Introduction to Structure Preparation and Visualization

Created with: Release 2018-4

Prerequisites: Release 2018-2 or higher
Access to the internet

Categories: Molecular Visualization, Structure-Based Design, Ligand-Based Design

Keywords: import files, protein-ligand complex, LigPrep, Protein Preparation Wizard

This tutorial gives an introduction to the Maestro interface and basic visualization tasks. You will learn how to prepare ligand and protein structures, an essential first step for modeling projects. This tutorial also demonstrates how to perform a virtual screen for potential inhibitors of FXa using the ligand docking application Glide. You will learn how to generate a protein receptor grid, dock a set of ligands into the receptor grid, and analyze the docking results.

Words found in the Glossary of Terms are shown like this: Workspace
File names are shown with the extension like this: `1fjs.pdb`
Items that you click or type are shown like this: **File > Import Structures**

This tutorial consists of the following sections:

1. Creating Projects and Importing Structures

At the start of the session, change the file path to your chosen Working Directory in Maestro to make file navigation easier. Each session in Maestro begins with a default Scratch Project, which is not saved. A Maestro project stores all your data and has a `.prj` extension. A project may contain numerous entries corresponding to imported structures, as well as the output of modeling-related tasks. Once a project is created, the project is automatically saved each time a change is made.

Structures can be imported from the PDB directly, or from your Working Directory using **File > Import Structures**, and are added to the Entry List and Project Table. The Entry List is located to the left of the Workspace. The Project Table can be accessed by **Ctrl+T (Cmd+T)** or **Window > Project Table** if you would like to see an expanded view of your project data.

1. Double-click the **Maestro** icon
    - (No icon? See Starting Maestro)

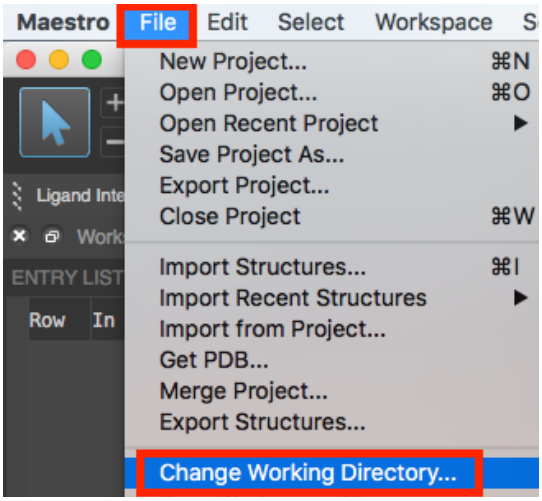
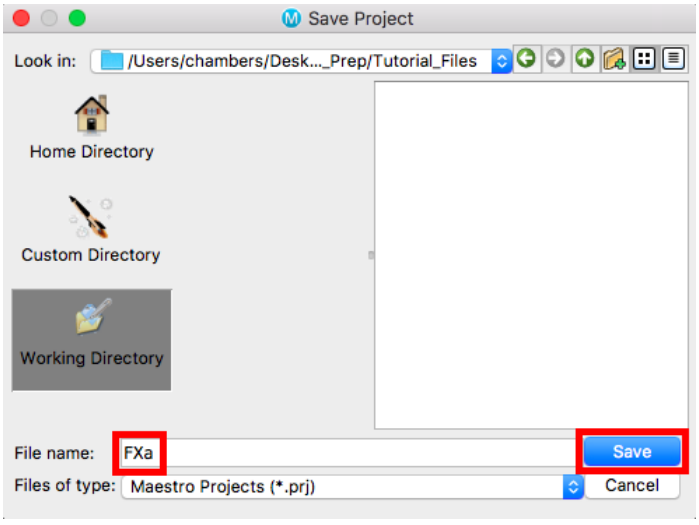


Figure 1-1. Change Working Directory option.

2. Go to **File > Change Working Directory**
3. Find your directory, and click **Choose**



4. Go to **File > Save Project As**
5. Change the File name to **FXa**, click **Save**
    - The project is now named `FXa.prj`

Figure 1-2. Save Project panel.

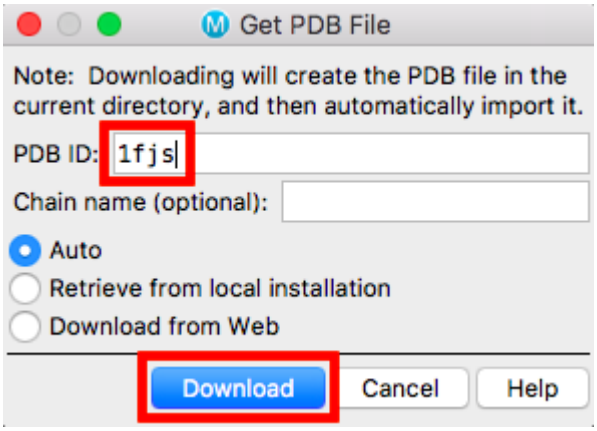


Figure 1-3. Get PDB File panel.

6. Go to **File > Get PDB**
7. For PDB ID, type **1fjs**
8. Click **Download**
    - 1FJS is loaded into the Workspace
    - A banner appears

Note: Banners appear when files have been imported, jobs incorporated into the Entry List, or to prompt a common next step. Here, preparing the protein will be covered in the following section.

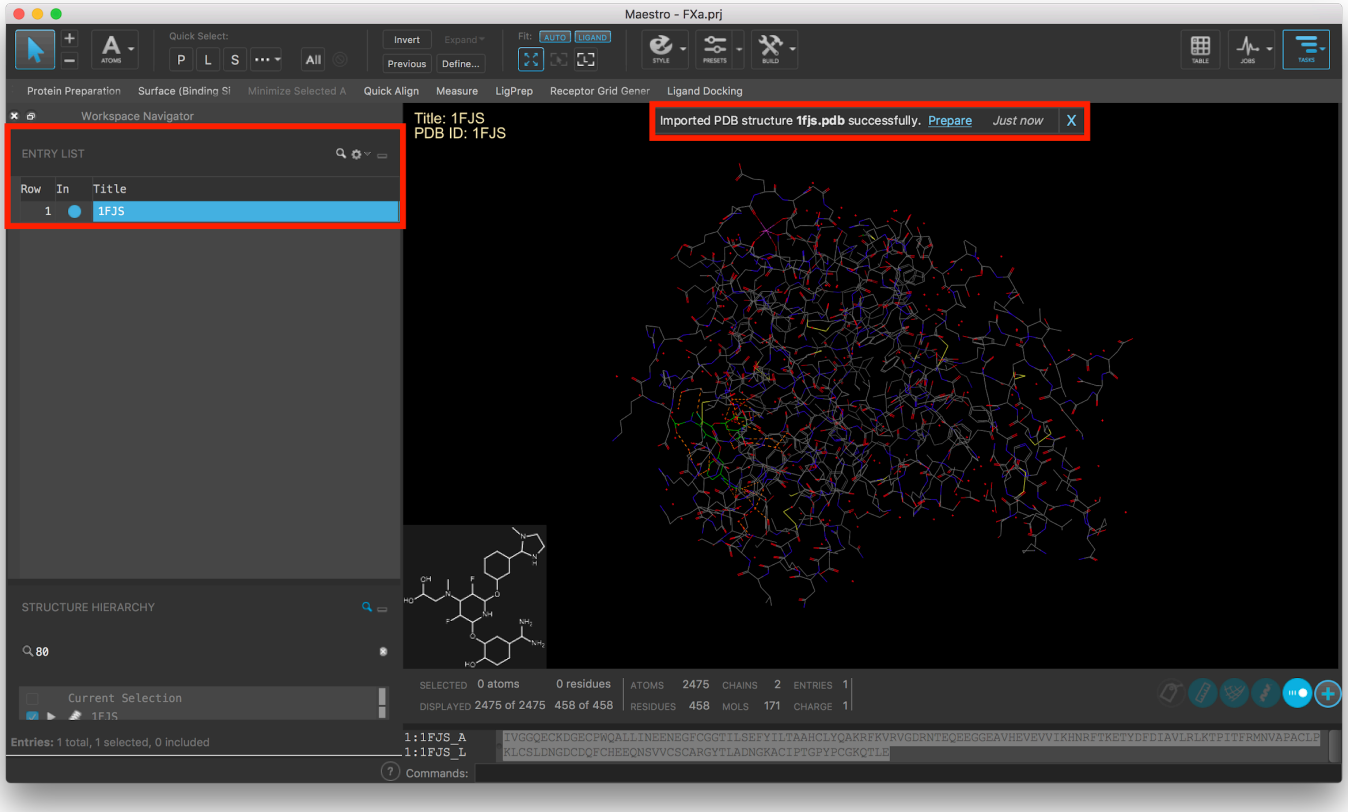


Figure 1-4. The Workspace after the structure is imported from the PDB, with the Entry List and banner highlighted.

Note: By default the structure corresponding to the imported file is both included in the Workspace and selected in the Entry List. Please refer to the Glossary of Terms for the difference between included and selected.

2. Preparing Protein Structures for Glide Docking Model

Structure files obtained from the PDB, vendors, and other sources often lack necessary information for performing modeling-related tasks. Typically, these files are missing hydrogens, partial charges, side chains, and/or whole loop regions. In order to make these structures suitable for modeling tasks, we will use the Protein Preparation Wizard to resolve common structural issues.

The Protein Preparation Wizard has processing, modification, and refinement tools that we will use on the `1FJS.pdb` structure. In the Import and Process tab, the recommended minimal processing tasks are checked by default. There are also options for filling in missing side chains and/or loops, depending on the needs of your structure. The Review and Modify tab shows you all the components of the complex, in separate sections: Chains, Waters, Ligands, and Hets. Here, you can choose which components of the complex to keep or remove.

The Refine tab allows for more detailed modifications to the PDB structure. The H-bond assignment section is used for optimizing the hydrogen bonding network – a process which samples water orientations and flips Asn, Gln, and/or His side chains at a specified pH value. Adjusting the pH will change the protonation states of residues and ligands accordingly, and is useful if you want to accurately reflect the experimental conditions. The Restrained minimization section fixes clashes that can occur with adding hydrogens or filling missing sidechains. By default, an RMSD of 0.3 Å is used, minimizing both the hydrogens and heavy atoms via harmonic penalty constraints. Optionally, hydrogen-only minimization can be chosen.

2.1 Process the protein structure

Figure 2-1. The Protein Preparation Wizard in the Favorites toolbar.

1. In the Favorites toolbar, click **Protein Preparation**
    - The Protein Preparation Wizard opens

Note: You can also click **Prepare** in the banner or find the Protein Preparation Wizard in the Task Tool

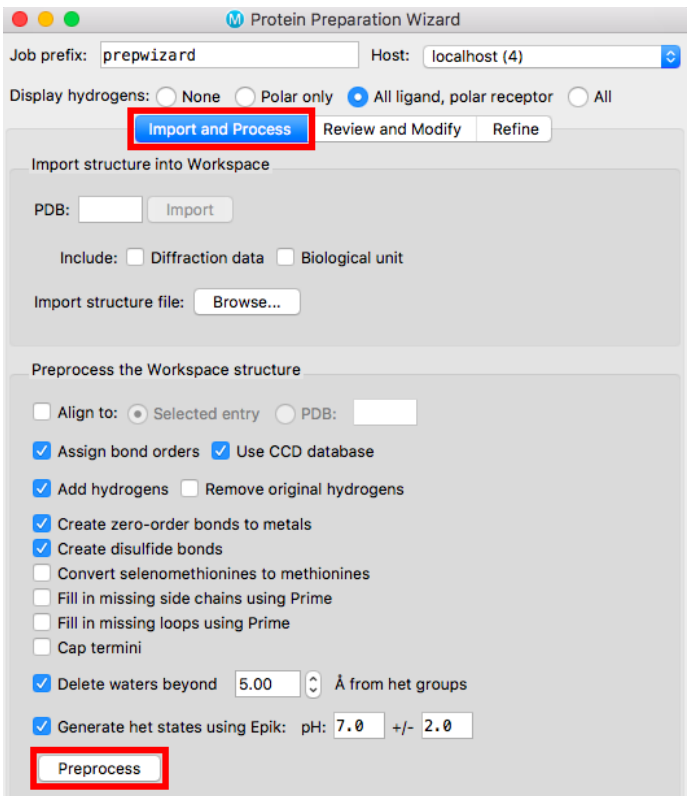


Figure 2-3. Import and Process tab of the Protein Preparation Wizard.

2. In the Import and Process tab, click **Preprocess**
    - A new entry is added to the Entry List and is included in the Workspace
3. In the Protein Preparation - Problems panel, click **OK**
    - The overlapping atoms are fixed in a later step

Note: The ligand bond order is fixed in the 2D Overlay

2.2 Review and Modify the structure

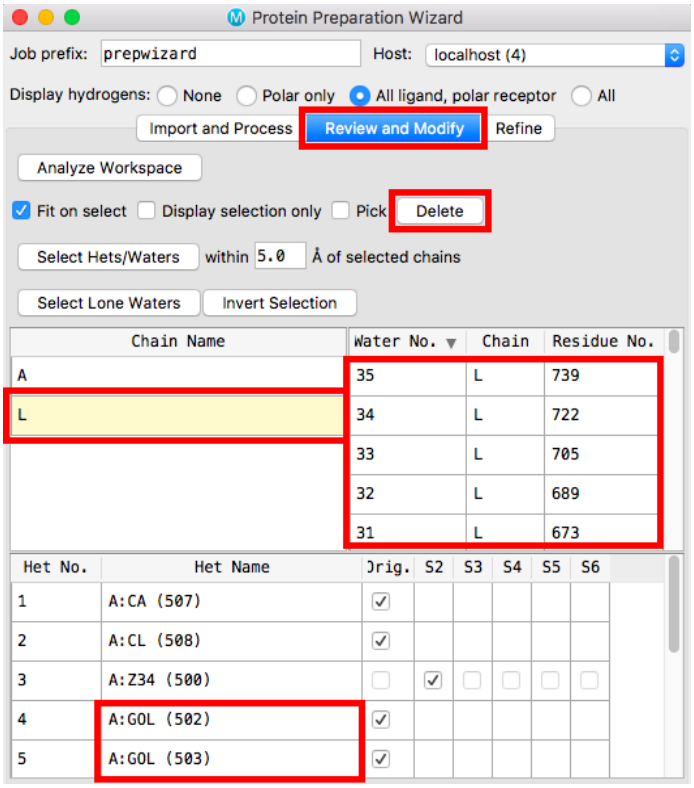


Figure 2-4. The Review and Modify tab before

1. Click the **Review and Modify** tab
2. Under Chain Name, click chain **L**
    - The Workspace zooms to the chain
3. Click **Delete**
    - The smaller of the two chains is removed
4. Shift-click to select all **waters**
5. Click **Delete**
6. In the Hets table, shift-click to select all **GOL** rows
7. Click **Delete**

Note: Depending on your system and research question, you may want to keep certain waters. See Protein Structure Preparation using the Protein Preparation Wizard or Protein Preparation Wizard Panel Help for more details.

removing unwanted components.

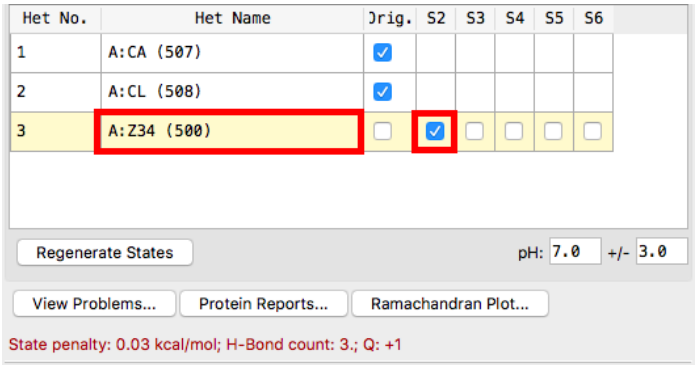


Figure 2-5. Generate different protonation states for the ligand.

8. In the Hets table, click **A:Z34 (500)**
   - Protonation states are generated
   - The lowest penalty state has been automatically checked
9. Click through the **different states**
   - Information is shown in red text at the bottom of the panel
   - The ligand updates in the Workspace
10. Check the **S2** box

2.3 Refine the prepared structure

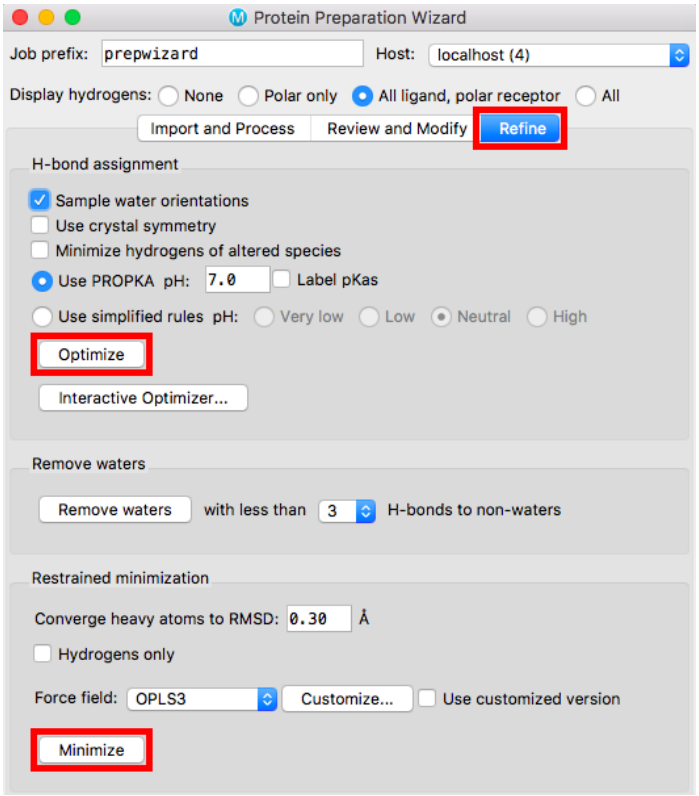


Figure 2-6. The Refine tab of the Protein Preparation Wizard.

1. Click the **Refine** tab
2. Under H-bond assignment, click **Optimize**
   - This step takes ~1 minute
   - A new entry is added to the Entry List
   - The overlapping atoms have been corrected, and side chains that have been flipped are now labeled in the Workspace
3. Under Restrained minimization, click **Minimize**
   - This step takes ~1 minute
   - A new entry is added to the Entry List

Note: Clicking **Interactive Optimizer** allows you to adjust any H-bond assignment

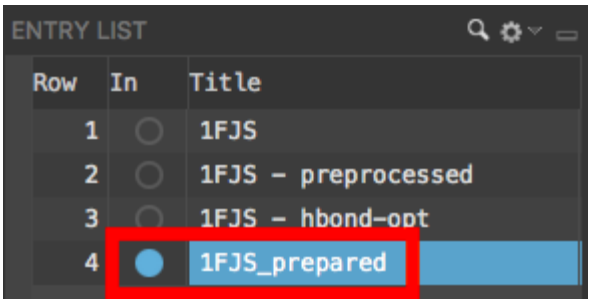


Figure 2-7. Change entry title.

4. In the Entry List, double-click **1FJS - minimized**
5. Type **1FJS_prepared** to rename the entry
6. Click the **Presets** button to zoom in to the ligand binding site.

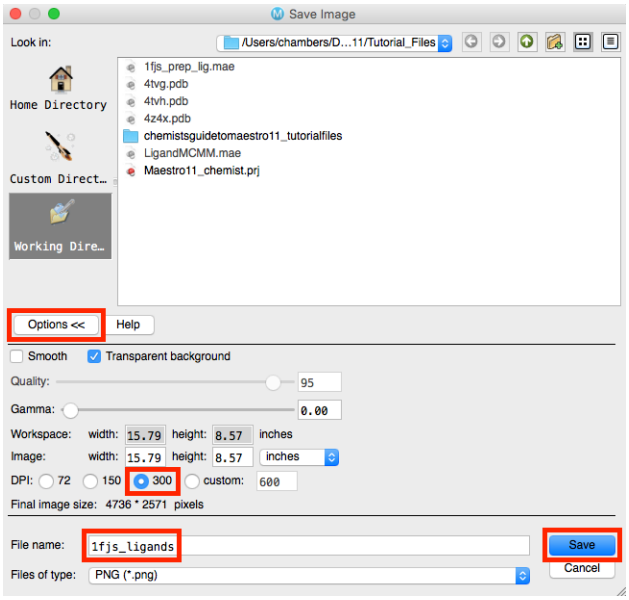


Figure 2-6. Save Image panel with Options open.

7. Click **Workspace > Save Image As**
   - The Save Image panel opens
8. Click **Options >>** to expand image details
9. Check **Transparent background** and **300** DPI
10. Change File name to **1fjs_ligands**
11. Click **Save**
    - A .png image of the Workspace is saved to your Working Directory

Note: If an item is highlighted in the Workspace, the image with be saved with the selection highlights

Note: Go to **Tasks > Browse > Workspace and Project Table Operations** for more options, such as **Ray Trace** for high-quality images

Questions For Comprehension:

1) Choose a PDB structure that you are interested in that has a ligand bound.
   a) What is the three letter code: _____
   b) What is the resolution of the PDB structure? _____
   c) Describe the structure in more detail below.

_____

_____

_____

2) Go through the same workflow from Section 2 with your chosen PDB.
   a) What errors were identified from preprocessing your protein structure?

_____

_____

_____

b) After going through the workflow in the Review and Modify tab, how many protein chains are present? How many solvent molecules, ions and ligands are bound?

_____

_____

_____

c) After going through the workflow in the Refine tab, identify all the pKa of histidine residues in your structure. What are the protonation states of these histidines?

_____

_____

_____

3) After preparing your structure, take steps #4-11 from section 2.3 to create an image of the ligand bound to your protein of interest. What residues are in the binding site?

_____

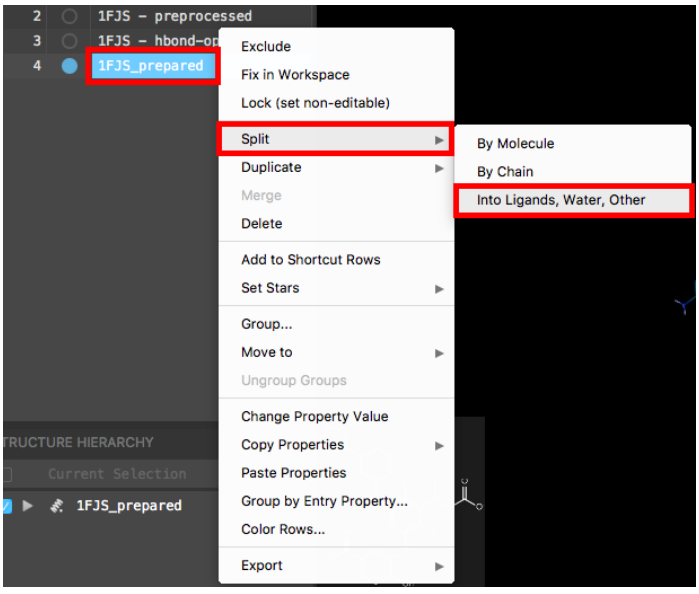
_____

3. Preparing a Ligand Structure

In this section, we will prepare the co-crystallized ligand from the 1FJS structure for use in virtual screening. This is a typical step for cognate ligand docking, as it provides important validation prior to screening a larger ligand data set.

The following steps provide an example of how you would prepare a ligand data set using LigPrep. Ligand files can be sourced from numerous places, such as vendors or databases, often in the form of 1D or 2D structures with unstandardized chemistry. Before being used in a virtual screen, ligands must be converted to 3D structures, with their chemistry properly standardized and extrapolated.

3.1 Split the prepared structure



1. In the Entry List, right-click on **1FJS_prepared**
2. Choose **Split > Into Ligands, Water, Other**
   - Two new entries appear in the Entry List
3. Include **1FJS_prepared_ligand**
   - Only the ligand is displayed in the Workspace

Figure 3-1. Right-click to split an entry into different components.

3.2 Run LigPrep

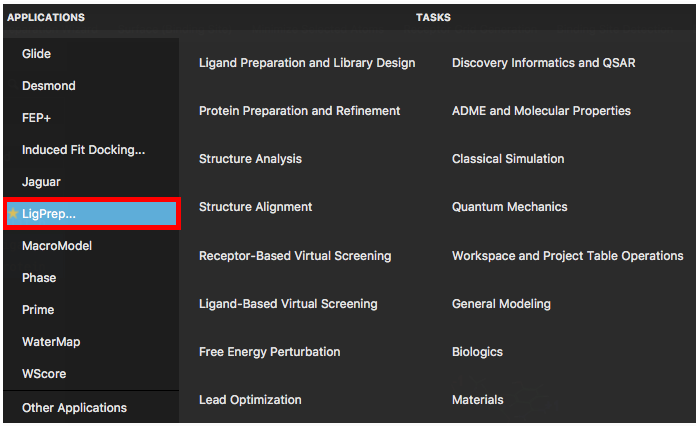


Figure 3-2. LigPrep application in the Task toolbar.

1. Go to **Tasks > Browse > LigPrep**
    - The LigPrep panel opens

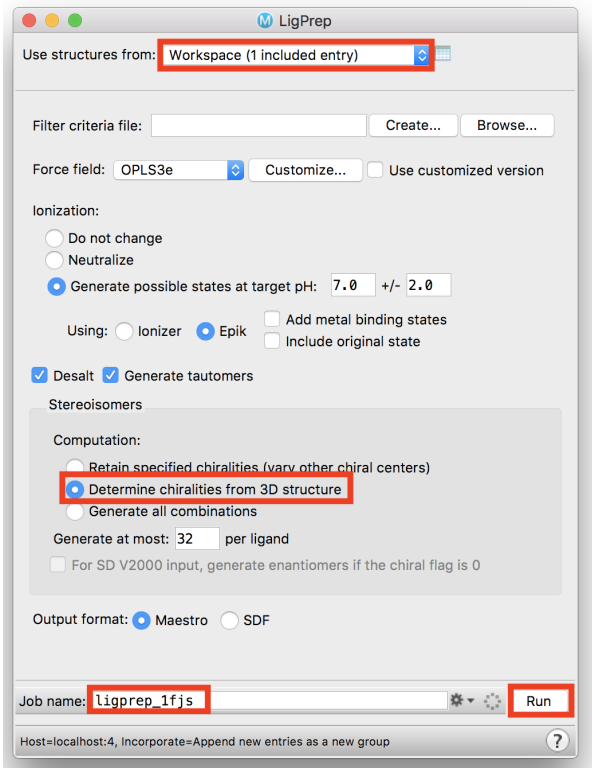


Figure 3-3. The LigPrep panel.

2. For Use structures from, choose **Workspace (1 included entry)**
3. Under Stereoisomers, choose **Determine chiralities from 3D structure**
4. Change Job name to **ligprep_1FJS**
5. Click **Run**
    - A banner appears when the job has been incorporated
    - A new group is added to the Entry List
    - The number of ligands in this group is shown in parentheses

Note: The Tile functionality is very useful for seeing the slight variations in chemistry for the generated structures. The Tile View can be turned on by clicking the + in the Workspace Configuration Toolbar in the bottom right corner and then clicking the Tile button.

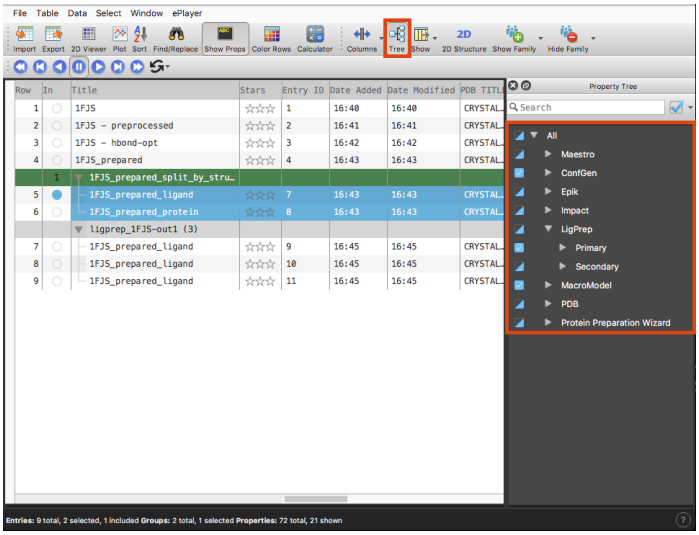


Figure 3-4. The Project Table with the Property Tree open.

6. Type **Ctrl+T (Cmd+T)** to open the Project Table
7. Click **Tree** to open the Property Tree
    - Different calculated properties can be toggled on and off
    - Click the **arrow** next to each application to view more properties

Questions For Comprehension:

4) Go through the same workflow from Section 3 with your chosen PDB.
    a) What ligand states were output from LigPrep? Draw them below:

b) How do these ligand states compare to the crystal structure? Use the Toggle tab to take a picture of the Epik generated states compared to the crystal structure.

4. Visualizing Protein-Ligand Complexes

In this section, we will explore ways to visualize structures in the Workspace. Object representation can be changed in a number of ways using the Style toolbox. Presets offers the ability to quickly render a structure in a number of styles, similar to PyMOL, to facilitate easy visualization. Presets can be used in a variety of ways, from de-cluttering your structure to creating publication-quality images. We will analyze the protein-ligand complex by looking at the interactions that are made and the surface of the binding pocket. Finally, we will save an image of the complex.

4.1 Use the Style toolbox

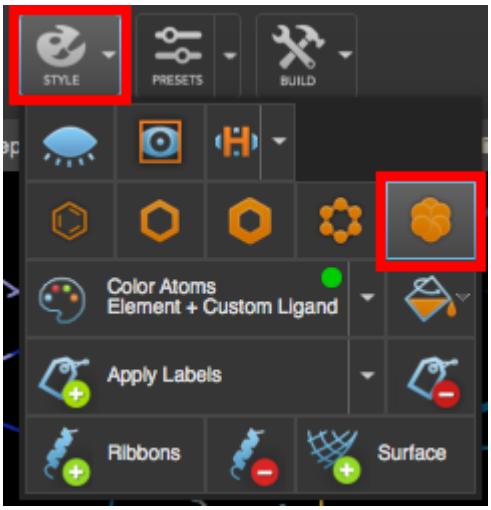


Figure 4-1. The Style toolbox with CPK representation highlighted.

1. Include entry **1FJS_prepared**
2. Type **L**
    - The Workspace zooms to the ligand
3. Under Quick Select, click **L**
    - The ligand is selected
4. Click **Style**
5. Choose **CPK** representation
    - The ligand is rendered in space-filling (CPK) representation
    - This is only applied to the ligand, since nothing else is selected in the Workspace

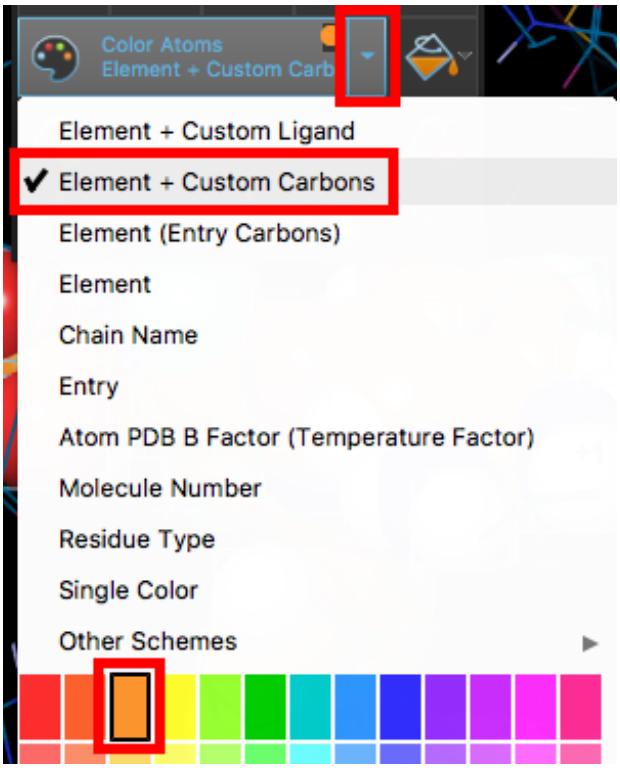


Figure 4-2. The Color Atoms menu.

6. Click the **Color Atoms arrow**
7. Choose **Element (Custom Ligand)**, and pick **orange** from the secondary menu
    - Ligand carbon atoms are orange
8. Under Quick Select, click **P**
    - The protein is selected
9. Type **Z**
    - The Workspace is zoomed to view the selected structure
10. In the Style toolbox, click **Ribbons**
    - Ribbons are added to the protein

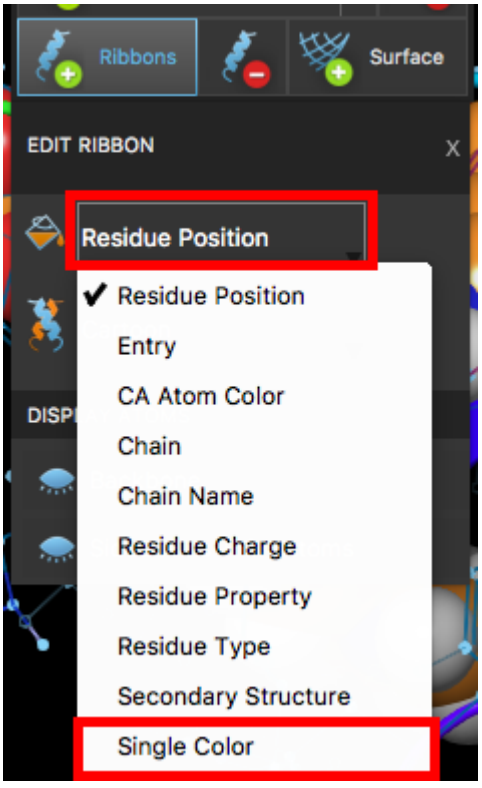


Figure 4-3. The Edit Ribbon panel.

11. Right-click on the **ribbon**
    - The Edit Ribbon panel opens

Note: Use the predictive highlighting to know when you will click on the ribbon.

12. Click **Residue position** in the color scheme
13. Choose **Single Color**

Note: Click the box to the right of the color scheme to choose different colors

4.2 Apply a Preset style

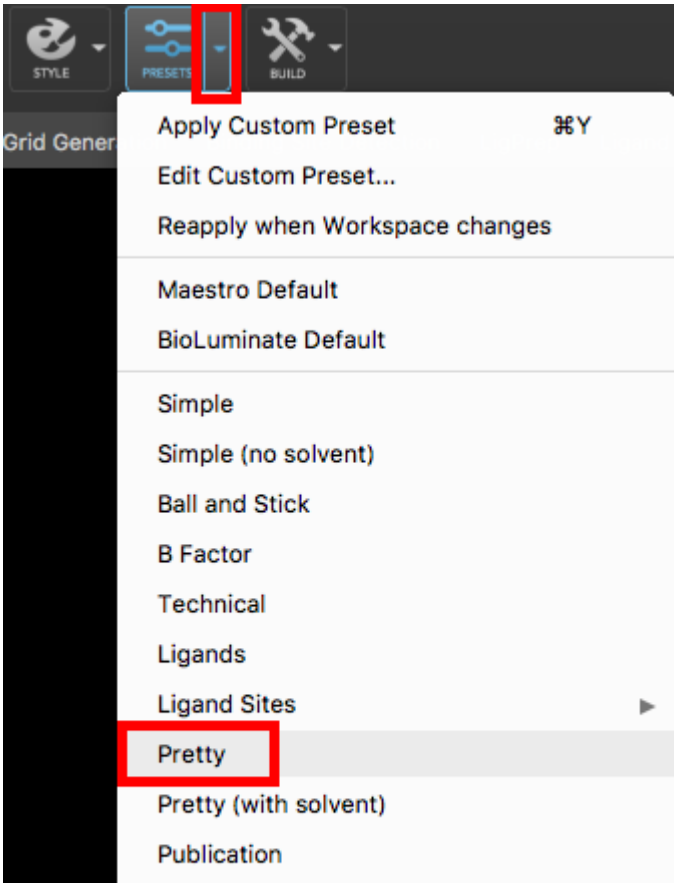


Figure 4-4. The Presets menu.

1. Click the **Presets arrow**
2. Choose **Pretty**
    - The Workspace is rendered with ribbons, a green thick-tube ligand, and side chains are hidden
3. Click **Presets**
    - The Workspace is redrawn with the Custom Preset
    - The Workspace zooms to the ligand

4.3 Visualize Interactions

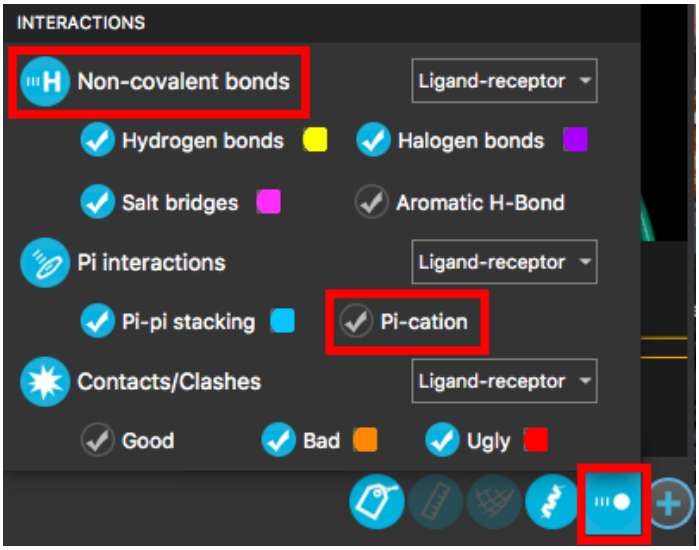


Figure 4-5. The Interactions panel in the Workspace Configuration Toolbar.

1. In the Workspace Configuration Toolbar, right-click **Interactions**
    - The Interactions panel opens
2. Turn on **Non-covalent bonds**
3. Turn off **Pi-cation** interactions

Note: Clicking the color to the right of each interaction opens the Preferences panel, where the interaction visualization can be customized

Note: The threshold for Contacts/Clashes is set to 0.89 for bad and 0.75 for ugly. These values correspond to the ratio of the distance between the two atoms and the sum of their Van der Waals radii.

4.4 Create a custom set

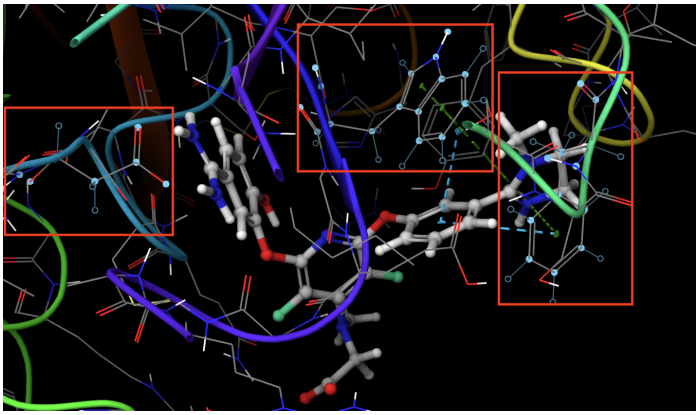


Figure 4-6. More options in Quick Select.

1. Type **R** to switch to residue picking
2. Ctrl+Click to select the binding site residues **PHE 174**, **TRP 215** and **ASP 189**

Note: Residues can be located by residue number and type in the Structure Hierarchy

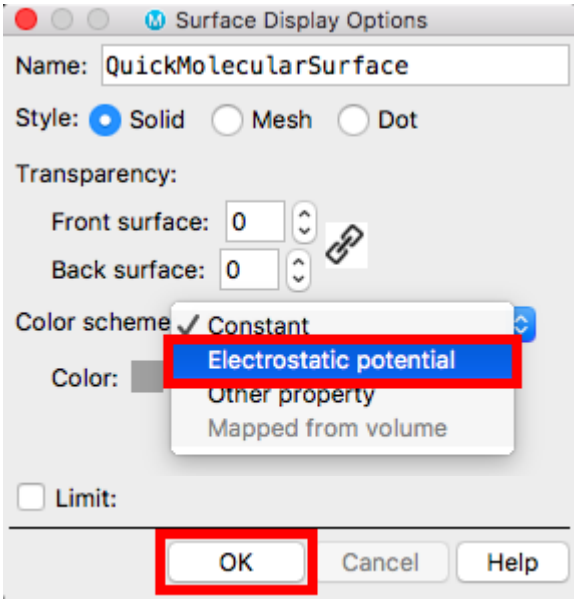


Figure 4-7. The Surface Display Options panel.

1. Right-click the **surface**
2. Choose **Display Options**
   - The Surface Display Options panel opens
3. For Color Scheme, choose **Electrostatic Potential**
4. Change the Min and Max values to **-0.1** and **0.1**, respectively
5. Click **OK**
   - The intensity of the surface colors is increased

4.5 Generate and manipulate a surface

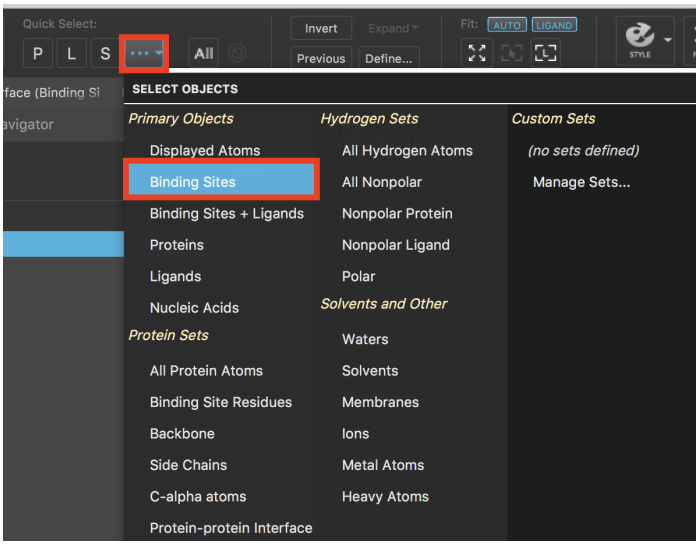


Figure 4-6. More options in Quick Select.

6. Under Quick Select, click **…** and choose **Binding Sites**
7. Click **Style** and choose **Surface**
    - A solid gray surface is applied
    - An S is next to the title in the Entry List, click to see surface options

Note: Click **Surface (Binding Site)** in the Favorites toolbar to perform the same task

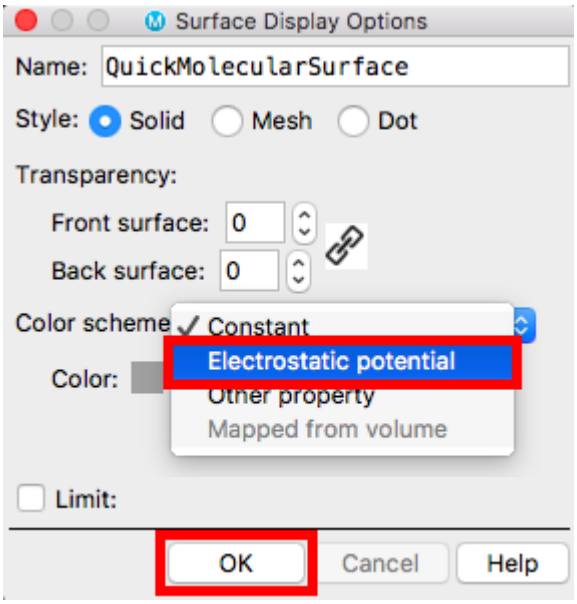


Figure 4-7. The Surface Display Options panel.

8. Right-click the **surface**
9. Choose **Display Options**
    - The Surface Display Options panel opens
10. For Color Scheme, choose **Electrostatic Potential**
11. Change the Min and Max values to **-0.1** and **0.1**, respectively
12. Click **OK**
    - The intensity of the surface colors is increased

4.5 Generate a 2D interaction diagram

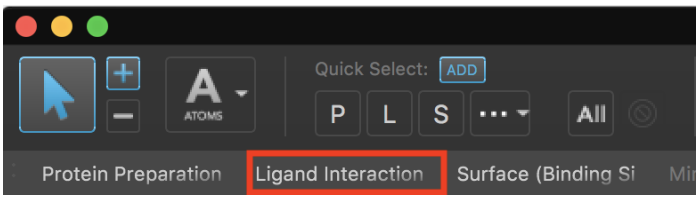


Figure 4-8. Ligand Interaction Diagram in the Favorites toolbar.

1. In the Favorites toolbar, click **Ligand Interaction**
    - The 2D Workspace - Ligand Interaction Diagram opens

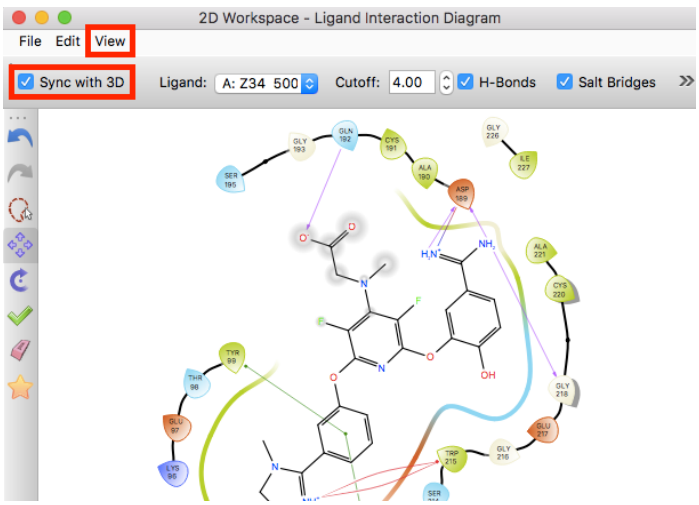


Figure 4-9. The Ligand Interaction Diagram with Sync with 3D turned on and LID legend open.

2. Check **Sync with 3D** and rotate the ligand in the Workspace
    - Ligand orientation is changed in the 2D representation
3. Choose **View > LID Legend**

Note: Images can be saved via **File > Save Screenshot**

Note: The residue icon point indicates the direction of the sidechain

Questions For Comprehension:

5) Go through the same workflow from Section 4 with your chosen PDB.
    a) What are the key interactions between the protein and ligand from your chosen PDB?

    _____

    _____

    _____

    _____

    _____

    _____

    b) Design another ligand that you think would bind with similar potency to the crystallized ligand. Justify your design below:

    _____

    _____

    _____

    c) Create a new entry and draw the ligand you designed in the 2D Sketcher (Edit > 2D Sketcher) and save it as MyLigand in the Entry list. LigPrep this ligand as well.

5. Generating a Receptor Grid

Grid generation must be performed prior to running a virtual screen with Glide. The shape and properties of the receptor are represented in a grid by fields that become progressively more discriminating during the docking process. To add more information to a receptor grid, different kinds of constraints can be applied during the grid generation stage. For a comprehensive overview of constraint options, see the grid generation videos on our website or the Glide User Manual (**Help**

> Help > User Manuals > Glide User Manual). In this tutorial, we will set a hydrogen bond constraint in our receptor grid.

5.1 Identify the binding site

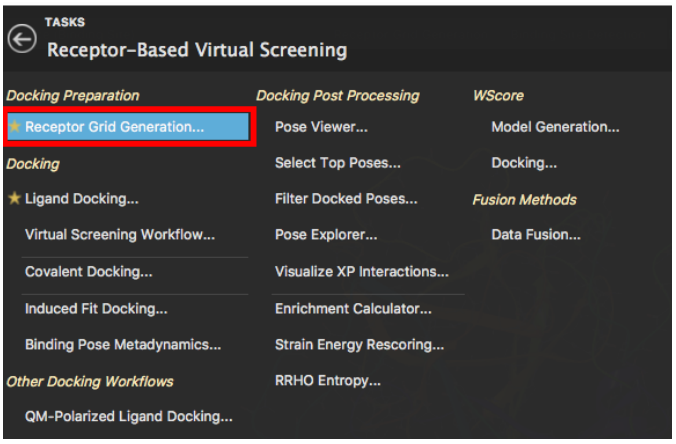


Figure 5-1. Receptor Grid Generation option in Receptor-Based Virtual Screening.

1. Click the **In** circle next to **1fjs_prepared** to include it in the Workspace
2. Go to **Tasks > Browse > Receptor-Based Virtual Screening > Receptor Grid Generation**
    - The Receptor Grid Generation panel opens

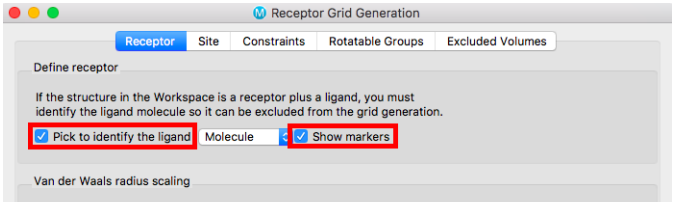


Figure 5-2. The Receptor tab of Receptor Grid Generation.

3. Under Define Receptor, check the boxes for **Pick to Identify the ligand (Molecule)** and **Show Markers**
    - A banner in the Workspace will prompt you to click on an atom in the ligand

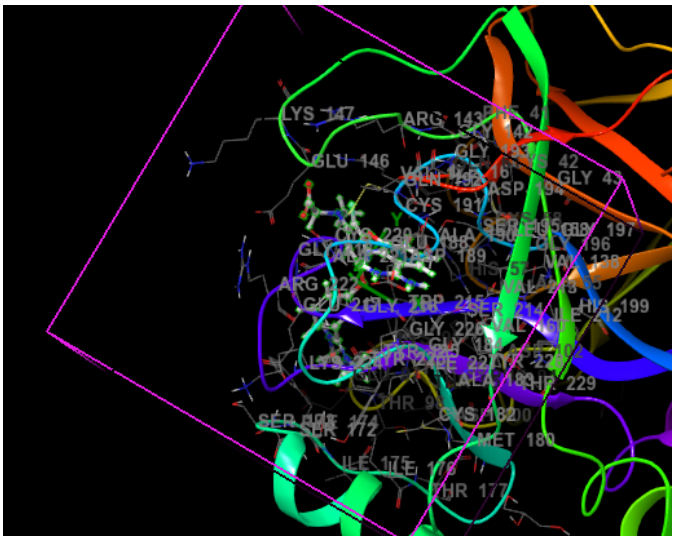


Figure 5-3. The ligand is defined to be excluded from grid generation.

4. Click on the **ligand**
    - The ligand is now highlighted with a purple box around it
    - The ligand will be excluded from the grid generation

Note: The purple bounding box defines the region that the docked molecule(s) can occupy to satisfy the initial stages of docking

5.2 Define the bounding box dimensions

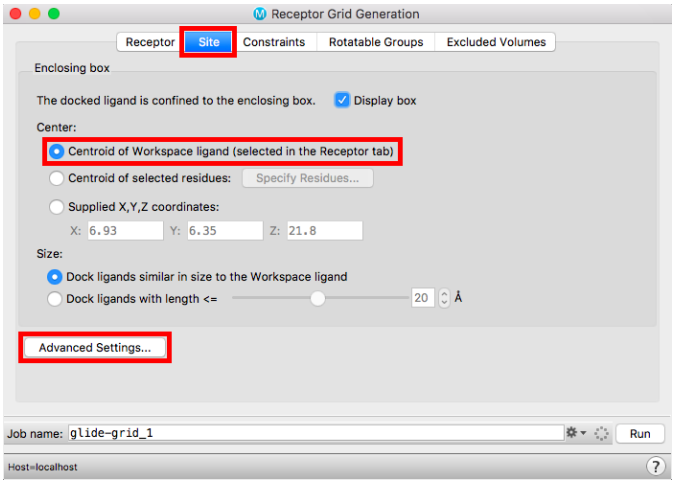


Figure 5-4. The Site tab of Receptor Grid Generation.

1. Click the **Site** tab
2. Select **Centroid of Workspace ligand (selected in the Receptor tab)**
3. Click **Advanced Settings**
    - A green inner bounding box appears

Note: The green bounding box defines the region in which the centroid of the docked molecule(s) must occupy to pass the initial stages docking

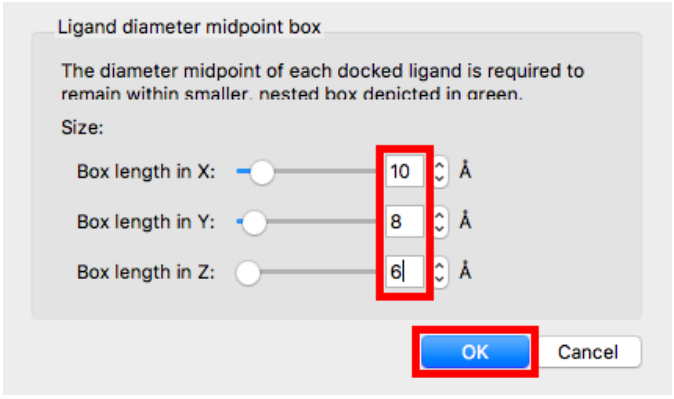


Figure 5-5. Ligand diameter midpoint box panel.

4. Adjust the settings for **X**, **Y**, and **Z** sizes to **10**, **8**, and **6** Å , respectively.
    - The shape of the green box is changed
5. Click **OK**

5.3 Set a hydrogen bonding constraint

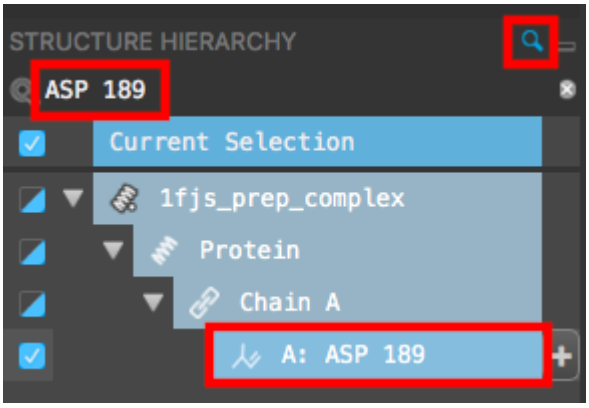


Figure 5-6. Search in the Structure Hierarchy.

1. Type **L** to zoom to the ligand
2. In the Structure Hierarchy, click the **magnifying glass**
3. In the search field, type **ASP 189**
4. Select **ASP 189**

Note: Please see the Introduction to Structure Preparation and Visualization tutorial for instructions on how to add residue labels and show H-bonds

5. Under Fit, click **Fit view to selected atoms**

Figure 5-7. Zoom to selected atoms.

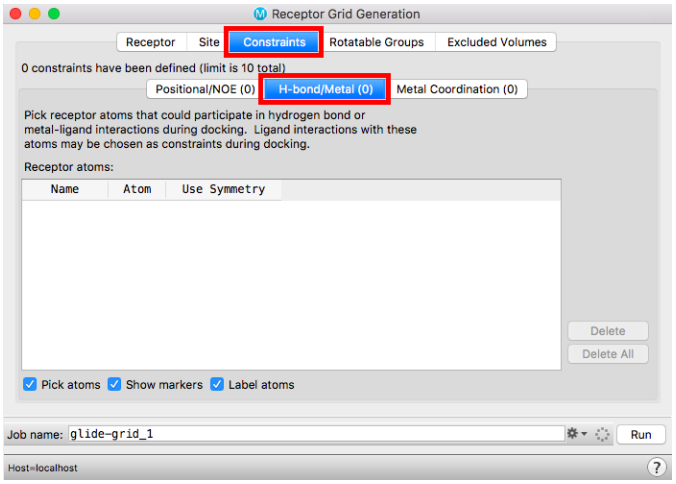


Figure 5-8. The Constraints tab of Receptor Grid Generation.

6. In the Receptor Grid Generation panel, click the **Constraints** tab
7. Click the **H-bond/Metal (0)** tab
    - A banner appears prompting selection of the receptor atom to be the constraint

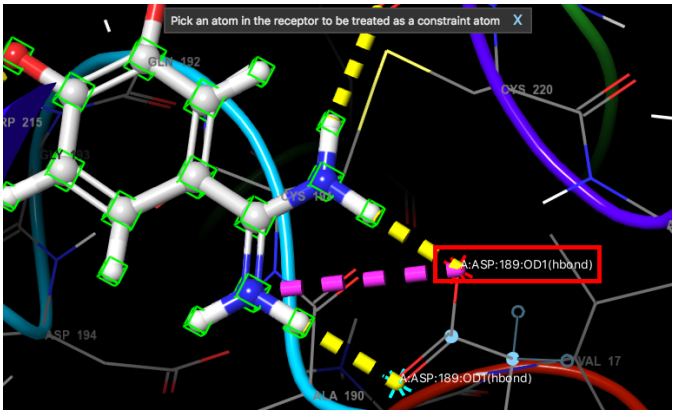


Figure 5-9. Constraint defined on ASP 189.

8. Click an **oxygen atom** of the ASP 189 sidechain
    - Both oxygens are highlighted
    - An H-bond constraint is defined in the Receptor atoms table

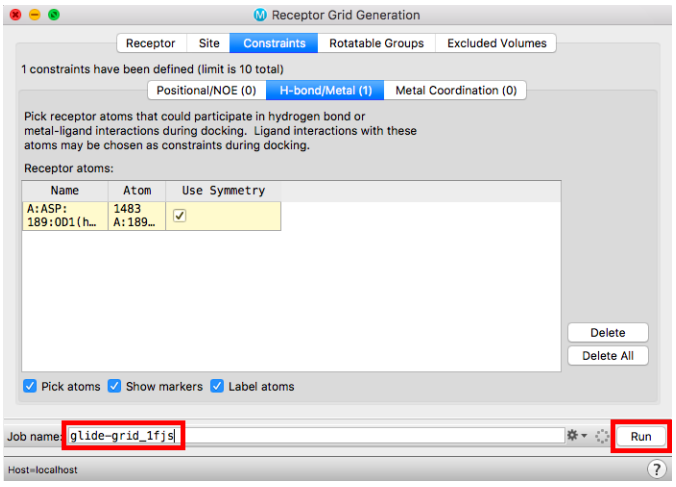


Figure 5-10. Run receptor grid generation job.

9. Change Job name to **glide-grid_1fjs**
10. Click **Run**
    - This job will take about a minute
    - A folder named glide-grid_1fjs is written to your Working Directory

Questions For Comprehension:

6) Go through the same workflow from Section 5 with your chosen PDB.
    a) Choose 3 sizes of the ligand diameter midpoint box to see which grid works best at redocking the crystallized ligand. What sizes did you choose? (Do not just use the same dimensions that were used in the 1FJS example).

    b) Choose 2 different constraints. What constraints did you choose and why? Reference literature to justify your constraints. Below.

6. Docking the Cognate Ligand and Screening a New Compound

The minimum requirements for running a Glide virtual screen are a grid file and a ligand file. It is strongly recommended that the grid file be generated from a protein prepared using the Protein Preparation Wizard and the ligand file be prepared using LigPrep. Additionally, you can choose the scoring function, set ligand- and receptor-based constraints, and define the output. Please see the Glide User Manual for more detail. In this section, we will include the hydrogen bonding constraint that was created in the previous step.

First, we will dock the cognate ligand, which is a helpful way to benchmark a virtual screen of compounds with unknown binding activity against a target. If you have followed on from the Introduction to Structure Preparation and Visualization tutorial, you can begin at section 4.2. The information gained from this step can help with evaluating poses and beneficial interactions, which is useful for hit finding. Second, we will dock the screening compounds from a prepared ligand file, `50ligs_epik.mae.gz`. Both jobs will use the receptor grid file that was generated in the previous step.

6.1 Prepare the cognate ligand (if needed)

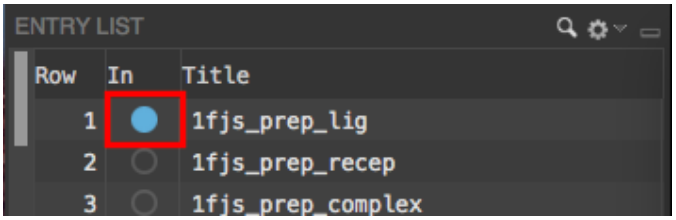


Figure 6-1. Include 1fjs_prep_lig.

1. Include **1fjs_prep_lig** in the Workspace
2. Go to **Tasks > Browse > LigPrep**
    - The LigPrep panel opens

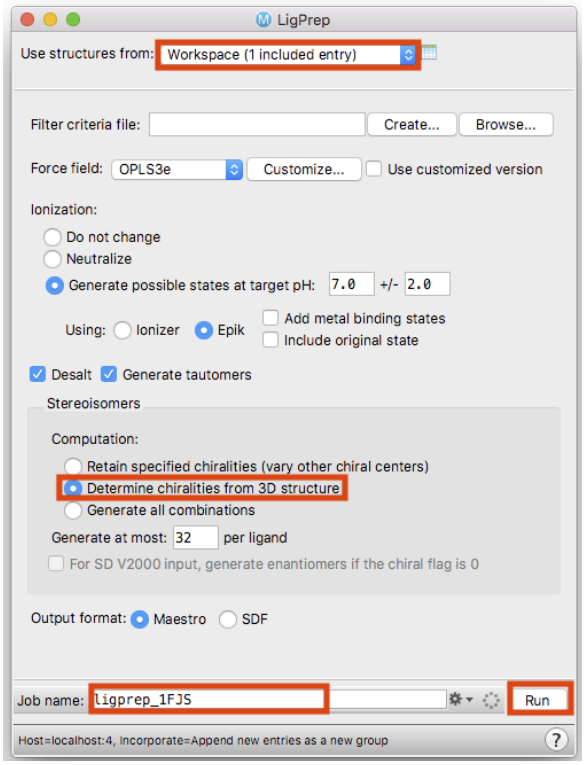


Figure 6-2. The LigPrep panel.

3. For Use structures from, choose **Workspace (1 included entry)**
4. Under Stereoisomers, select **Determine chiralities from 3D structure**
5. Change Job name to **ligprep_1FJS**
6. Click **Run**
    - A banner appears when the job has been incorporated
    - A new group is added to the Entry List

6.2 Dock the cognate ligand

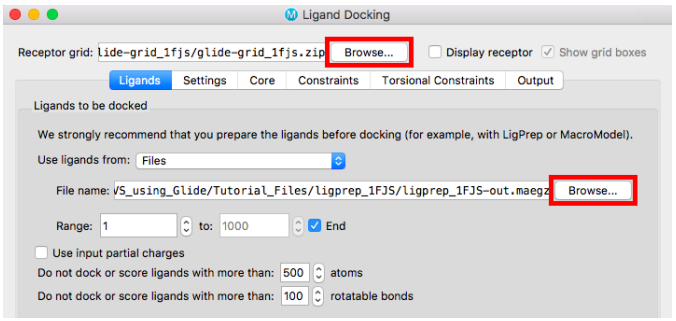


Figure 6-3. The Ligands tab of the Ligand Docking panel.

1. Go to **Tasks > Browse > Receptor-Based Virtual Screening > Ligand Docking**
    - The Ligand Docking panel opens
2. Next to Receptor grid, click **Browse** and choose **`glide-grid_1fjs.zip`**
3. In the Ligands tab, for Use ligands from, choose **Files**
4. Next to File name, click **Browse** and choose **`ligprep_1FJS-out.maegz`**

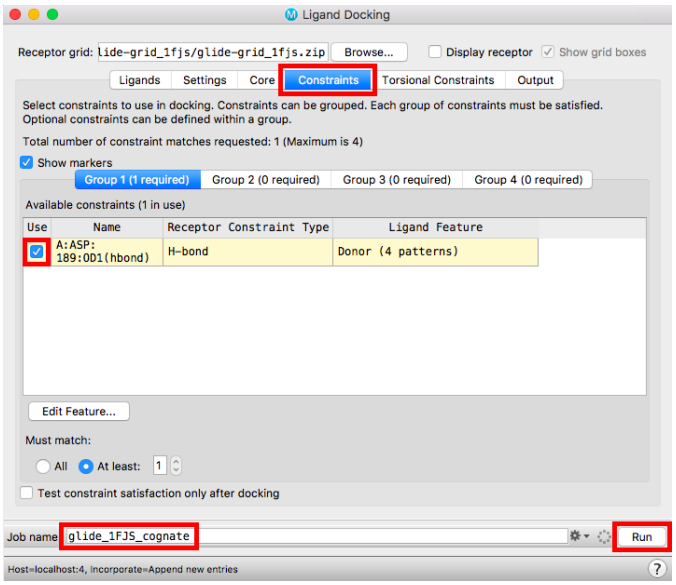


Figure 6-4. The Constraints tab of the Ligand Docking panel.

5. Click the **Constraints** tab
6. Under Use, **check** the H-bond constraint for ASP 189
7. Change Job name to **glide_1FJS_cognate**
8. Click **Run**
    - This job takes about a minute
    - A banner appears to show that files have been incorporated
    - A new group is added to the Entry List

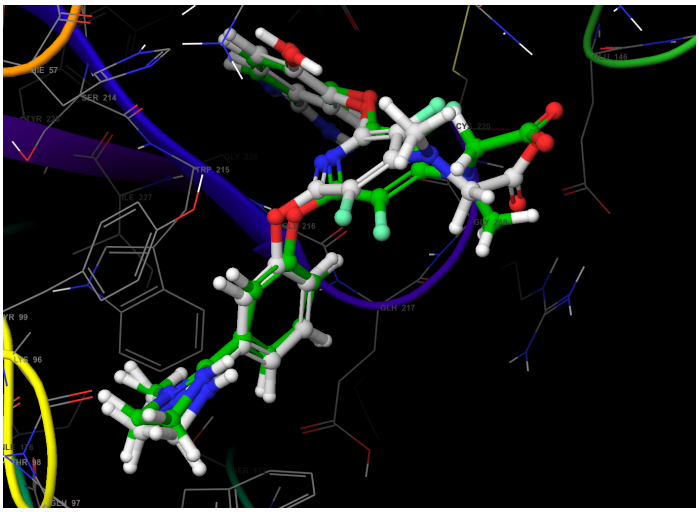


Figure 6-5. Binding pose of the docked cognate ligand (green) compared to the crystal (gray).

9. Double-click the **In** circle next to **1fjs_prep_complex**
    - The entry is fixed in the Workspace
10. Include the **first ligand result** of the glide_1FJS_cognate-pv1 group
11. Include other **ligand results** in turn
    - H-bonds to ASP 189 are conserved
12. Double-click the **In** circle next to **1fjs_prep_complex**
    - The entry is no longer fixed in the Workspace

6.3 Visualize the results using Pose Viewer

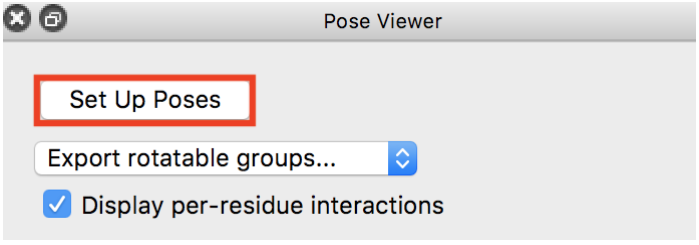


Figure 6-1. Pose Viewer panel.

1. Go to **Tasks > Browse > Receptor-Based Virtual Screening > Pose Viewer**
2. Select newly generated group titled **`glide_1FJS_screening_pv`**
3. Click **Set Up Poses**
4. Check **Display per-residue interactions**
5. Step through the results using the **right** and **left** arrow keys
    - Ligand poses are displayed in the

	Workspace ○ Residues are colored according to their interaction energies, ranging from red (negative, favorable) to blue (positive)

6.2 Analyze the results

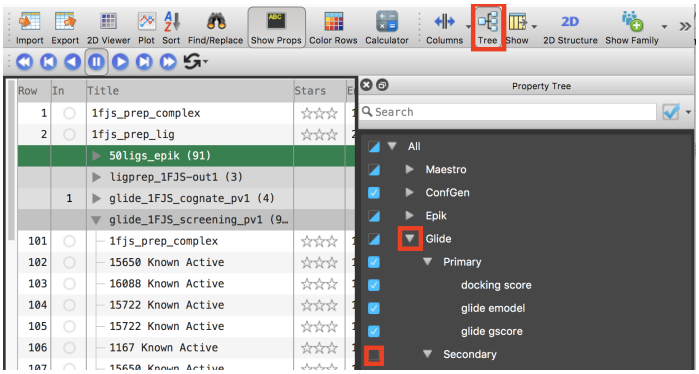


Figure 6-2. Glide Primary properties shown in the Project Table.

1. In the Project Table, click the Property **Tree** icon
   - The Property Tree appears on the right of the Project Table
2. Click the **All** box twice
   - All boxes are deselected
3. Click the **Glide** box
4. Click **Secondary**
   - Only the Glide Primary properties are shown

Note: Please see Knowledge Base Article 1027 for more information on the difference between docking score, Glide gscore, and glide emodel score.

6.3 Visualize pre-docked XP results

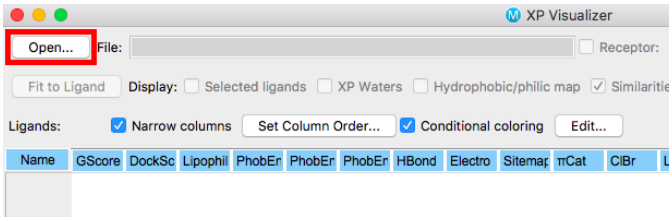


Figure 6-3. The XP Visualizer panel.

1. Go to **Tasks > Browse > Receptor-Based Virtual Screening > Visualize XP Interactions**
   - XP Visualizer opens
2. Click **Open**

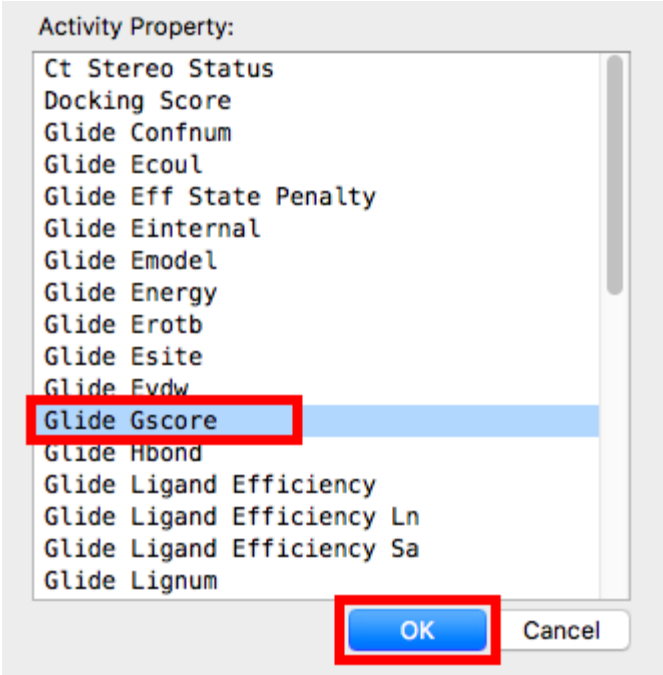


Figure 6-4. Choose the Activity Property.

3. Choose **`factorXa_xp_refine_pv.maegz`** and click **Open**
4. Choose **Glide Gscore** as the activity property, click **OK**
    - The table is populated with the XP results
    - Individual terms of the scoring function are colored as red (penalty) or blue (reward)

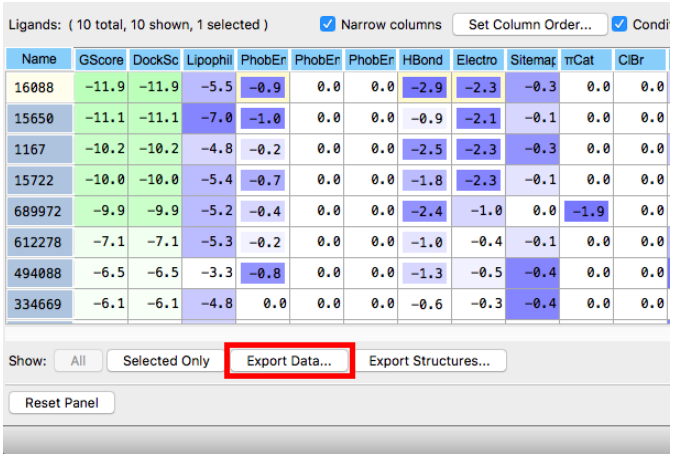


Figure 6-5. The XP Visualizer showing rewards (blue) and penalties (red) to the Glide Gscore.

5. Click **Export Data** to export the spreadsheet as a `.csv` file

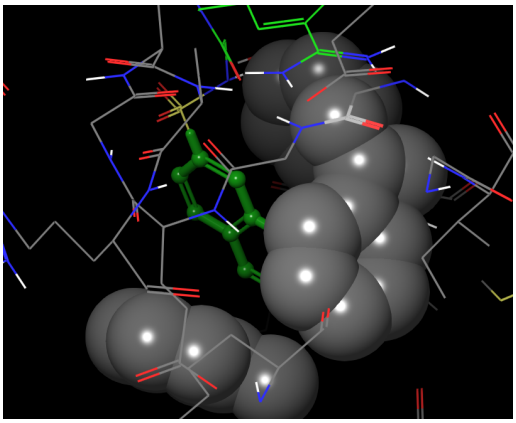


Figure 6-6. Hydrophobic enclosure reward

6. Click on the **indented colored entries** to visualize in the Workspace

shown in the Workspace.	

Questions For Comprehension:

7) Go through the same workflow from Section 6 with your chosen PDB and with the cognate ligand and the ligand you designed on pg 14 (Question 5B).
   a) Fill out the table below:

	Cognate Ligand - Restraint 1	Cognate Ligand - Restraint 2	MyLigand - Restraint 1	MyLigand - Restraint 2
Grid 1	GScore:	GScore:	GScore:	GScore:
	Hbond:	Hbond:	Hbond:	Hbond:
Grid 2	GScore:	GScore:	GScore:	GScore:
	Hbond:	Hbond:	Hbond:	Hbond:
Grid 3	GScore:	GScore:	GScore:	GScore:
	Hbond:	Hbond:	Hbond:	Hbond:

   b) Which Grid resulted in a docking that was best aligned to the crystal structure for the cognate ligand? Is there a way to quantify that alignment?

   c) Which restraint resulted in a docking that was best aligned to the crystal structure for the cognate ligand?

   d) Compare and contrast the docking of MyLigand with the Cognate ligand.

e) **Prepare a Powerpoint presentation** for your class that discusses the results of your docking. Be sure to include at least one slide per section of this tutorial. Finally, in this presentation, outline next steps that would be good to take for further drug discovery of your PDB target.

7. Virtual Screening with Glide

Structure files obtained from the PDB, vendors, and other sources often lack necessary information for performing modeling-related tasks. Typically, these files are missing hydrogens, partial charges, side chains, and/or whole loop regions. In order to make these structures suitable for modeling tasks, we used the Protein Preparation Wizard to resolve issues. Similarly, ligand files can be sourced from numerous places, such as vendors or databases, often in the form of 1D or 2D structures with unstandardized chemistry. LigPrep can convert ligand files to 3D structures, with the chemistry properly standardized and extrapolated, ready for use in virtual screening.

In this section, virtual screening ligands will be prepared. These preparation steps are a necessary part of a virtual screen and must be done before docking.

7.1 Import ligand files

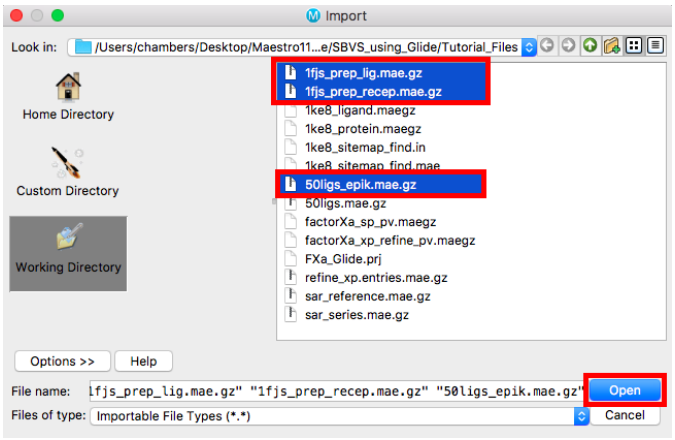


Figure 7-1. The Import panel, with non-contiguous files selected.

1. Go to **File > Import Structures**
2. Ctrl-click (Cmd-click) to select files **`50ligs_epik.mae.gz`**
3. Click **Open**
   - Structures are in the Entry List
   - A banner appears confirming entries have been imported

7.2 Dock the screening compounds

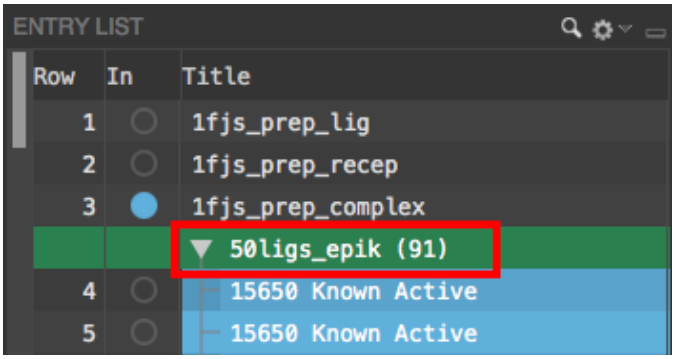


Figure 7-2. Select 50ligs_epik in the Entry List.

4. In the Entry List, select the group **50ligs_epik**

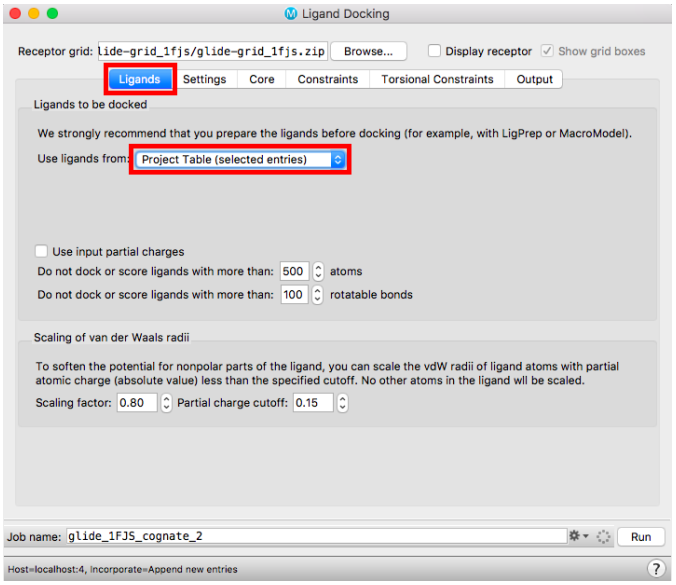


Figure 7-3. Use ligands from selected entries.

5. In the Ligand Docking panel, click the **Ligands** tab
6. For Use ligands from, choose **Project Table (selected entries)**

Note: Keep `glide-grid_1fjs.zip` as the receptor grid

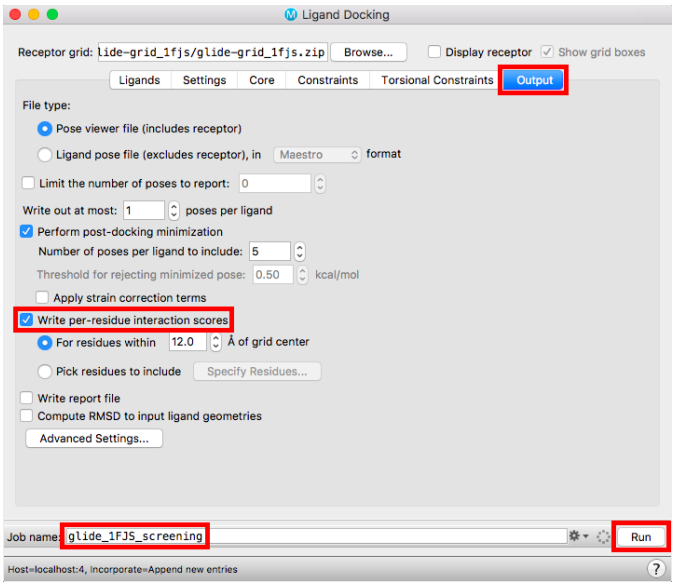


Figure 7-4. The Output tab of the Ligand Docking panel.

7. Click the **Output** tab
8. Check **Write per-residue interaction scores**
9. Change Job name to **glide_1FJS_screening**
10. Click **Run**
    - This job takes a few minutes
    - A banner appears to show that files have been incorporated
    - A new group is added to the Entry List

8) Go through the same workflow from Section 6 with your chosen PDB and answer the following questions.
   a) Did any of the ligands dock into your protein grid? _____
   b) How could you evaluate the quality of these docking models?

      _____
      _____
      _____
      _____
      _____
      _____

   c) Where could you find known actives and inactives against this protein?

      _____
      _____
      _____
      _____
      _____
      _____

8. Conclusion and References

In this tutorial, we imported and prepared a protein and ligand file, then visualized and analyzed the protein-ligand complex. A raw PDB file was made suitable for modeling purposes using the Protein Preparation Wizard, and the cognate ligand was extrapolated using LigPrep in the same fashion that would be used for a multi-ligand file. These steps would be the starting point for many computational experiments, virtual screening with Glide, molecular dynamics simulations (Desmond), and lead optimization (Prime, MM-GBSA). Structures visualization options were able to be chosen manually using the Style toolbox, as well as with one click using Presets. The Workspace Configuration toolbar allowed for toggling various components in the Workspace and the 2D view in the Ligand Interaction Diagram gave another way to analyze information.

For further information, please see:
Maestro 11 Training Portal
Protein Preparation Wizard Panel Help

9. Glossary of Terms

cognate ligand - a ligand that is bound to its protein target

Entry List - a simplified view of the Project Table that allows you to perform basic operations such as selection and inclusion

included - the entry is represented in the Workspace, the circle in the In column is blue

incorporated - once a job is finished, output files from the Working Directory are added to the project and shown in the Entry List and Project Table

Project Table - displays the contents of a project and is also an interface for performing operations on selected entries, viewing properties, and organizing structures and data

Scratch Project - a temporary project in which work is not saved, closing a scratch project removes all current work and begins a new scratch project

selected - (1) the atoms are chosen in the Workspace. These atoms are referred to as "the selection" or "the atom selection". Workspace operations are performed on the selected atoms. (2) The entry is chosen in the Entry List (and Project Table) and the row for the entry is highlighted. Project operations are performed on all selected entries

Working Directory - the location that files are saved

Workspace - the 3D display area in the center of the main window, where molecular structures are displayed