acids have side chains, whereas glycine has only hydrogen that makes glycine acryl amino acid. And here, we will show you how the representation of glycine happened. You see here, glycine is selected, you will see the selection, and then again, the stick format is given here.

So, you could see the glycine, and now this glycine, if you see it, only has hydrogen, which is why it is a unique amino acid to mention. **(Video Ends: 04:51)**

(Refer Slide Time: 04:52)



So, returning from where we started, we talked about side chains. Now, we will get two amino acids and their side chains, and then it will be utilized to measure the distance between those two residues. So, in the picture, you will see two amino acids whose distance is first identified and then calculated. Distance will help you understand whether there is a non-covalent bond formation.

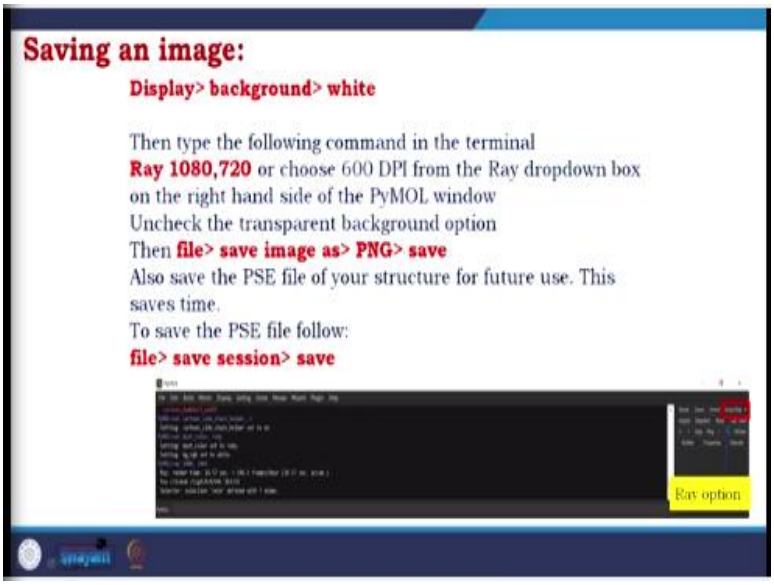
So, wizard to the measurement, what is happening? You must click on the two atoms whose distance you want to measure. Remember, do not keep the distance label from PYMOL in your diagram; label the distances separately in the PowerPoint; why? Because you measure it but do not label it, if you mark it in the PowerPoint, you would have control of changing them whenever you want.

Change the dash color to any color you choose; you see that dashed, so usually we present in ruby color; use the following command, set underscore color name, red gap float, green float, blue float, which is the color RGB code. So, with this, I will again switch to PYMOL, and I will show you. **(Video Starts: 06:35)** Again, this is the window; you have remembered that we have already selected the serine, and now what we are selecting is lysine.

So, when you are selecting a lysine, you know that you get the nitrogen, and we will check the distance between the serine and the lysine's oxygen and nitrogen. So, when you do that, you can see that, in this case, the distance is 3 Angstrom, which clearly shows that these two amino acids are in non-covalent interaction, which are hydrogen bonds. So, if you remember what I talked about, I am repeating you will not take the name or the colors given here; you go back and do it in the PowerPoint.

So, you could make it a ruby-colored generally default but do not put the distance. **(Video Ends: 07:58)** This is the interaction between the serine, the catalytic residue, and the lysine, the secondary catalytic residue, which helps in the catalysis by taking the proton from the serine, making the setting O minus.

(Refer Slide Time: 08:13)



Coming back, we will show how to save an image. So, you go to display, then to the background, and make it white. If you remember, I have already done it because, as I told you, you want to save an image on a white background whenever you want. So you do not spend unnecessary ink when you take a printout. Then you must type the following command in the terminal Ray 1080, 720 or choose 600 DPI from the Ray drop-down box on the right-hand side of the PyMOL window.

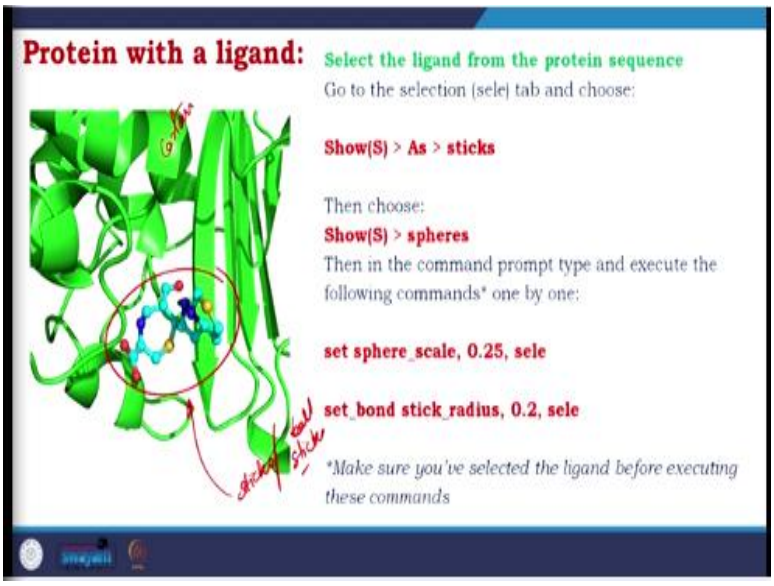
You have two options, and then uncheck the transparent background option. Then file, and save the image as PNG; if you want to save it in PNG, there are other options than save. Also, if you are careful, you would like to save the dot PSE file of your structure because if you do that, for any future reference regarding the interaction you want to make an image, you could change it, so that is always clever.

To save the dot PSE file, save session save; if you remember, I have shown you that option. And this is the Ray option if you remember again; in the pop Ray in the Raster3D, those are the technology that comes from them, and now it is integrated into any visualization platforms. So, with that, I would again switch and show you the condition we have in the PyMOL. **(Video Starts: 09:58)** As I mentioned, we want to save it.

So, if you see, now you change the parameters, the DPI values, and all so that you can make a high-resolution picture. See, the development of the formation of the rendering is happening. Rendering means if you carefully see the screen, the picture gets sharper at the edges, becoming better. See, now you get this portion, it is already selected the part you want to save, so you put any name on that, and you could save it, so you save it.

Also, as I told you, when you go to file and save the session, you put save session or save session as and then save it. So, your name and, as I told it would be saved in the format of dot PSE or dot PZE, these are the two formats that save your session. **(Video Ends: 11:26)** Returning, we talk about protein. Still, a protein or enzyme, the enzyme beta-lactamase, is there. We are talking about the interaction of protein or enzyme with its ligand, with which substrates are essential to study.

(Refer Slide Time: 11:55)



So, to do the study, you have to select the ligand from the protein sequence; you have to go to the selection, which is select tab and choose. So, as sticks, your protein would be represented in a cartoon only; if you see your protein is represented in a cartoon here and then your ligand would be presented as sticks, balls sticks are the options to represent the ligand molecule generally.

After that, you choose, show spheres, then in the command prompt type and execute the following comments individually, set sphere to underscore scale, 0.25, sele. So, what are you doing? You are optimizing the side of the stick and sphere; set underscores bond stick underscores radius, 0.2 sele. Ensure you have selected the ligand before executing this command because it will apply to every bond if you do not select it.

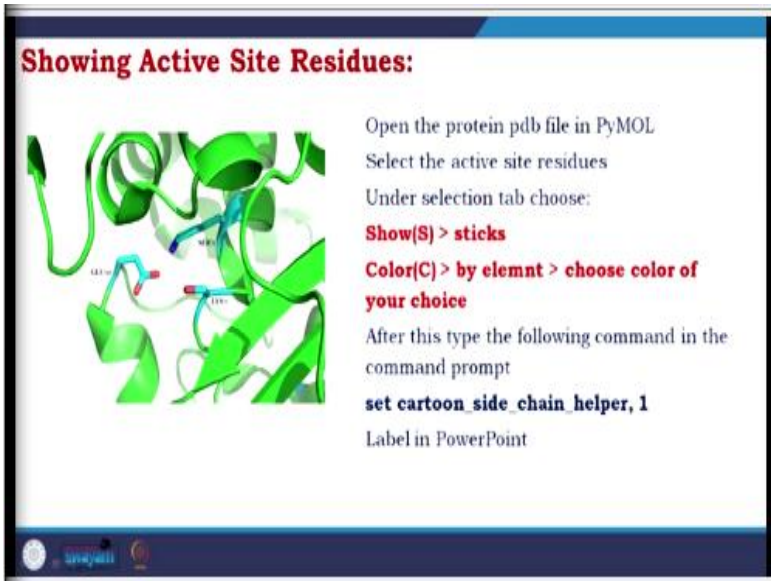
You could only differentiate between the ligand with others by selecting it and then selecting the parts. So, let us again go back and look at how it is possible. **(Video Starts: 13:31)** We delete the previous one and are loading a new structure. See, there are so many pdb files, and you choose a file, see here the chosen file is shown. If you see the guys in the red sphere or phosphates here, which come through the condition of that crystallization because this structure is solved by protein crystallography?

Now, you could remove water or keep them; you could remove the phosphate or keep them. So, all these things, whatever, here we remove all of them so that the structure becomes clean. And you see, the only thing attached to the protein is the ligand; this is again the same protein, the beta-lactamase from mycobacterium tuberculosis, and that drug is Tebipenem.

So, you could select the drug, as I told you, and you could add those commands to make the ligand easier to view. See, if you put everyone's sphere, it will look like that, so you have to be careful at each stage, and that is the only way you can develop a good figure. Also, if you look carefully, you can remember through code; I have already shown you the two Tebipanem confirmations, even in a structure with beta-lactamase.

Here you will see that the four-membered rings are present or absent, and the unavailability means it is making a covalent bond which is the normal case where the oxylation reaction has happened. Now, you see the representation of the ligand with the ball and the stick. **(Video Ends: 15:59)**

(Refer Slide Time: 16:02)



I am going back to my presentation, showing active site residues. So, although we have already shown the residues of serine and glycine, these are the active sites. So, every time what do you have to do? You have to open the protein pdb file in PyMOL and then select the active site residues; how do you choose them? It is a very interesting question; many beginners who come

and take training with me ask me. So, there is no rule; generally, we go to the literature and read about the protein, which gives us knowledge about the active site.

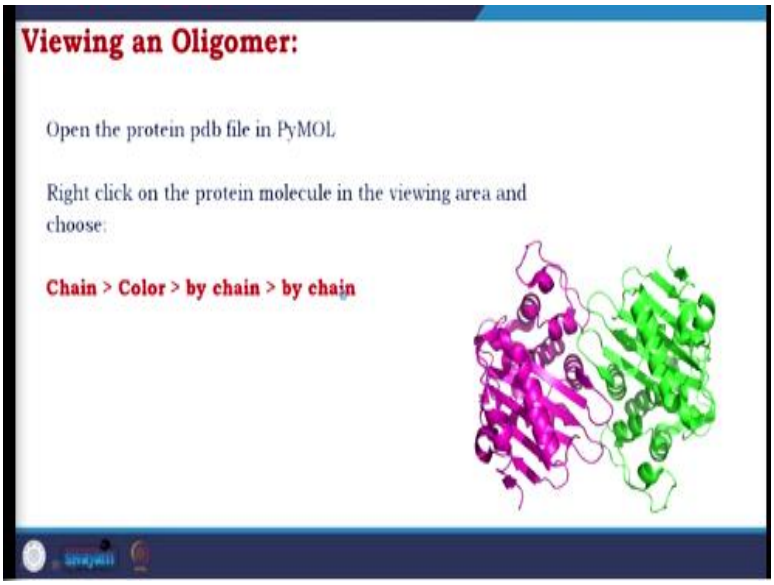
But if you are becoming an expert, then you can trace the substrate and the ligand, just now we have discussed that you could go back, you could trace, you could see the interaction, and you could identify who are the amino acids that are making the interactions with the substrate and they are the active site residues. So, under the selection tab, you have to choose, show the sticks, color by the element, and choose the color of your choice.

And then, type the following command in the command prompt, set cartoon underscore side underscore chain underscore helper, one, and then, as I always say, label that in PowerPoint. So, you will get things like that; you see serine 70, serine, lysine 73, and glutamate 166. When we go back, we will show you that residue but different numbering. If you remember, I told you these are the standard numbers. But each of the proteins and enzymes from every organism has its numbering.

So, let us go back and see the numbering here. **(Video Starts: 18:11)** Here, the serine is 84, and then we go to glutamate, which is 182, and then we will show lysine, which is 87. So, if you see, these are the three amino acids that perform the two reactions I talked about in the Coot class, which involve oxylation and deoxylation reactions. Here you see that lysine has taken the proton from the serine, and the serine attacks the beta-lactam ring and forms a covalent complex, which I have talked about.

So, now you know this is the active site and how those active site residues are closer to the substrate, you would see the periphery. **(Video Ends: 19:21)**

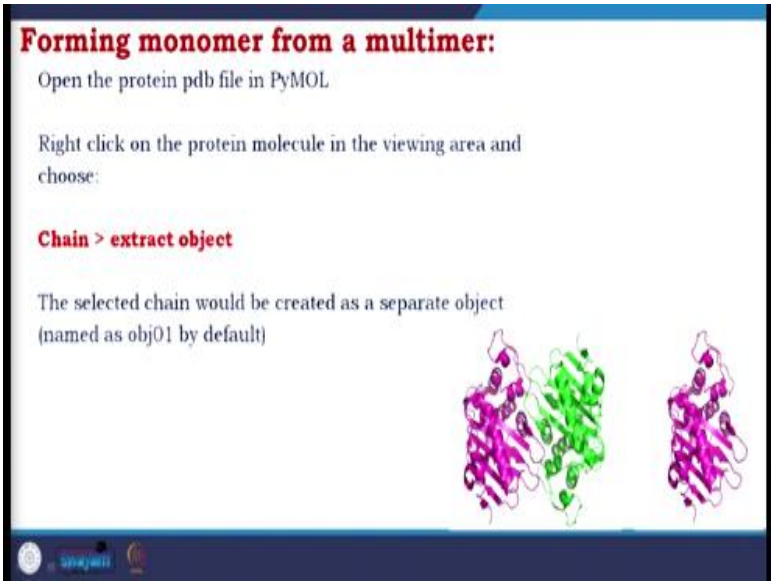
(Refer Slide Time: 19:22)



Let us go back to viewing an oligomer, I have already shown you a dimer, but here we will come up with a different type of representation. You see the oligomers, but here you will see some other oligomers to show their property. So, as usual, you have to open the protein in PyMOL, then you have to right-click on the protein molecule in the viewing area, and you have to choose chain color by the chain, then by the chain.

So when you do that, you will get this dimer; you see that pink and green color representing each monomer.

(Refer Slide Time: 20:04)

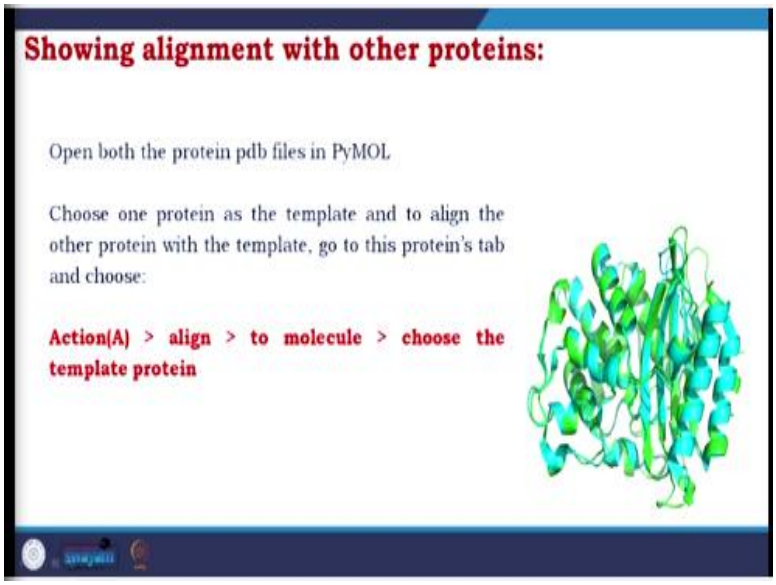


So, again before going into the monomer for a multimer, check it in the PyMOL. **(Video Starts: 20:16)** First, you must clean everything and load an oligomer. So, if you see there is a dimer, but you could only be confirmed when you go to color and color by the chain, two colors, so that means it is a dimer so that we will come again. So, it is clear to you now that this is a dimer. **(Video Ends: 20:53)**

So, again how are they forming? How could you select a monomer?
Again, you have to open the protein if it is not open; in our case, it is already available; then right-click on the protein molecule in the viewing area, choose the chain, and extract the object. The selected chain would be created as a separate object, which you could name as obj01 by default but have your name.

So, there is the dimer, and it would be a monomer which would be a result; remember I told you, you should always make a monomer before doing the alignment. So, let us see how to make the monomer here. The good part is now the commands are written here. **(Video Starts: 21:45)** So, you could easily follow them. So, you extract objects; see, I have shown you differently, now this is a different way to make it. So, you have a monomer now, so this is how you always convert the oligomers to monomers. **(Video Ends: 22:06)**

(Refer Slide Time: 22:07)

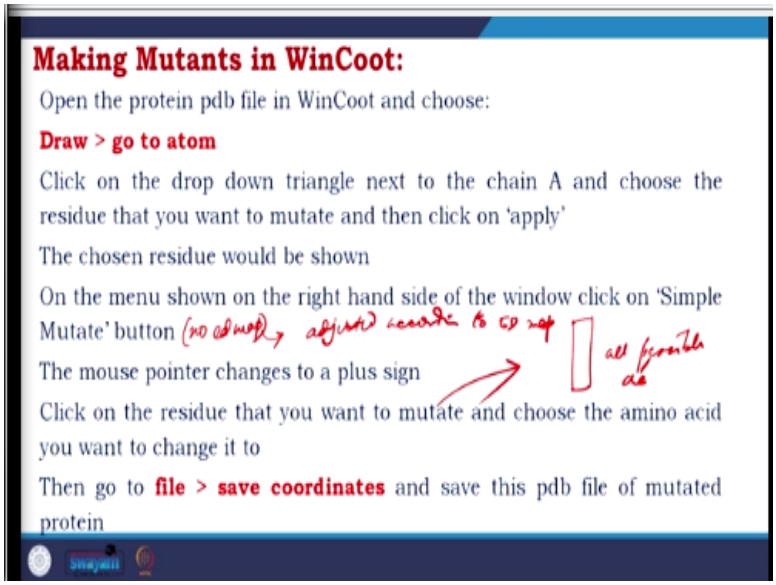


Now what to do with the monomers? You could do an alignment with other proteins and the two monomers. To do that, again, you have to open the two proteins; if one protein is loaded, you must open the second one. So, you have to choose one protein as the template, and to align the other protein with the template, go to this proteins tab and choose the action, which is the A.

Suppose you remember the five things, action, show, height, label, and color. So, A, then you go to align, then to molecule, choose that template protein. So, this is an example of alignment, and you see that these two proteins are looking at the alignment, you could see that these two proteins are the same or have identical folds with very high identity. **(Video Starts: 23:09)** So, let us switch and see in the PyMOL window how we could see the alignment.

So, first, as usual, you have to open the pdb file; one pdb is there, then you have to open another file. So, the second protein is there, and then you have to do the alignment. So, as you remember, you have done the alignment, and it shows that the RMSD value is 0.727 with 177 atoms, which means the alignment, is quite good. **(Video Ends: 24:01)**

(Refer Slide Time: 24:02)



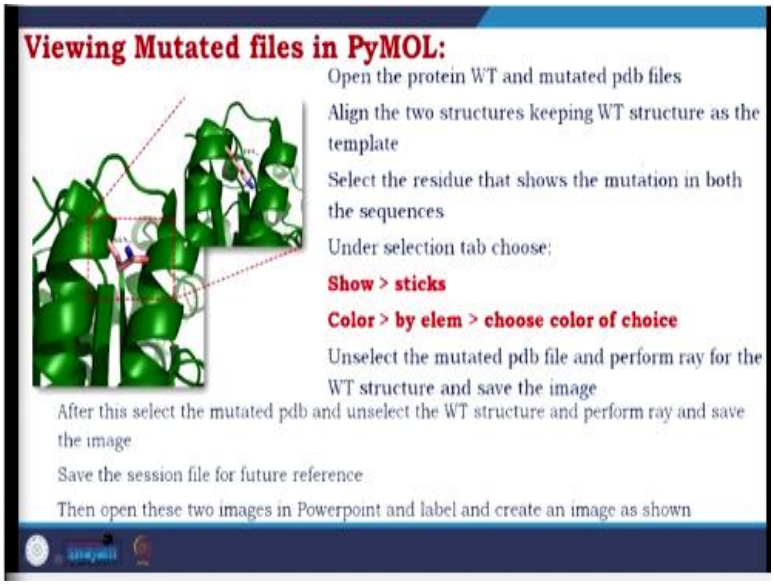
So, now making mutants. You could make mutants in PyMOL, but as I told you, for modification, we believe we trust more on the other type of softwares, so we do it in WinCoot. So, what to do in WinCoot? You open the protein pdb file; I hope you remember, I have given the demonstration, you go back, and you will see you draw; you go to draw and then go to atom.

Click on the drop-down triangle next to chain A, choose the receipt you want to mutate, and then click apply.

The chosen residue will be shown on the window's menu on the right-hand side. Click the simple mutate button if you do not have the electron density map. So, simple mutate, no ED map. But when you have an ed map, the mutation would be adjusted according to the ED map, so there are two options. The mouse pointer changes to a plus sign; click on the residue you want to mutate and choose the amino acid.

So, when you do that, if you remember, you will get a list of all possible amino acids. Now you choose the amino acid you want to change it to, go then to file, save coordinate, and save this pdb file of mutated protein, so you have done mutation to your protein.

(Refer Slide Time: 25:55)



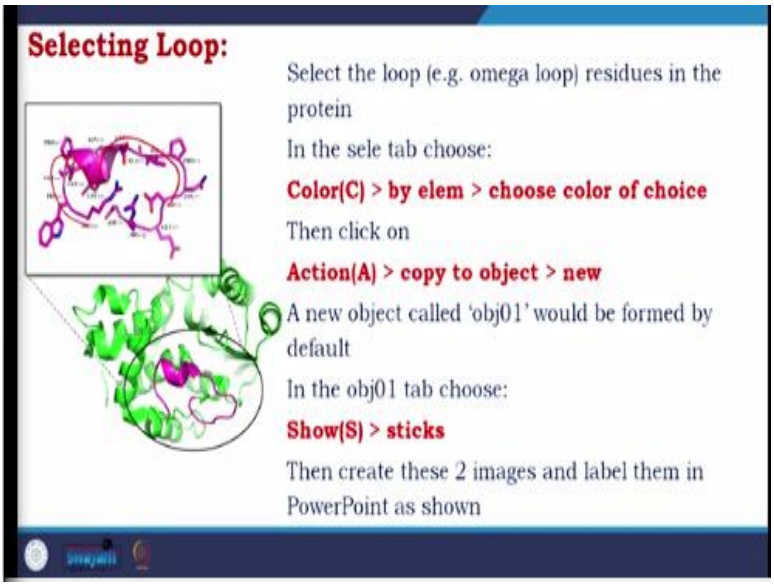
Now you want to view the mutated file in PyMOL. Again, you open the wild protein type and mutated Pdb files; you align the two structures keeping the wild type structure as the template. You select the residue that shows the mutations in both the sequences under the section tab choose, show, sticks. So, you make them in sticks color by elem, select the color of choice. Unselect the mutated pdb file and perform ray for the wild-type structure and save the image.

So, here you see the residue was glutamine 39, and you mutate it to lysine to make it lysine 39. So, that is something you have mutated already in Coot, and now you see it in PyMOL and save it in higher resolution for publication. **(Video Starts: 27:02)** To demonstrate that we again come back to the screen, and now we will see the mutants. We will also compare the wild type and the mutant to have a similar picture.

And as I said, we could have made the picture in Coot; why did we not do it? Because when we do that in Coot, we cannot get a nice, high-resolution picture. So, here we are making a mutation like finding a mutation, the serine 84, the catalytic residue is mutated to alanine. So, on one side, there is serine; on the other side, there is alanine, you see the difference in the same protein, but serine in place of serine alanine is observed.

Now you have to select them, you have to make the stick representation, and now you will see the difference. So, in one place, you have the serine; in another, you have the alanine; you could easily see the absence of oxygen. CH 2 OH is serine, CH 3 is alanine, coming back to. So, again here is what I forgot to tell you would save the session for future reference, and you now take them to the PowerPoint and make the figure according to how you like to have them. **(Video Ends: 28:43)**

(Refer Slide Time: 28:44)

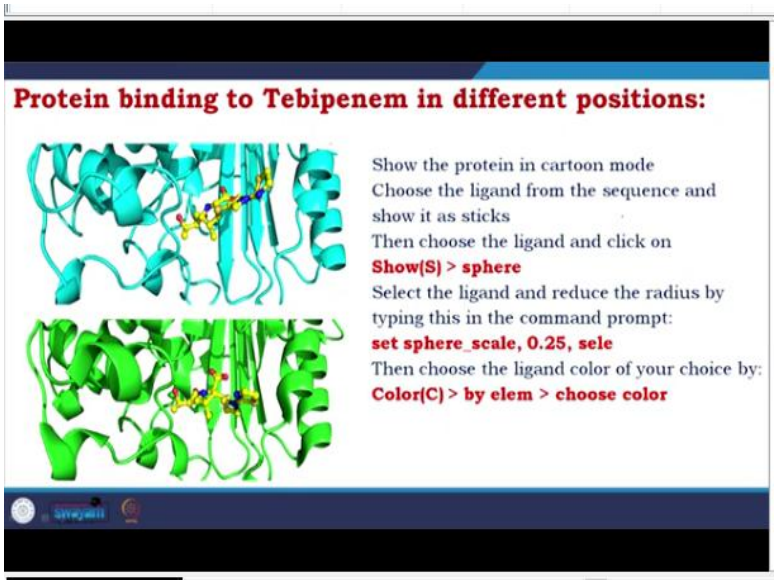


The selection of loop, loops plays a very critical role in function**.** Here you see a big loop, and if you see kind of carefully, you will see there is an omega type of representation. So, it is called the omega loop; this loop is characteristic in a spatial class A beta-lactamase, and another course of beta-lactamase is also found. So, what do you have to do? You have to select the loop residues in the protein. In the sele tab, you have to choose the color by elem, choose the color of choice, then click on the action, copy to object, new.

A new object, obj01, would be formed by default, or you could have given your name. In the obj01 tab, choose show sticks. Then you would have created these two images and labeled them in PowerPoint. **(Video Starts: 30:02)** Again, go back and look at how we are taking the omega loop as I was talking about and selecting it. So, we have to keep it in the cartoon, and then you have to select the omega loop; you have to know the numbering representing the loop.

And then, you have to work on selecting the loop, and by doing that, you have to know where the loop residues are and then color it again by element. So, you will see that in a nice omega loop, you have to represent them by stick by making a separate object. As I told when you do that, you could save it as object 01. This is the loop, and the molecular representation will describe the composition of the amino acids in the loop. Also, you see a 310 helix, part of the omega loop. **(Video Ends: 31:18)**

(Refer Slide Time: 31:20)



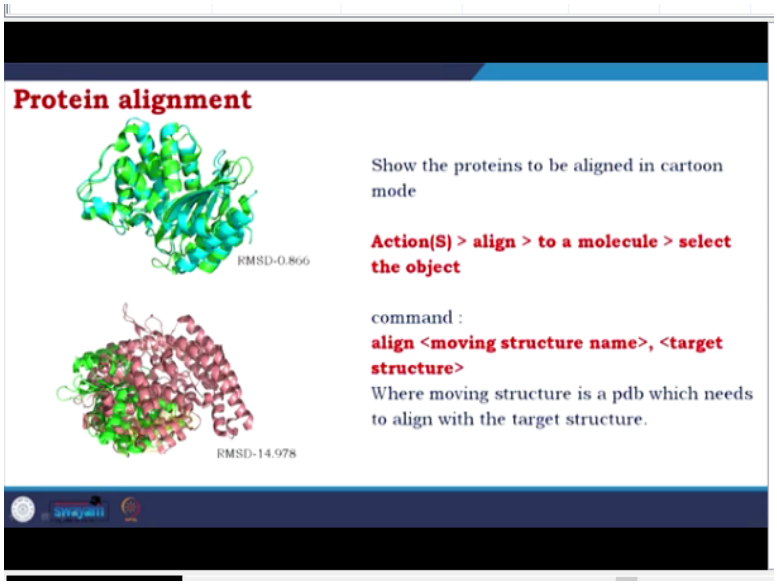
Again, I am switching. So, if you remember, I have shown you the different cell binding of the Tebipenem in 2 structures. This is because, in one, there is a mutation; as I have shown you earlier, the mutation is lysine to alanine, and because of this mutation, the lysine could not have taken the proton from the serine, and the serine could not react. So, two blessy structures we have are with Tebipenem, but in one that Tebipenem could not react where the mutation of lysine to alanine happened and one where reaction the first step the oxylation happened.

So, that is the scenario to do that; first, we have to show the protein in cartoon mode, then choose the ligand from the sequence, and so it sticks, then choose the ligand and click on such a sphere because remember, we want to show the Tebipenem ball and stick mode, select the ligand and reduce the radius by typing these in the common prompt set sphere underscore scale 0.25 sele you are fixing the sphere then choose the ligand color, color whatever you want, I want to show you that. **(Video Starts: 32:51)**

So, I switch here again. Again, you have to delete the object and load both the Tebipenem complexes, teb mm, and teb ca; teb ca is already loaded. So, another one is teb mm. So, the two blac structures are the same as the two states of the Carbapenem drug, Tebipenem. So, we have made them here; if we align them, it would be easier to take a look. We could also remove the other parts of the water and phosphates and then do the alignment.

So, when you see the alignment, you see how good it is now, and we could probably make them and bring them to the center. So that you could see the 2 Tebipenems, remember when we saw these Tebipenems in Coot, we have already known that they are differentially bound, but now bringing them in PyMOL, the pictures would be much better as you see, you could have focused it, orient it and represent a much better structure than what you have seen in Coot. So, here you see the 2 Tebipenems; I am switching. **(Video Ends: 34:22)**

(Refer Slide Time: 34:23)



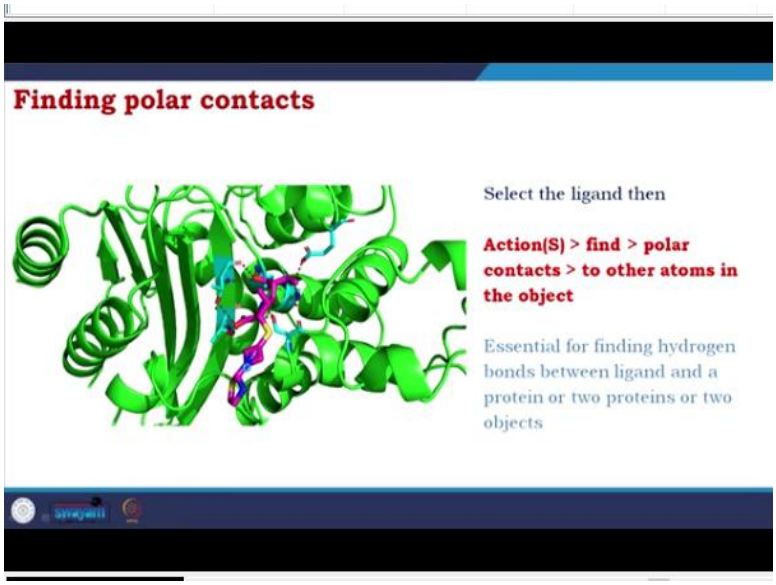
Protein alignment, I have shown you the alignment; if the proteins have suitable for matching, they have a low RMSD value. If the matching is not excellent, then the RMSD value would be high. As you see here, RMSD is 0.866 when the two proteins have nearly identical fold and sequence, whereas it is not RMSD 14.978. So, as usual, you must first show the proteins aligned in cartoon mode.

Then action, aligned to a molecule, select the object, then you have to put the common align moving structure name target structure. So, align first the structure that must be moving and then the steady structure. So, first, the moving structure, where the moving structure is a pdb, which needs to align with the target structure. **(Video Starts: 35:35)** So, I will go to the PyMOL window and show you the alignment.

So, you have to load the pdb file as I am doing here, and then you need two pdb files because you have to do the alignment. So, there are two pdb files. And when you are doing the alignment between them, we will see that aligned to molecule, you will see good alignment, and the RMSD value is 0.727 means they are perfect as too close structure. And now, we will see another alignment where we are trying to align two different structures.

And when you do that, you see the RMSD is 14.337, which swing that the two structures have significantly less fold similarity. So, understanding these comparing structures is a very utility tool. **(Video Ends: 37:03)**

(Refer Slide Time: 37:04)



Now coming to finding polar contracts, as I told when you want to understand the deeper label of science, you have to find out the interactions; inside the protein, what are the interaction with the protein with the ligand, and all these things and that is called finding polar contacts. First, you must select the ligand with whom you want to see the interactions, then go to action and find polar contacts to another atom in the object.

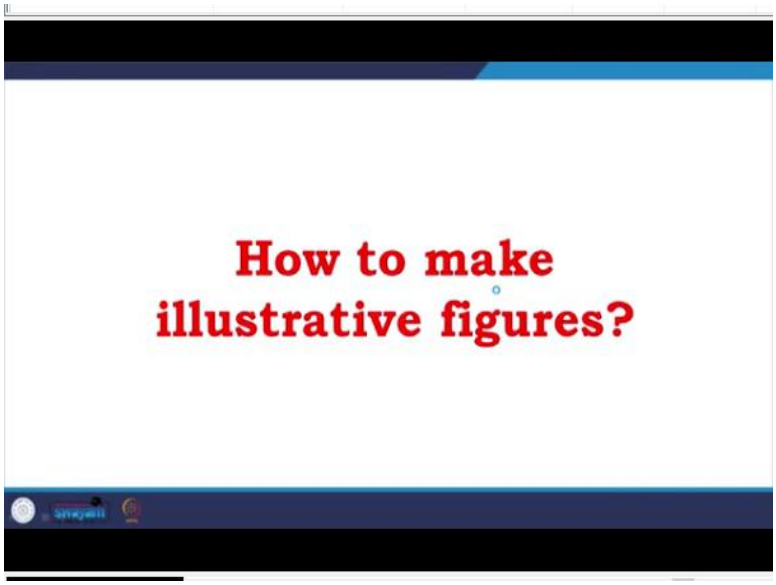
And it is essential for finding hydrogen bonds between a ligand and a protein or two proteins between 2 proteins or between 2 objects. So, let us see the PyMOL demonstration of finding polar contracts. **(Video Starts: 37:52)** You must load the pdb file. And once you are loading the pdb files, what you have to do you get the polar contacts. So, if you see here, you have identified the polar contacts with Tebipenem; Tebipenem is a Carbapenem, as I told you. So, here you go, and you find polar contact with the other atoms in the object.

Here, the yellow lines indicate the possible hydrogen bonds that could form between the protein and the ligand. The water molecules present in the protein means that are catalytically important. So, now, you do them, color them, come back to PowerPoint, and label them. **(Video Ends:**

39:10) So, we have shown in these two classes how to get many different things and do that through PyMOL.

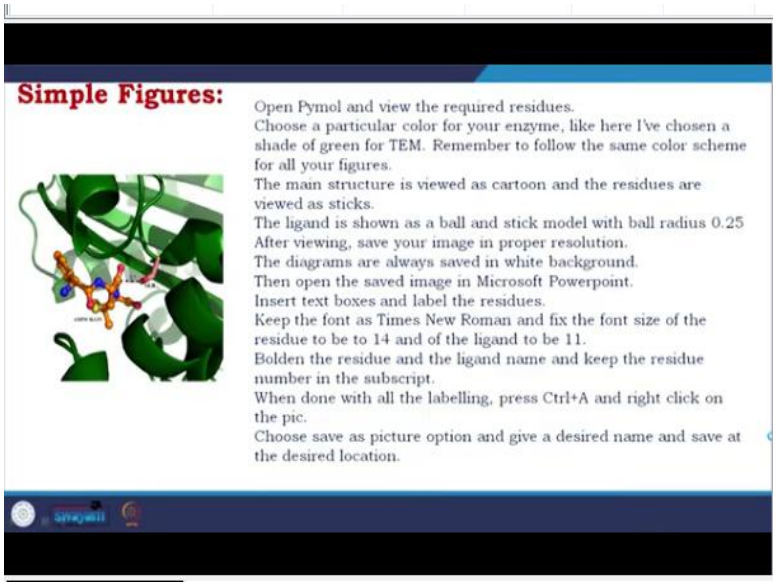
But one of the highly critical things through PyMOL is to make good figures, probably in these two classes. I have told you about these multiple times. So, now this is time we are going and showing you how you could make publication quality figures using this software.

(Refer Slide Time: 39:42)



So what is the software used? You would require the following software PyMOL which is the major software; WinCoot, which will help you to guide, and Microsoft PowerPoint, which will help you get more control over the structure. If you use the labeling coming through the software, you lose control, you probably want to show some interaction, or you do not want to show you will not get those facilities. You will not get those advantages if you allow the PyMOL software by itself to label.

(Refer Slide Time: 40:23)

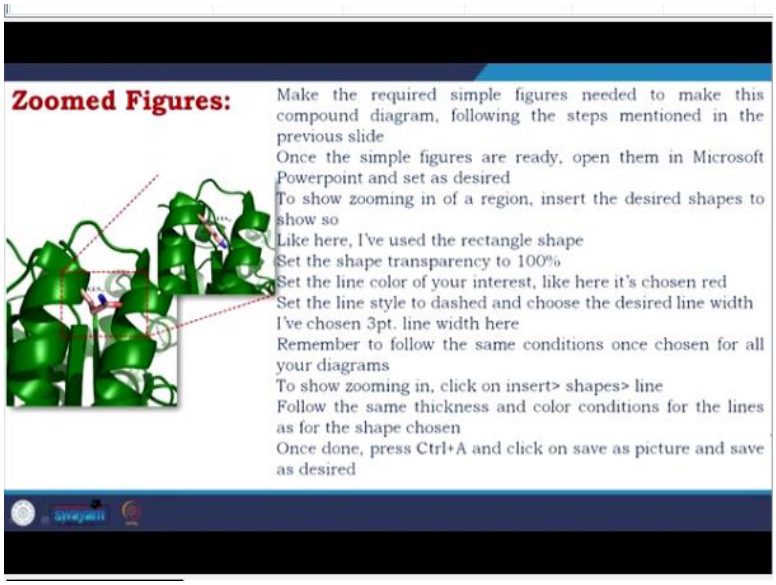


First of all, you could make a simple figure. To make a simple figure, you open PyMOL and view the required residues; you choose a particular color for your enzyme, and like here, we have chosen the shade of green for the enzyme tm, and you remember to follow the same color scheme for all of your fingers. So that you make a continuity like your two proteins and you make it through 2 different colors, it is helpful to make the signs visible.

The main structure is viewed as a cartoon, and the residuals are viewed as sticks. The ligand is shown as a ball and stick model with a ball radius of 0.25. After viewing, save your image in proper resolution; the diagrams are always saved in the white background, as I have talked about, then open the saved image in Microsoft PowerPoint, insert text boxes and, label the residues, keep the font as time New Roman and fix the font size of the residue to be to 14 and of the ligand to be of 11.

Bolden the residue and the ligand name and keep the residue number in the subscript; when done with all the labeling, press control A and right-click on the pic, choose the save as picture option and give a desired name and save it at a desired location. So, this is one of the high-resolution pictures developed you could see. So, you could also follow this theme and make it a simple figure.

(Refer Slide Time: 42:08)



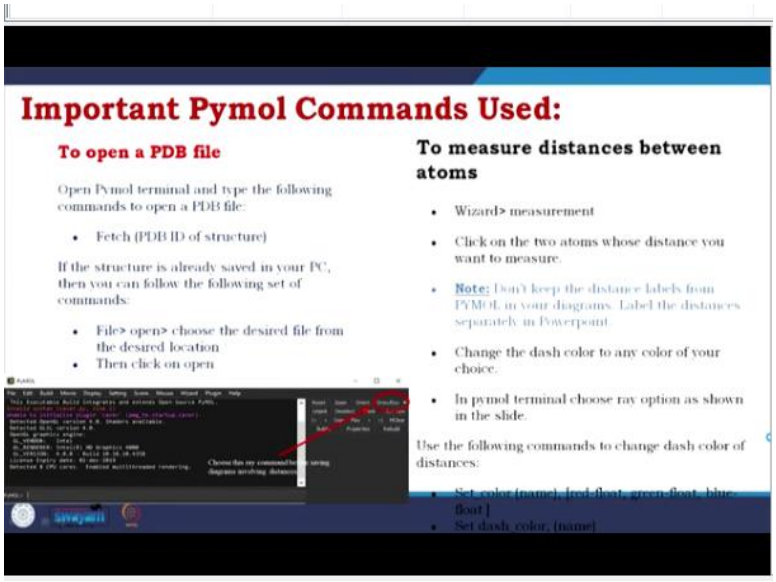
Zoom the figures. So, you have one mutation for one residue. You make a mutation module to show the zooming of it. So, make the required sample figures to make this compound background following the steps mentioned in the previous slides. So, following that, you could make a simple figure. Once the; simple figures are ready, you open them in Microsoft PowerPoint and set them as desired. So, here you are, not creating an image to mix a few images to make a detailed image that explains the science you want to project.

To show zooming in on a region, insert the desired steps to show that. Then here you see you have used rectangle shapes; you could use rectangle shapes, you could use any other shapes, set the shape transparency to 100%, set the line color of your interest like here we have chosen red, set the lines style to dashed and choose the desired line width. So, these are all not needed scientifically, but more carefully doing that, you will make more attractive publication-quality figures.

Set the lines style to dashed and choose the desired line width. We have chosen 3 points and line width here, and you always remember to follow the same condition once chosen for all your diagrams. Continuity again shows the author's maturity in writing the manuscript and creating the figures or describing a project as an undergrad; as a master, you do projects when presenting the project results.

If you make continuity in figure making that shows how good and mature you are, click on insert, shapes, and line to show zooming in. Follow the same thickness and color conditions for the lines as for the shape chosen. Once done, press control A and click on save as a picture and save as desired in the desired location.

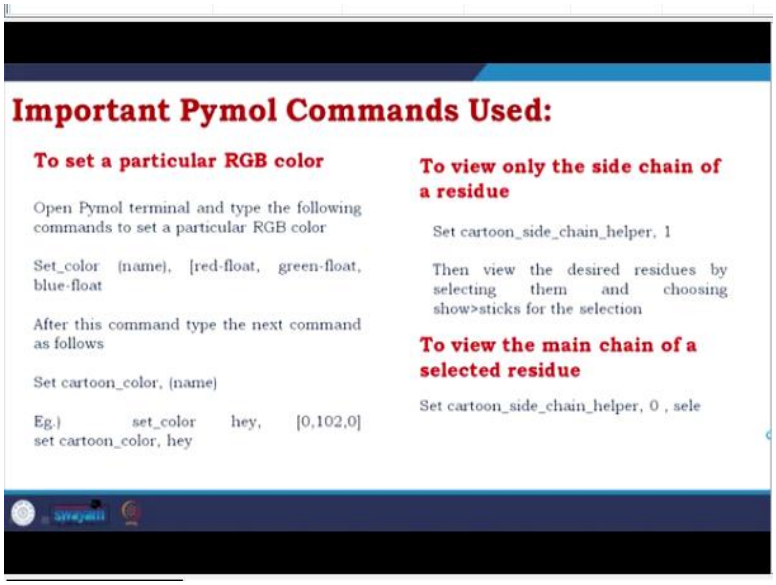
(Refer Slide Time: 44:35)



Important Pymol commands used I have made a list to open a PDB file. I have shown you to open the Pymol terminal and type the following commands Fetch PDB ID the common line. Suppose the structure is already saved on your computer. In that case, you can follow the following file, open it, choose the desired file from the desired location, and then click open to measure the distance between atoms wizard measurement.

Again we have shown that you click on the two atoms whose distance you want to measure. Please do not keep the distance label from the Pymol, as I have talked about earlier. Label the distance separately in PowerPoint, change the color, and then in the Pymol terminal, choose the Ray option and use the following command to set the color and dash options.

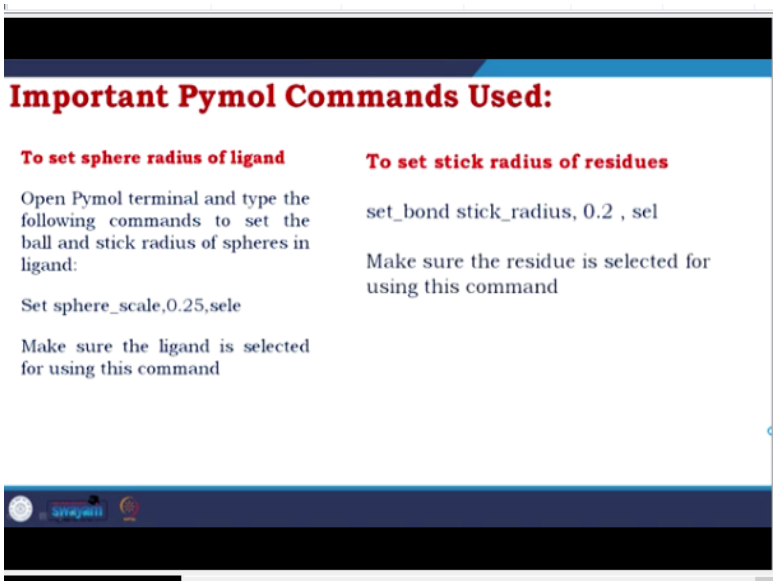
(Refer Slide Time: 45:28)



To set a particular RGB color, open the Pymol terminal and type the following commands to set a particular RGB color. Set underscore color, name, and float red, green, and blue RGB. After this command, type the next command as follows. Set the cartoon to underscore color and provide the name. For example, as we have shown earlier, you use hey color, which is 0, 102, 0. To view only the side chain of a residue set cartoon underscore, side underscore, chain underscore, helper, 1.

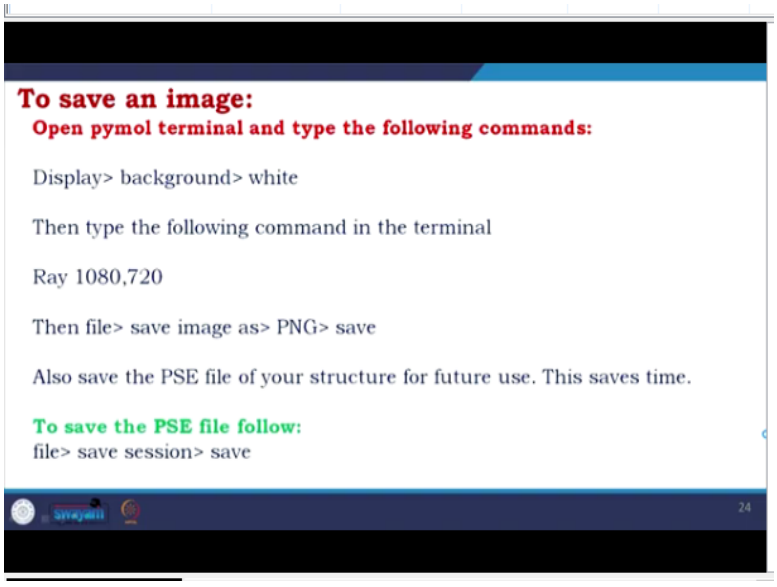
So, I have listed them, and though I have demonstrated them, I have listed them again so that you do not make a mistake and all these are correctly organized. Then view the desired residues by selecting them and choosing show sticks for the selection. To view the main chain of a selected residue set cartoon underscore side, underscore, chain underscore helper, 0, sele.

(Refer Slide Time: 46:29)



To set the sphere radius of the ligand, open the Pymol terminal and type the following command to set the ball and stick radius of the sphere in ligand, set sphere underscore, scale 0.25 sele, and ensure the ligand is selected for using this command. To set the stick radius of residues, set underscore bond stick underscore radius 0.2 sele. Make sure the residue is selected for using this command.

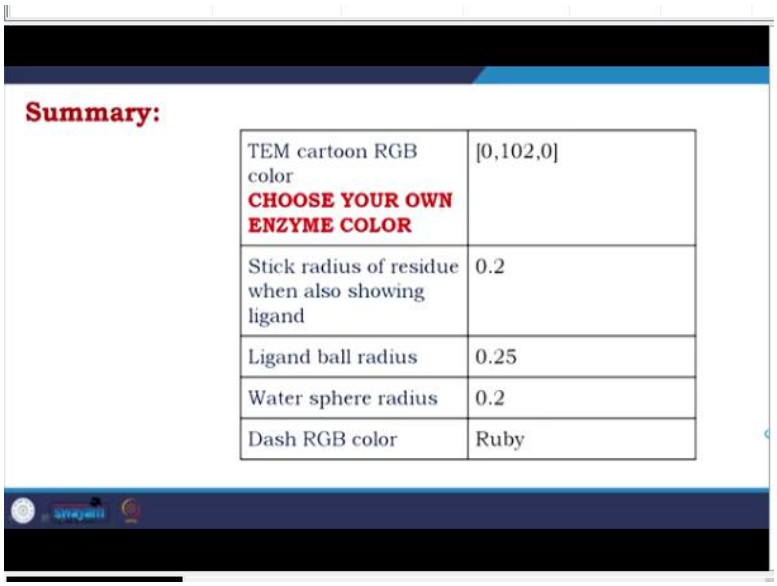
(Refer Slide Time: 46:58)



Lastly, open the Pymol terminal to save an image and type the following command. Display background white; you remember I repeatedly told you to make your terminal white, then type the following command in the terminal Ray 1080,720. Then file save images PNG save. Also,

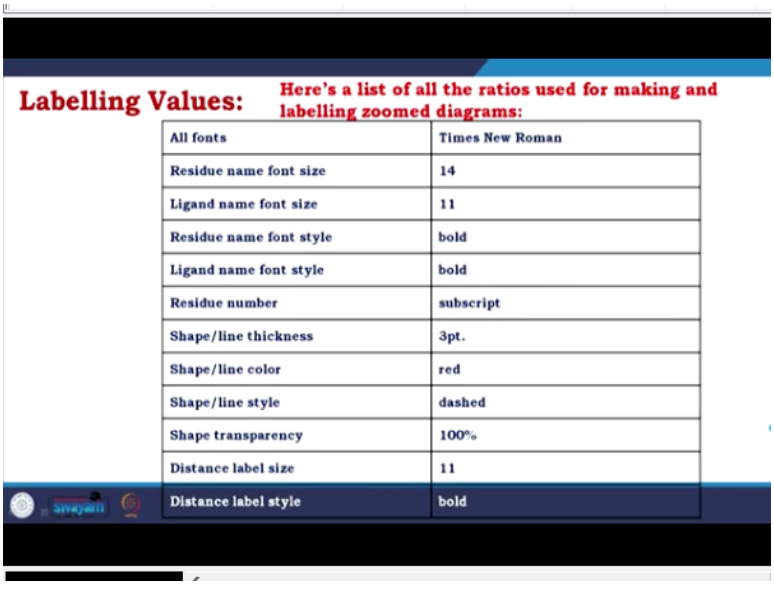
save the dot PSE file as I again talk about this; that dot PSE file helps you to bring the same thing again. To save it, file save session save.

(Refer Slide Time: 47:35)



So, the summary, if you have TEM, which is a beta-lactamase we have shown here TEM cartoon in RGB color, you have to use 0,102,0. You could choose your enzyme colors, stick radius of residue when also showing ligand 0.2, Ligand ball radius 0.25, water sphere radius 0.2, dash RGB color ruby.

(Refer Slide Time: 48:06)



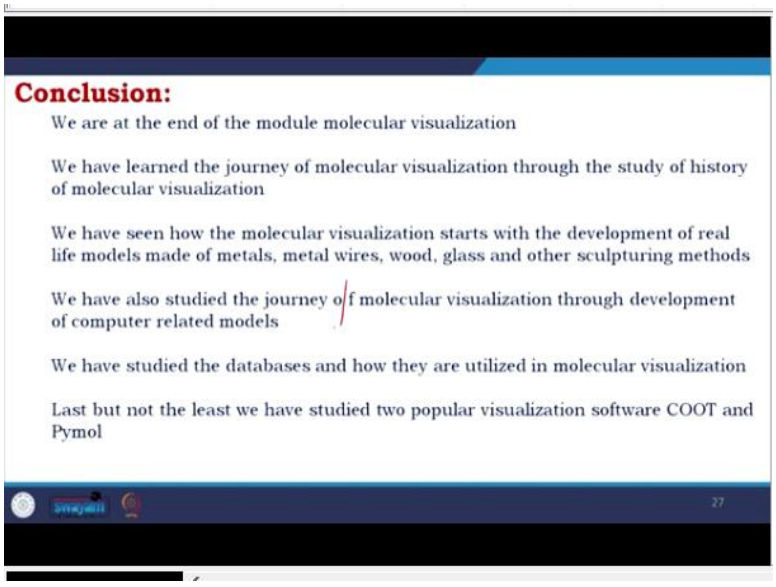
Labeling values is a list of all the ratios used for making and labeling zoomed diagrams, all fonts Times New Roman: residue name font size 14, ligand name font size 11. Residue name font

bold, Ligand name font bold, Residue number, subscripts, shape, line, thickness 3 points. Shape, line, the color red, shape, line, style dashed. Shape, transparency 100%, distance label size 11, distant label style bold.

So, is it mandatory to do that? No, there is nothing to be fixed, but we have given you those numbers; if you apply, you will be very surprised and very elect to see that you have made a beautiful figure publication, a quality figure out of Pymol. Once you do that, you make all changes according to how you like, you might not like, you know, the font of Times New Roman and all the things you could do, but the ratios are important.

So, with this, I hope you can learn more as I told you at the time of Coot, I am repeating this Pymol, Coot, and all the related softwares like VMD and Chimera. They are coming up with thousands of choices. How to know them? To play, I have given all the links, download this software, got the pdb file from protein databank, and did anything and everything; the more you play, the more you spend time and learn. So, with that conclusion, this module.

(Refer Slide Time: 50:03)



We are at the end of the module molecular visualization; we have learned about molecular visualization's journey to study molecular visualization's history. We have seen how molecular visualization starts with developing a real-life model made of metals, metal wires, wood, glass, and other sculpturing methods. You probably remember it is a very significant story of Byron

Bends. If you do not remember, Byron Bend is a wire made through machines that could be a low-thickness bent wire.

And two structures, one of an antibody and another of disulfide mutants; the model for both of them was created. Byron Bends created the physical model, and when both of the structures arrived at the meeting we saw, we read about the experience of Dr. Richardson, who worked on them. To his surprise, he finds them with identical folds; this is the first time people have seen two sequences with 9% identity providing identical folds.

So, these sculpturing objects not only help people to see the models but also help develop new science. Then we changed from the journey of sculpturing to computer-related models. We have studied the databases like pdb, pdb E, pdb J, the NMR database, the electron microscope database, and the nucleoside database. All are marked together to global pdb now, and what type of files do they have? The PDB file, the MMC file, the MTZ file, the XML file, and the FASTA file.

All types of important files have been studied, and then we have studied different softwares. We majorly divide them into two parts, one visualization, software with modification, and another only visualization platform to make good quality figure in movies. We have chosen each representative and one from them. The modification software Coot is the representative, and Pymol is the representative of the visualization.

And with that, we learned how to study that software and know and use it, and we end up making an example of how to make a high-quality, publication-quality figure. I hope all these things will help my young generation of structural biologists to be excited to learn more through visualization, as I always believe seeing is believing. So, you will see more I expect you will be more excited, and with this, I will have made the new generation more excited about the 3D structure of biological macromolecules.

Thank you very much; you guys are wonderful at listening; as I always say, keep listening and asking us questions if you have doubts. Thank you very much.