Bioinformatics
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Lecture – 60
Demonstration on AutoDock

Demonstration on Autodock; Autodock is used to get the interaction between protein and ligand mainly, and in this demonstration we will cover two aspects. One is rigid docking and the another is virtual screening in structure based web design.

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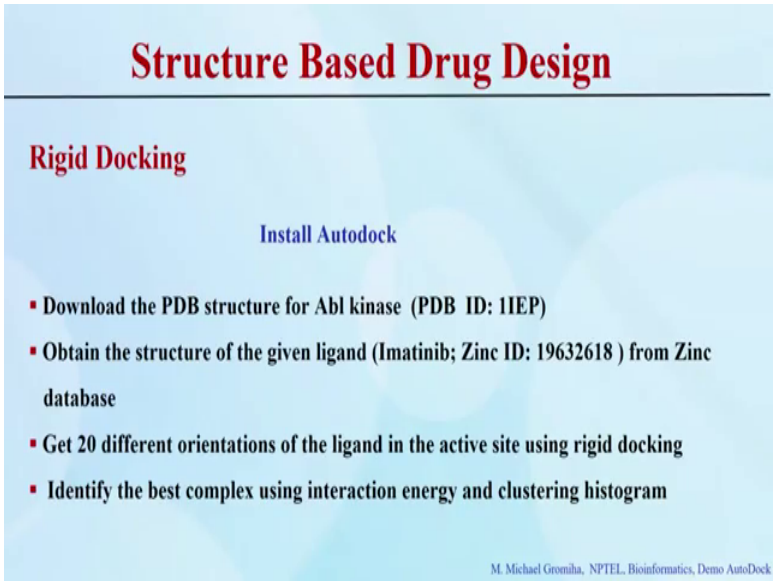

In the rigid docking first you install autodock you can go to the autodock website and you can follow the procedures to install autodock. And here we require a protein as well as ligand in the example we use the protein Abl kinase and the ligand Imatinib. We will get the PDB structure of this Abl kinase from the protein data bank the id is 1IEP and the ligand information from any of the small small databases like zinc database or the pubcam and so on.

So, then we get different orientations for this ligand and you try to get the best complex between the protein Abl kinase and the ligand using interaction energy as well as the clustering histogram.

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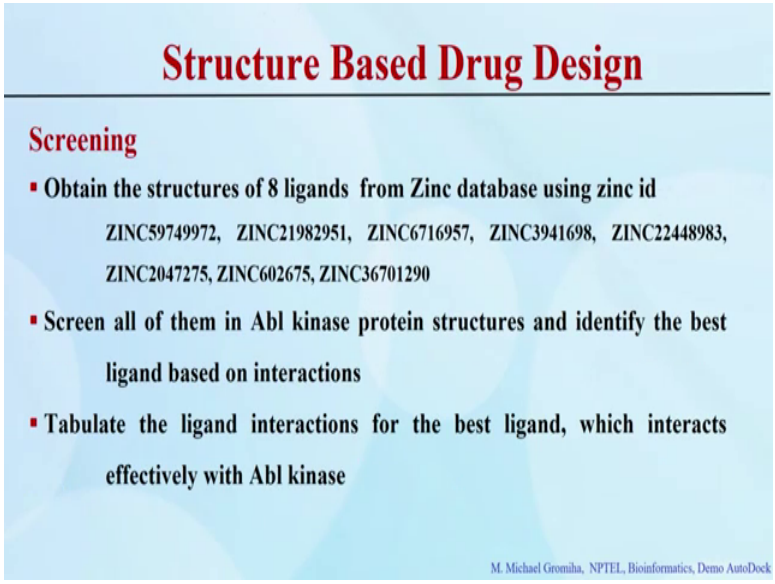

In the case of screening because several ligands. So, we need to identify which ligand is the best to interact with this particular target here the target is Abl kinase. So, we have several ligands you can obtain all these ligands from zinc database, and your screen all these ligands with the target that is abs Abl kinase protein, then I am find the best ligand based on the interactions right. And then we tabulate all the interactions and based on the interactions you can identify the best one, which is interact effectively with these Abl kinase.

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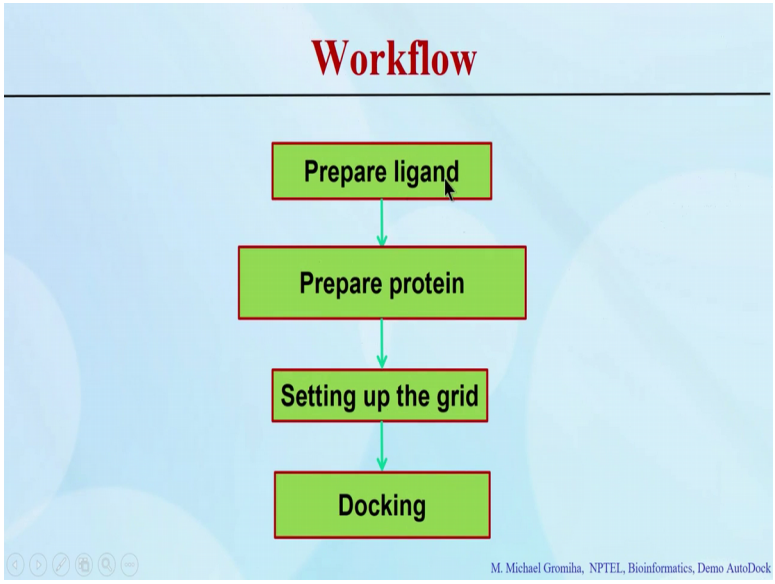

The workflow includes first a preparation of the ligand and prepare the protein, I discuss the details similar classes and we set up the grid for the interactions and finally, we introduce the docking and you get the score between protein as well as the ligand.

Now, we watch the detailed procedure right to get the complex as well as how to get the score, and how to identify the best ligand among the pour up different ligands obtain from the sink database.

In this session we will see how to do docking using autodock, in order to get the protein structure here we are going to use PDB database or interest of the protein is Abl kinase.

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So, go to PDB database and then give the PDB id 1IEP.

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And download the PDB format. So, if you install primal then click 1IEP structure.

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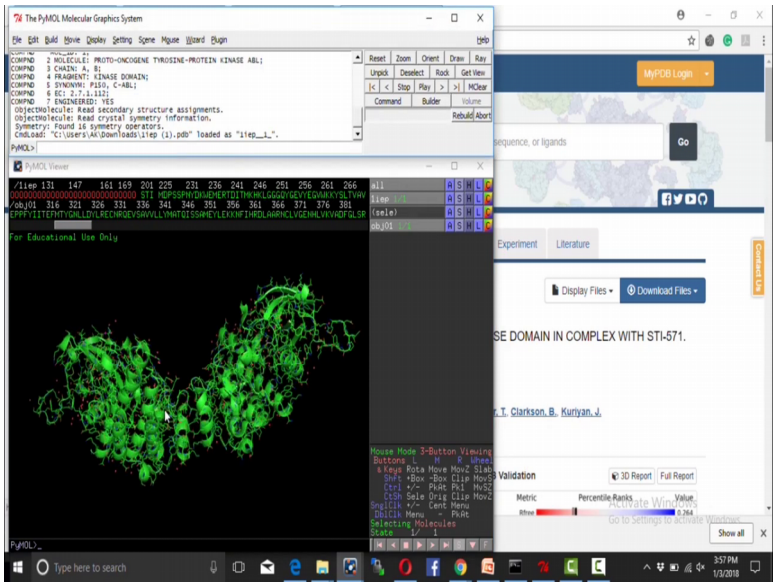

So, this particular protein structure has two different chains a and b chains as you see here, in our docking experiment we are going to consider any of the one of the chain for the docking purpose. So, in this we have to select molecule.

So, select any one chain. So, that will be like sele. So, save them as an a separate object copy to object. So, that will be obaj 02. Now you can save this file save molecule obj o2. So, now, you can save them kinase in a PDB format.

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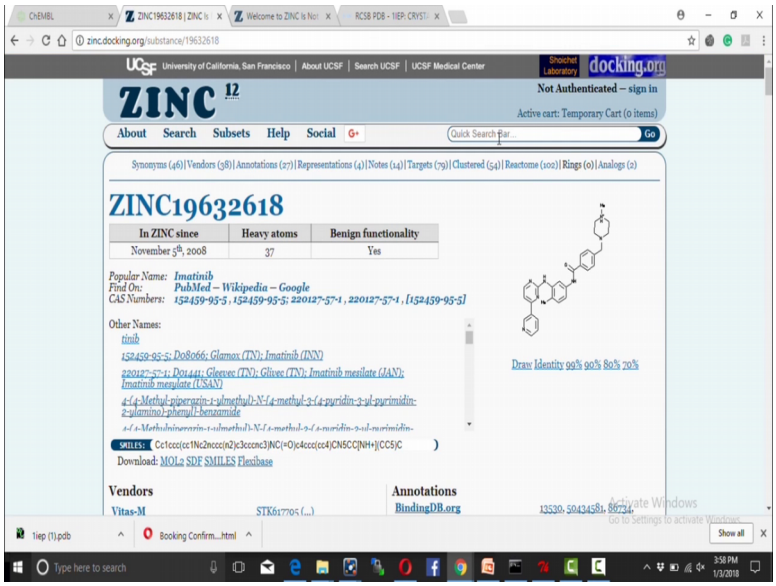

So, once you retrieve the protein then you have to retrieve the ligand, then go to zinc database then type. In search go to search option and then click on text and then give the imatinib and then click enter.

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Then you will get to this you will navigate to this page, which includes the structure of the imatinib and then the various properties of this ligand we have to download this structure in the format of mol 2. So, click mol 2. So, once you downloaded the structure, protein and ligand structure is ready for the docking.

So, you have to open the autodock gui click first we have to prepare the ligand. So, click on ligand in got to input, open whatsoever we have downloaded the imatinib structure you have to input here. So, this is the id zinc 19638618 it is in the mol 2 format just open this.

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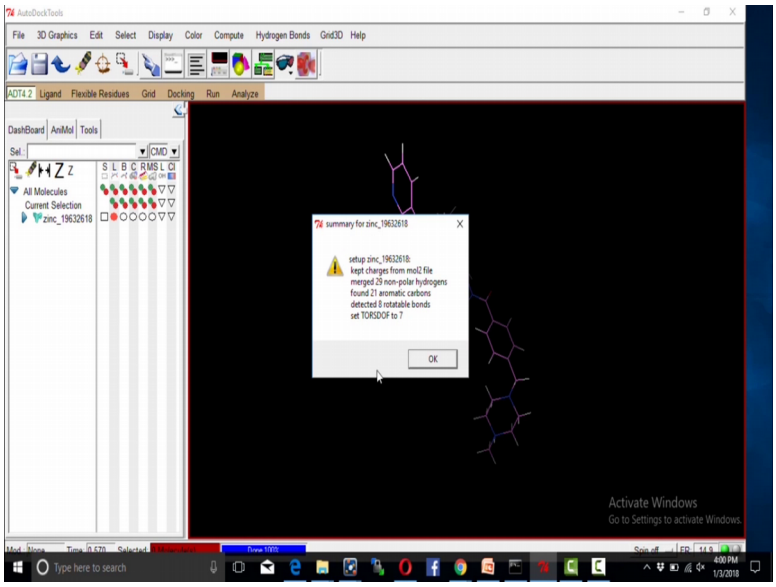

So, this is giving you the information about this ligand, which consist of 29 non polar hydrogen atoms and 21 aromatic carbons and 8 rotatable bonds. So, click. So, this is the structure of your interest, now go to ligand torsion tree deduct the root. This helps you to identify the rotatable bonds in your ligand. So, go to ligand again torsion tree choose torsions.

So, in this we can identify how many bonds are rotatable in the auto in the given ligand. So, in this we can see 7 bonds are rotatable out of 32 bonds. So, there is a amide bond in the given ligand, now we are allowing them also rotatable. So, make amide bond rotatable and click done, go to ligand output save as PDBQT. So, we have to save ligand and the protein in the PDBQT format for the docking purpose.

So, Q stands for the charge, T is the atom type which is specified by the autodock. So, save as PDBQT so, I have save the ligand. Now, we have to prepare the protein. Go to file read molecule so, you have only taken any one of the chain from the 1IEB. So, open that chain now kinase PDB. So, this is the 3 D structure of the Abl kinase.

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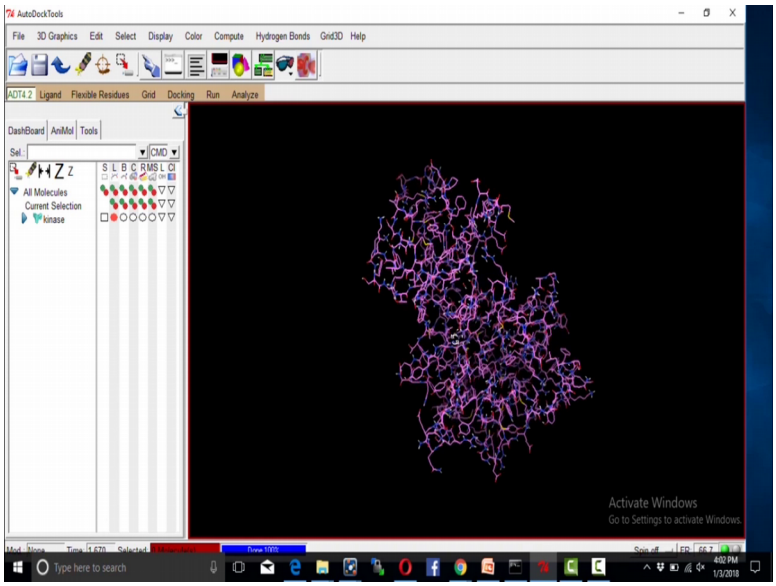

In this auto dock we can see various or display options, L stands for line B stands for ball and stick C for coil and R stands for ribbon. So, I am going to change the option form lines to ribbon. So, this is the 3 D structure of Abl kinase. So, as we all know kinase has two domains this is the kinase domain.

So, we have to prepare the protein first, go to edit and hydre click on hydrogens add polar only q ok. Go to edit atoms assign AD4 atom type, edit charges you have to add charges you can use to charge options one is Kollman charges another one is gasteiger charges. So, I am giving here Kollman charges.

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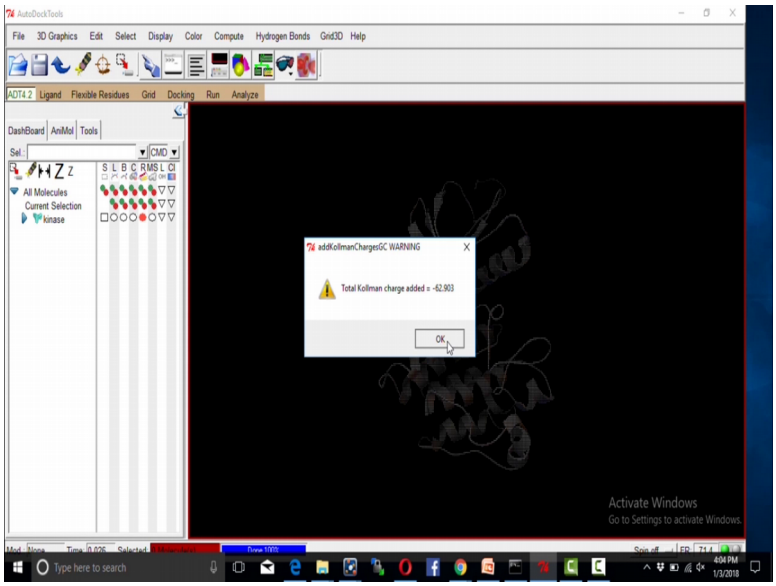

So, Kollman charge will be computed based on the quantum mechanics. So, once you have added the charges then file save PDBQT.

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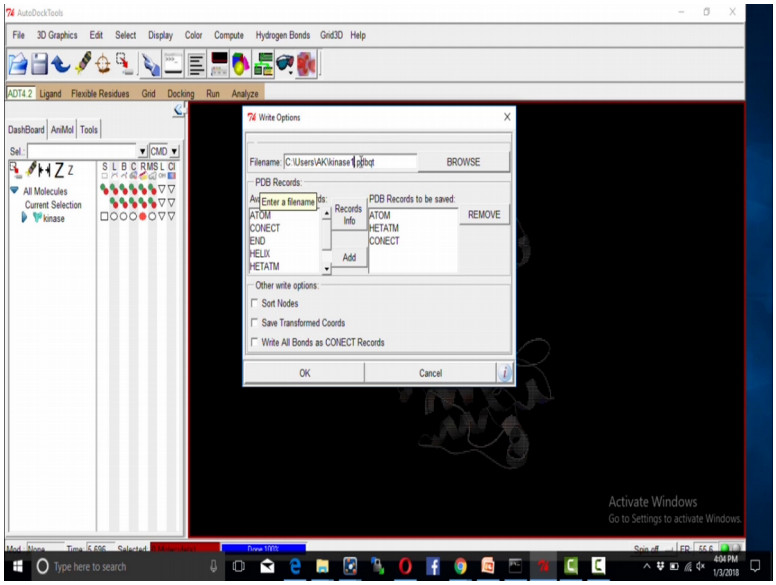

User defined any name, save in your working directory. Now, we have prepared the protein on the ligand

The next step you have to set the grid for the autodock. So, go to grid option click on macromolecule open. So, open this kinase PDBQT which you have prepared currently,

want to preserve the charges. So, this is the structure then you have to open the ligand go to grid box.

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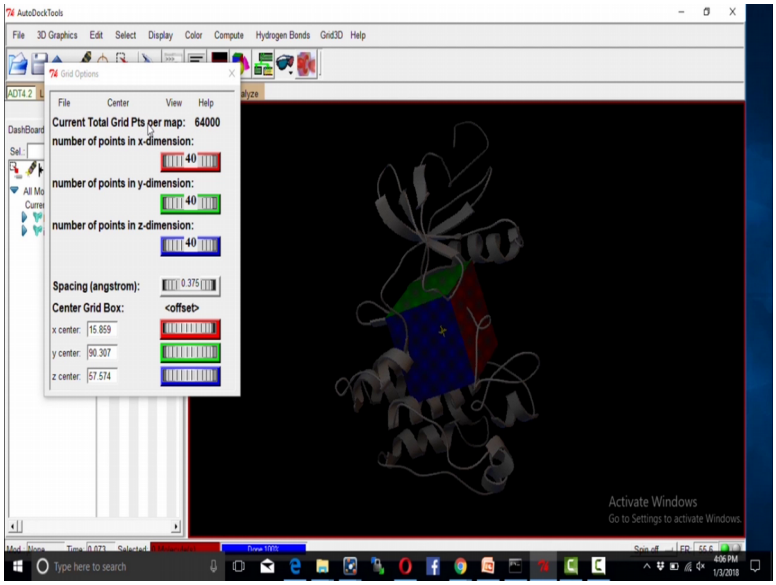

So, here we are going to set up the grid for autodock where you are allowing the ligand to do the conformation search in this specifies space. So, if you know the binding site then you can specify the binding site area; if you do not know the binding site information then you can set the whole protein in a grid. So, that it will do a blind docking

So, in this case I know in the kinase protein we know the binding site. So, I am going to give the grid box in the specified area. So, go to center on macromolecule, then adjust the grid box such that it will cover only this portion. So, you can increase the XYZ dimensions so, that it will cover this space.

Go to file close saving current go to grid output save as a GPF file format. This is grid parameter file you have to save with the GPF extension, then you have to run the autogrid run autogrid specify the executable unspecify your grid parameter file.

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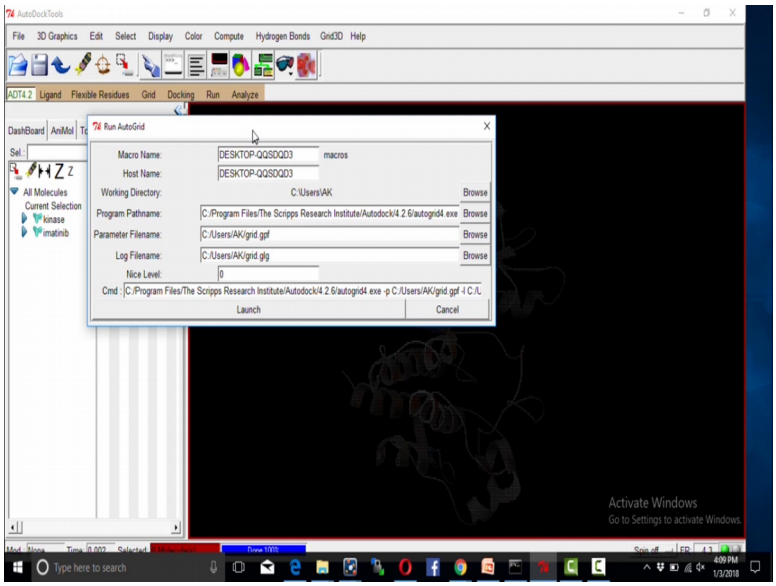

And then click launch it will take some time 5 to 10 minutes to finish the calculation. Usually this grid helps you to identify the interactions between the atom types. So, once you finish the grid we will move on how to do docking.

So, already I am having the output file for the grid. So, after grid it will create the GLG file for the grid. So, this GLG file will be having the information about the different map type, different atom types along with its interactions. So, this will create different map types based on the atoms that you can see here kinase dot A C d e HD maps. So, these are the some of the map file nothing, but these are the interaction mapping of the different atom types in the given specified grid.

So, once you finish this go to docking macromolecule set rigid name.

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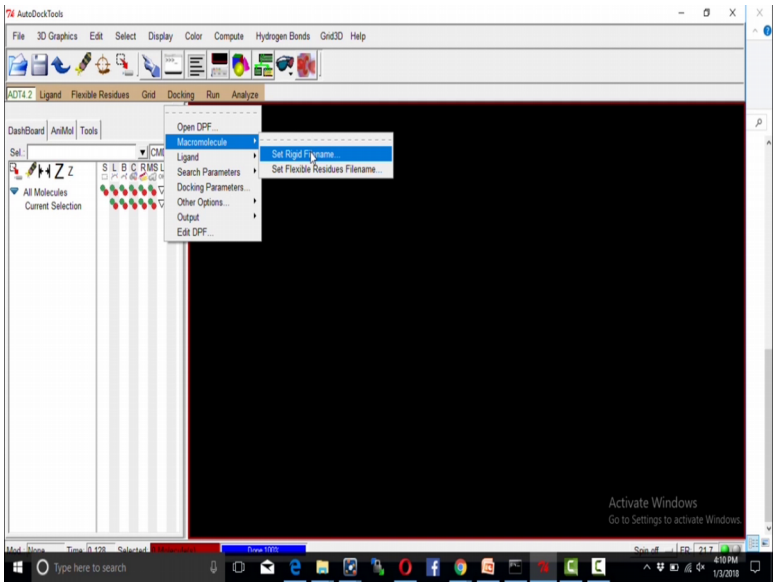

Because in this case we are going to keep protein as a rigid particle and then we are allowing the ligand to move around in this specified grid box. So, open this PDBQT file macromolecule is nothing, but your protein. So, ligand open.

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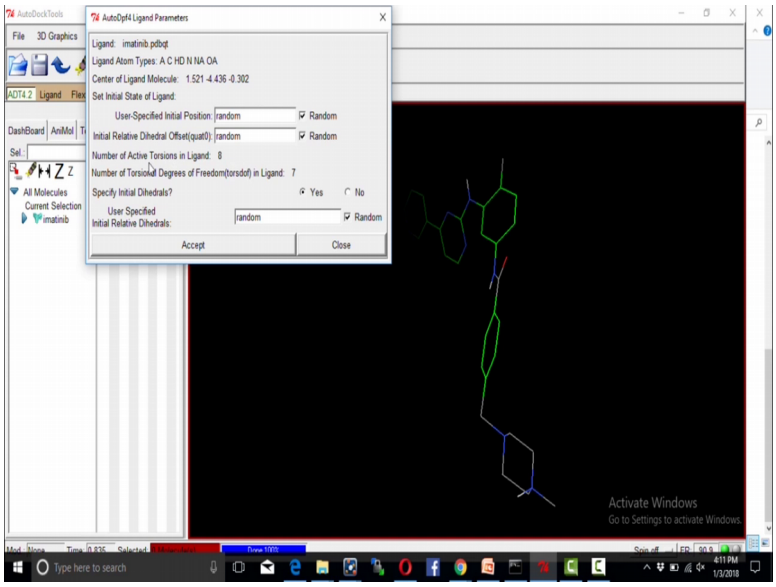

So, this will ask you the options like how many bonds are rotatable. So, here the degrees of freedom is active torsions in the ligand is 8. So, accept this again go to docking; here I am going to use genetic algorithm for doing the conformation search for the ligand in the given grid.

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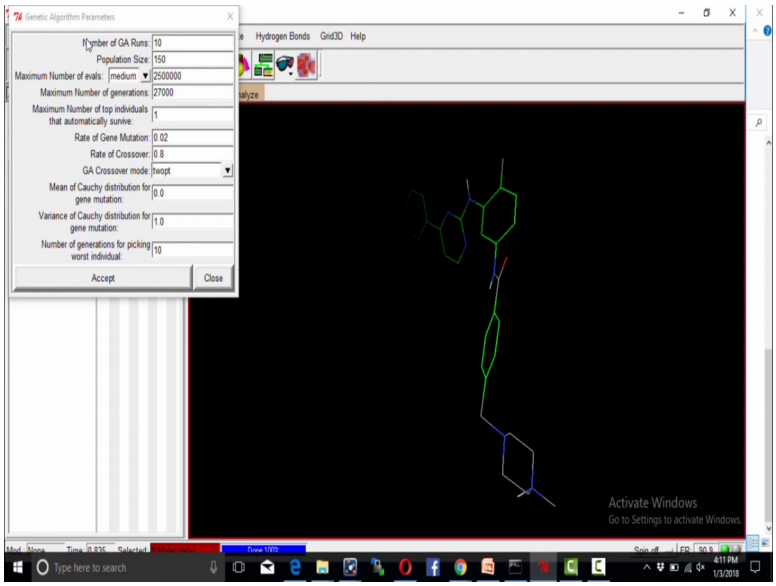

So, here I am going to mention how many conformations you want you can change this options number of GA runs. So, here I need twenty different orientation. So, I am going to specify 20 and accept this. Go to run again other output Lamarckian GA you have to save this file in a dpf file format dpf was nothing, but docking parameter file. So, you can give any user specified name with a dpf extension.

Once you once you created the dpf file, you have to run the autodcok. So, click run, run autodck you have to specify the autodock executable file, then your dpf file, then click launch. So, this will take some moment to generate the various conformers and then it will create a dlg file; dlg file is nothing, but docking log file. So, this is the dlg file. So, go to analyze docking you can open this dlg file from the goi of the autodocking cell open.

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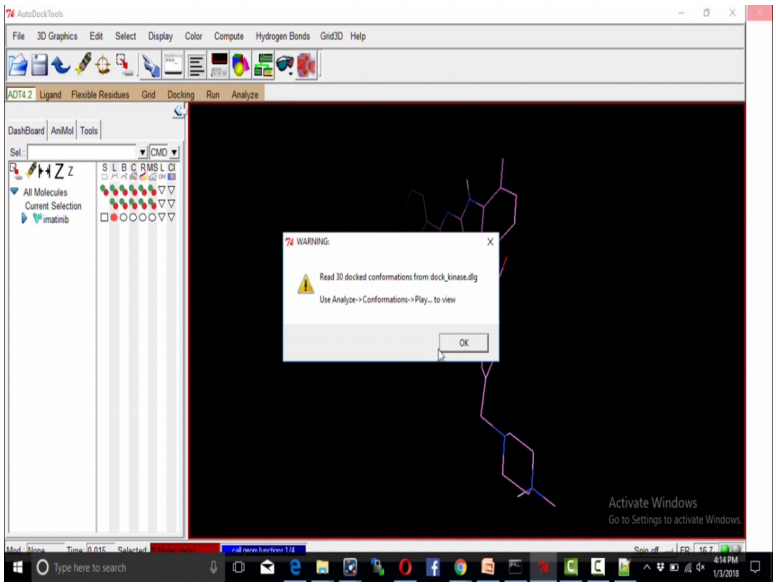

So, this is giving the different conformers of imatinib ligand, this is the docked conformation of the imatinib ligand, go to analyze again macromolecule, open, this is your interest of protein, then change into ribbon, go to analyze conformations and play.

So, in this; in the dlg file, you can see the different conformations using clustering histogram you can identify which conformation is the best conforma binding conformation for the specified protein.

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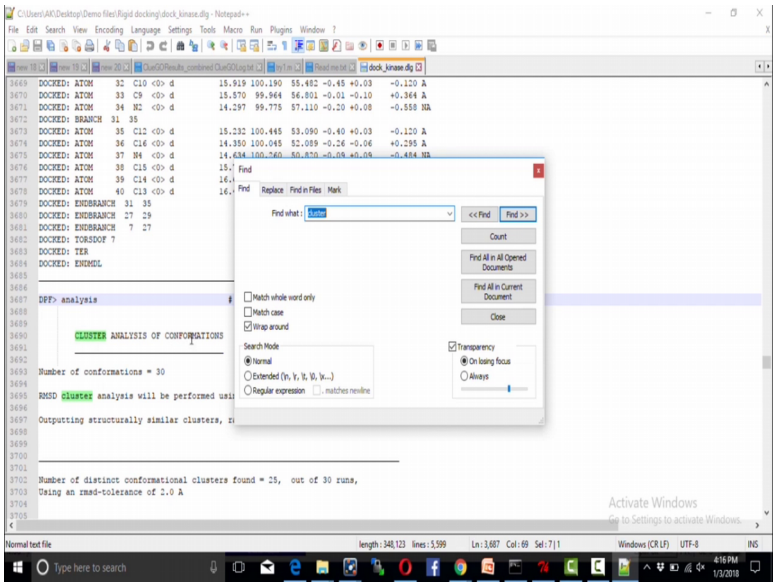

So, this is the clustal analysis of the conformation in the dlg file. So, you can see here I have generated 30 different conformation for this imatinib.

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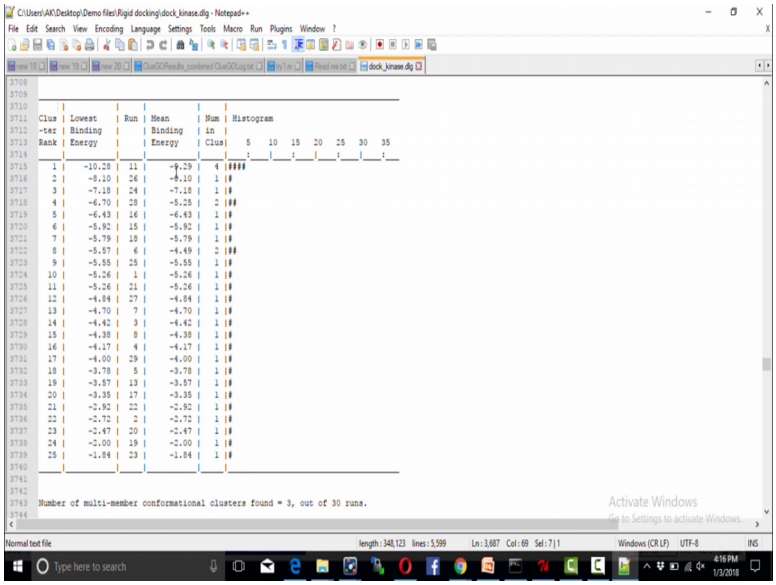

So, here we can see minus 10.28 is the best lowest binding energy for the given imati given ligand, which forms four different four different conformations will fall in this same energy bindings energy binding lowest binding energy.

So, this is the best conformations for the imatinib.

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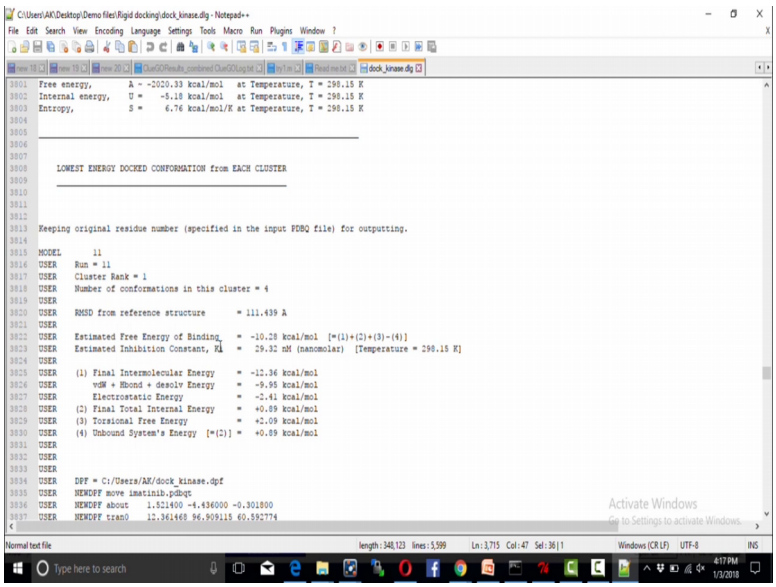

So, go to 11th run 11, you can see here the binding free energy is minus 10.28 kcal and which will be giving you the inhibition constant 29 nano molar and various inter molecular energy parameters along with the xyz coordinates of the given ligand.

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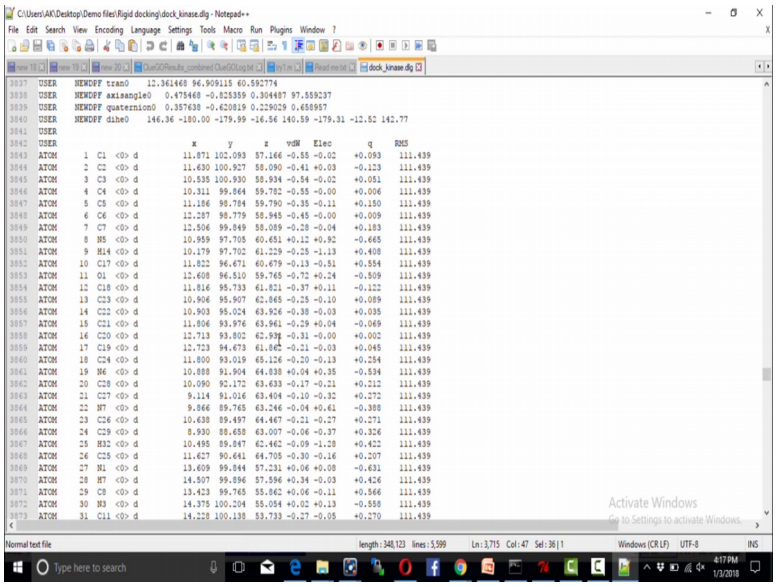

So, this is how the dlg file looks in the after docking. So, go to analyze clustering show the clusters.

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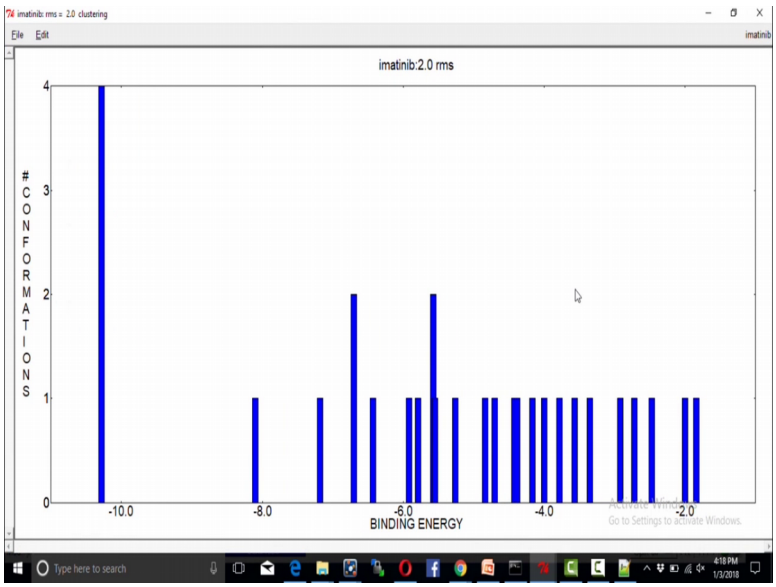

So, here as I showed earlier you can see the four conformation which fall in the category of minus 10 kcal. So, you can select this. So, there are four different you can see here.

Change the ligand conf style to the lines so, that you can see the ligands here, go to this click this option. So, you can save this conformation in a PDBQT format or else PDB format.

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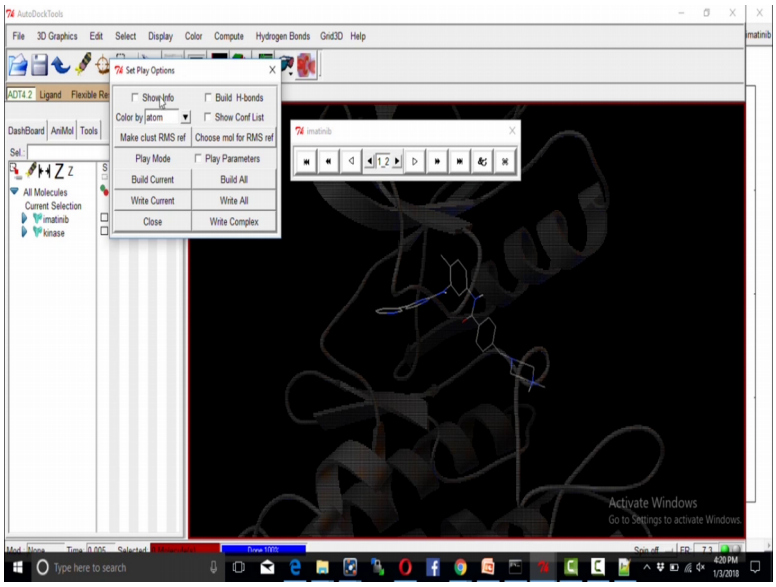

So, I order to do that you have to click this button and then click build current, and then write current. So, this is your best dockedpose. So, you can save as dockedpose 1 the PDBQT format.

So, you can open this file in the primal you can visualize the intra interactions using primal. So, you can use this option build hydrogen bond that will also show you the hydrogen bond which is.

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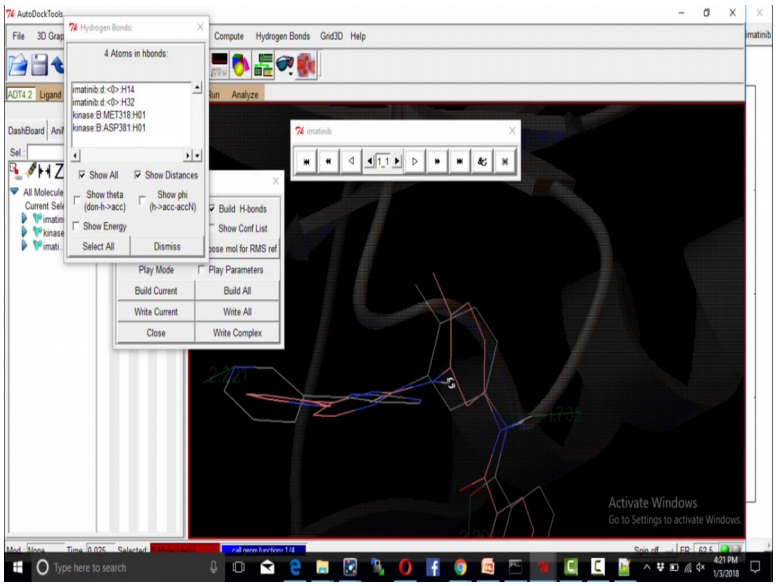

So, which is specified between the ligand and protein protein molecule, here we can see there is hydrogen interaction between the methionine and the ligand with this with this pyridine. Here we can see there are two hydrogen bonds, one is with methionine one is with the aspartic accept. So, this amide forms the hydrogen bond and this carboxyl group also forms in a hydrogen bond.

So, from autodock excel you can visualize the hydrogen hydrogen bonds or else you can open the primal and then you can visualize the hydrogen bonds. So, here we finished with the rigid docking, now if you have multiple ligands, then how to do the docking high proper docking using screening using the autodock vina.

So, in order to do that you have to install the autodock vina, you need to files for it one is configuration file another one is the PDBQT of your protein file, you needed for the screening purpose. So, I will show you how to prepare that. So, already we have seen there are 8 ligands you can go to zinc database, then get the retrieve the mol to file format of those ligands.

Then you have to prepare the ligands, as I told you as I told you earlier you have to go to autodock then prepare the ligand by input open click on input open and open the particular mol to file format given give open see then detect the root and choose torsions and save in a PDBQT file format.

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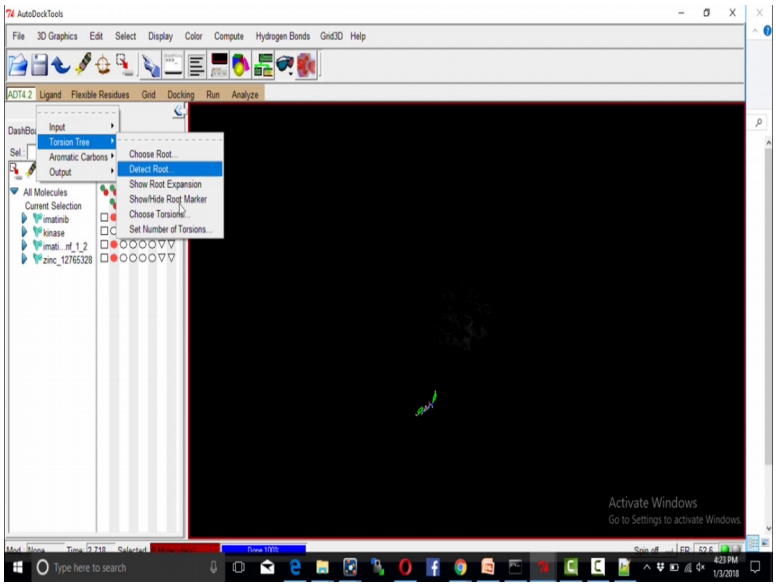

So, like this you have to prepare all the ligands, once you prepared all the ligand and the PDBQT of your protein.

Then you have to do screening, screening you need this configuration file.

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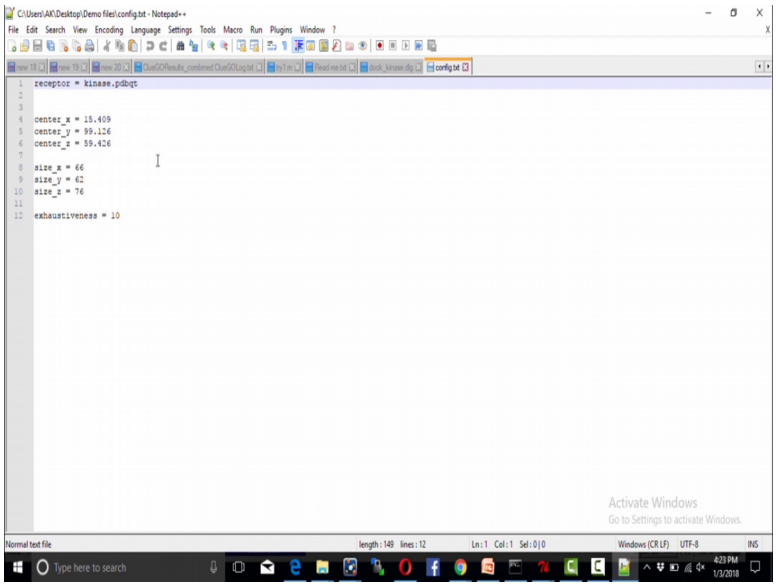

So, this configuration file will be having the details of the receptor which is a kinase in the PDBQT in the PDBQT file format, and then the xyz of the grid this detail also you have to specify. This is the gird I have used for the rigid docking. So, the exhaustiveness for particular ligand how many conformer it should generate; If you want to generate

more conformer, then you can increase the exhaustiveness by giving like 20 or 30. Here I am changing to 20 and save this config dot txt, thing you have to create screen dot bat file.

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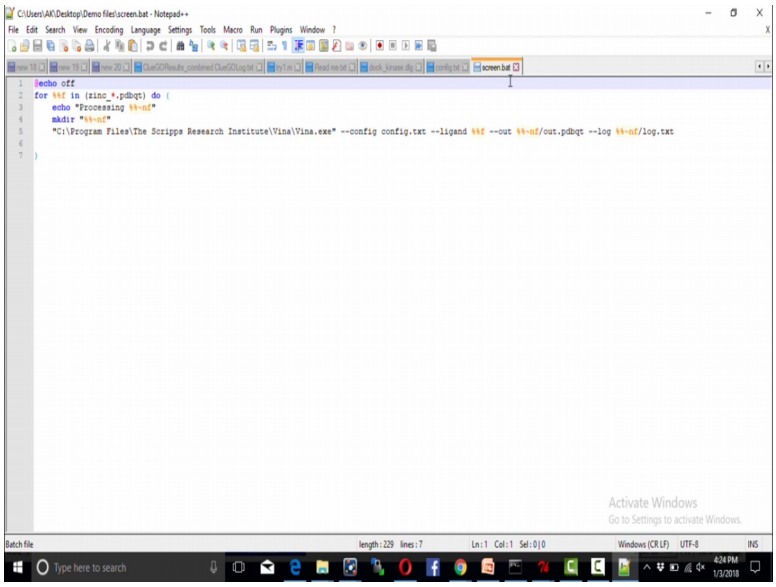

So, screen dot bat file will be having the information of we are having different zinc ids. So, we have prepare the PDBQT file format using the autodock, then you have to you have to specify the zinc. So, it will take all the zinc ids which will start with zinc extension.

So, this script helps you to take the different ligands in the PDBQT format, then it will dock with the specified protein structure and then it will create a separate folders for each ligand with along with the different conformers. So, likewise it will create like zinc with the extension of the id.

So, go to this once you created screen screen dot bat, got to this specified file got to the command prompt. So, this working directory should contain the detail of the PDBQT of your protein the PDBQT of various ligand whatever you want to do this screening and the configuration file and the screen dot bat file.

So, you have to run the screen dot bat file. So, just type screen dot bat and give enter. Once you get enter. So, the program will run and it will give it will produce the different folders starts with zinc along with this id. So, you can open this any one id, then you can

see here out dot PDBQT which will be having the conformation of the particular ligand. So, you can open the autodock then click analyze docking open autodock vina result.

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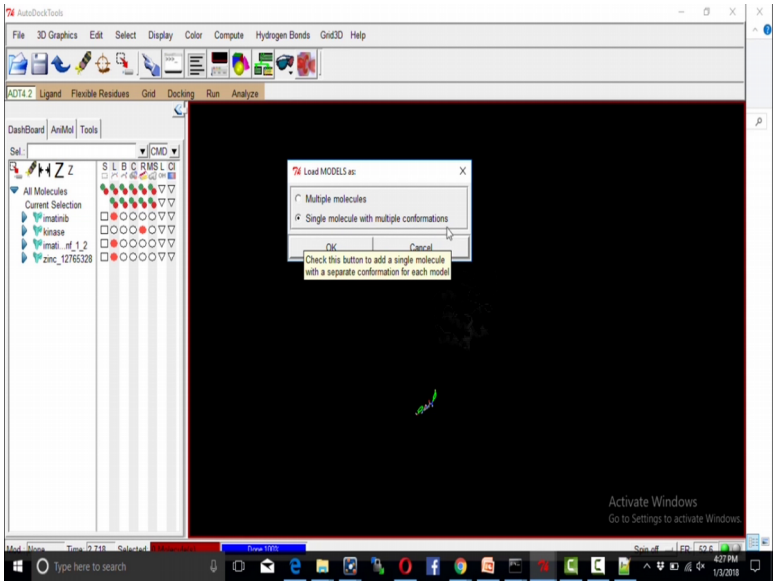

So, it has multiple conformations. So, single molecule with multiple conformation give.

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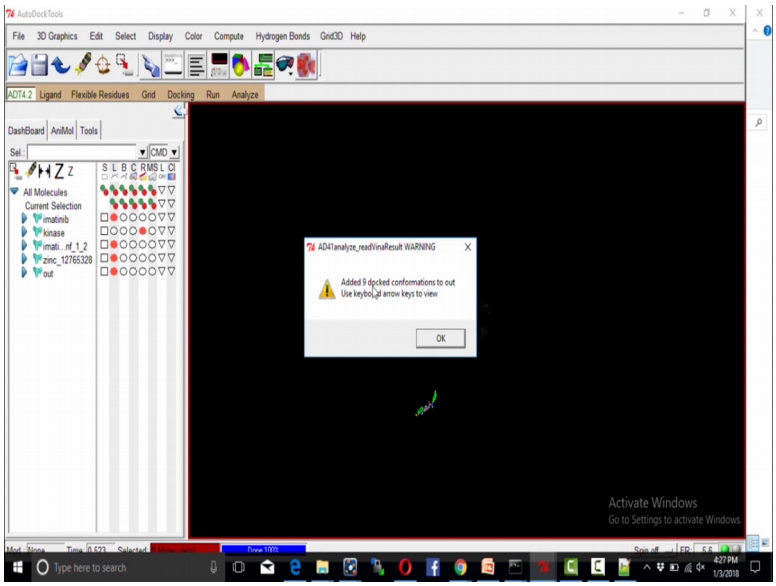

So, there are 9 docked conformation. So, you can analyze the interactions using primal or else yeah.

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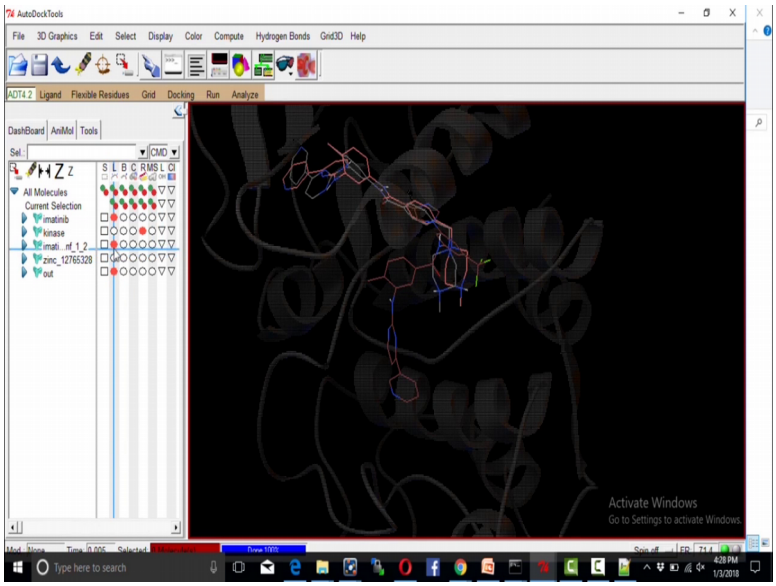

So, this is the structure of the ligand. So, we can see here with the energy. So, the first docked conformer which will be having minus 9.1, the second pose minus 8.9, third one you can see minus 8.8 minus 8.4 likewise. So, if you want to display the interactions, then go to analyze macromolecule docking show interactions.

So, this shows the packet binding packet of this particular ligand. So, these are the different residues, which is in the vicinity of the particular ligand.

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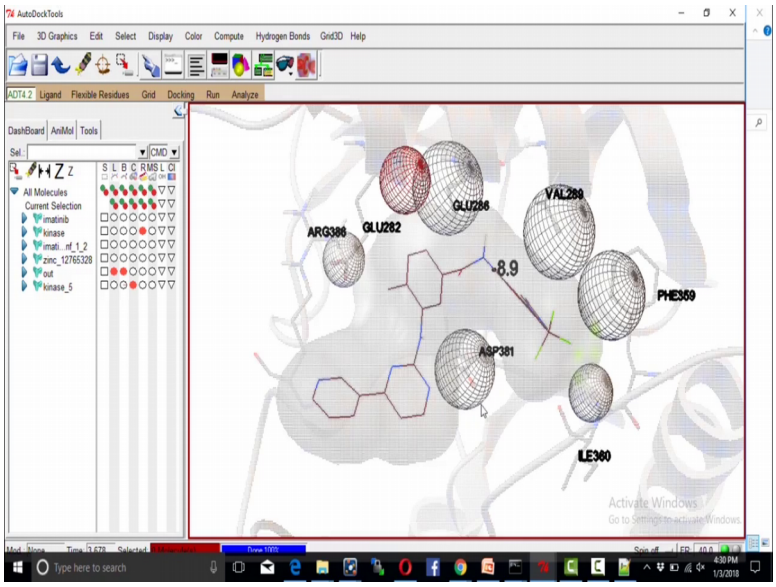

So, you can analyze the interactions using this in by clicking this interaction window. So, from the different ligand you have to select which ligand binds with Abl kinase effectively using screening excel. So, these are the different types of interaction which which will be given from this ligand, which will be given from this ligand.