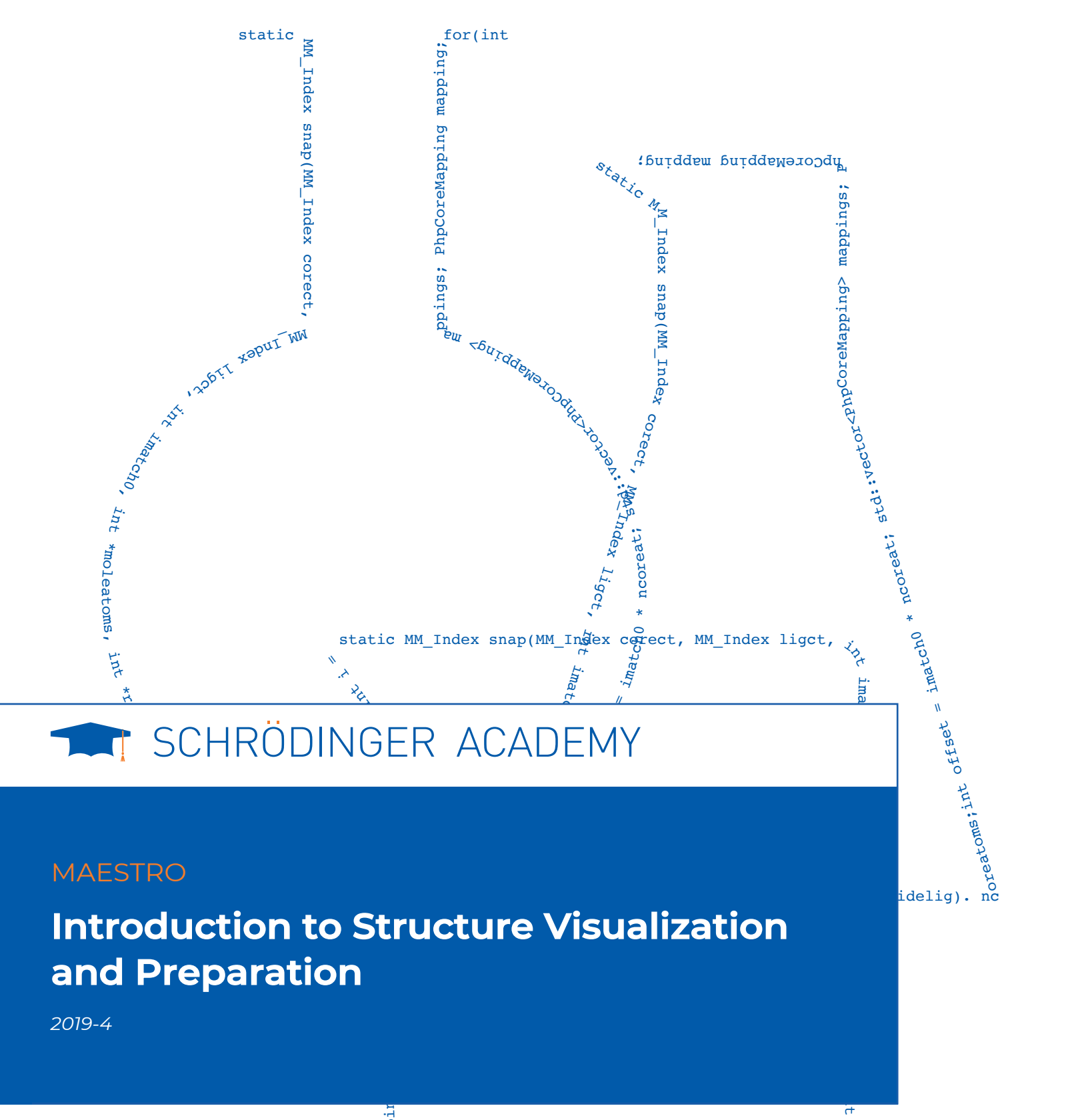


SCHRÖDINGER ACADEMY

MAESTRO

Introduction to Structure Visualization and Preparation

2019-4

Introduction to Structure Preparation and Visualization

Created with:	**Release 2019-4**
Prerequisites:	**Release 2019-2 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, custom set**

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.

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 is written using a 3-button mouse with a scroll wheel.

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)

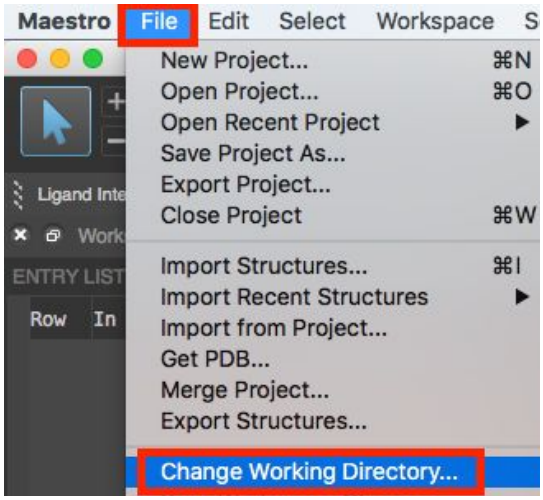


Figure 1-1. Change Working Directory option.

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

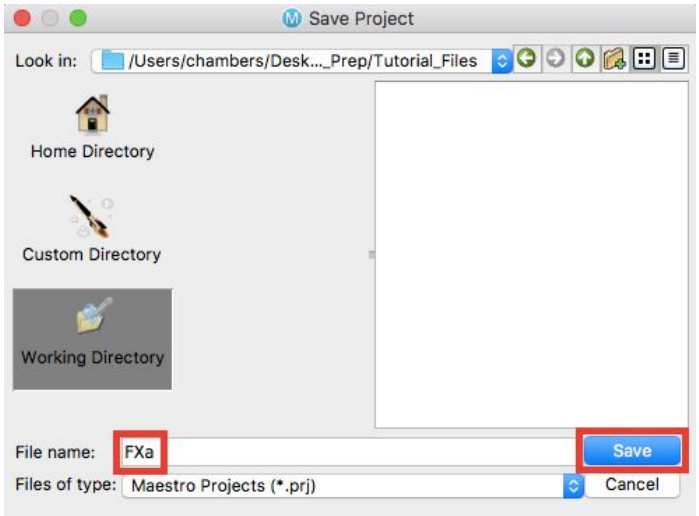


Figure 1-2. Save Project panel.

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

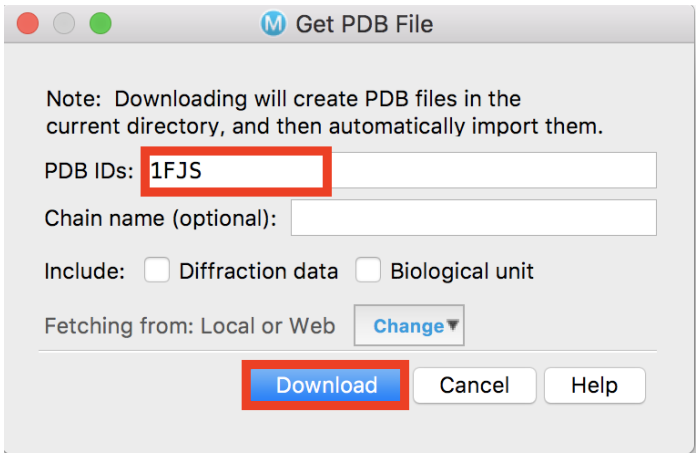


Figure 1-3. Get PDB File panel.

6. Go to **File > Get PDB**
7. For PDB IDs, 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.

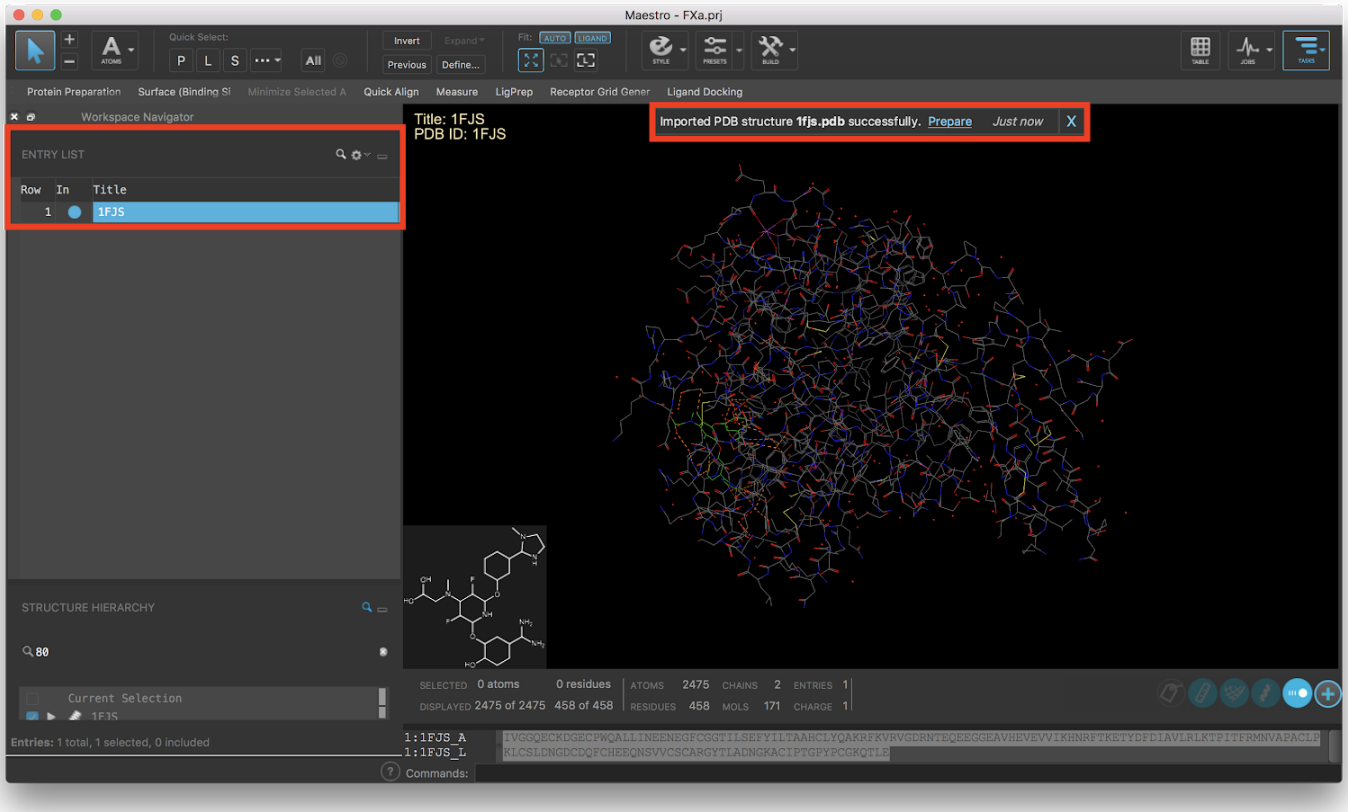


Figure 1-4. The Workspace after the structure is imported from the PDB, with the Entry List and banner highlighted.

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

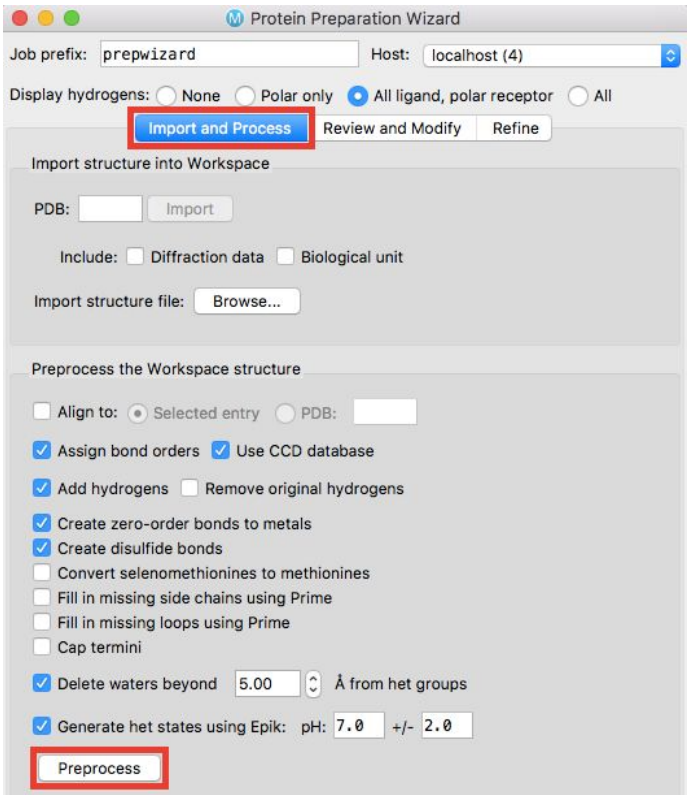


Figure 2-3. Import and Process tab of the Protein Preparation Wizard.

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

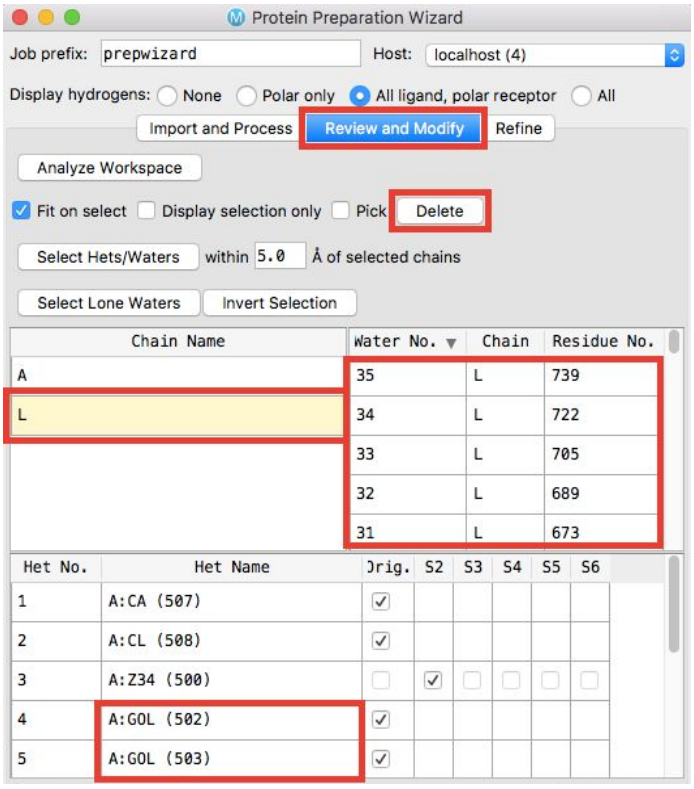
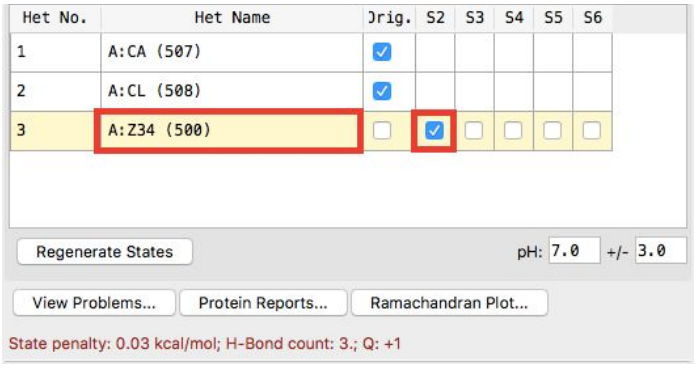


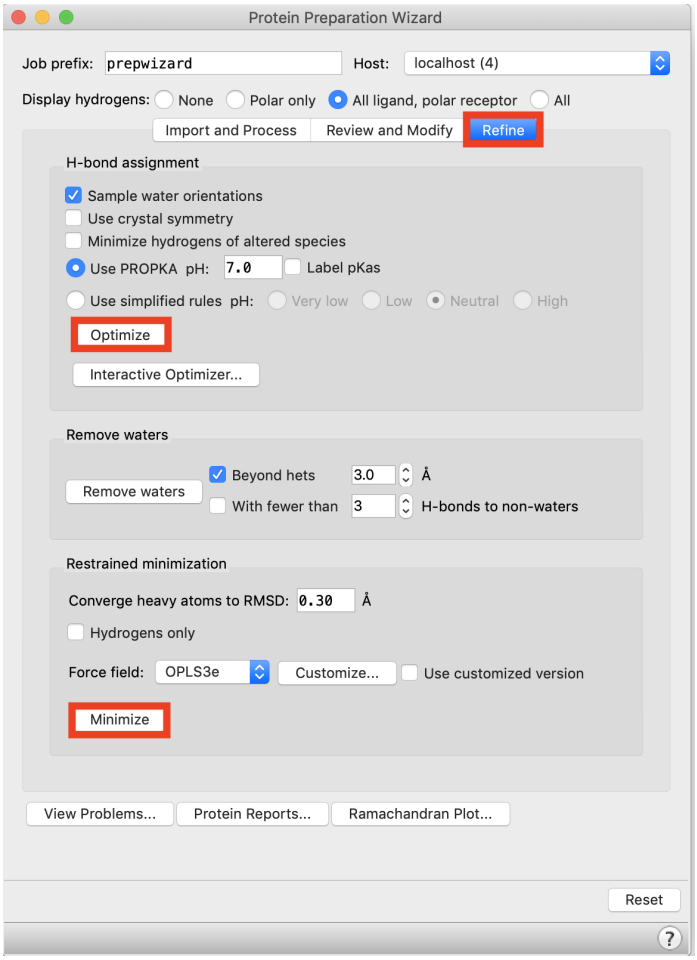
Figure 2-4. The Review and Modify tab before removing unwanted components.

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
    - GOL standard for glycerol, which is a crystallographic artifact that is not biologically relevant
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.
 *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

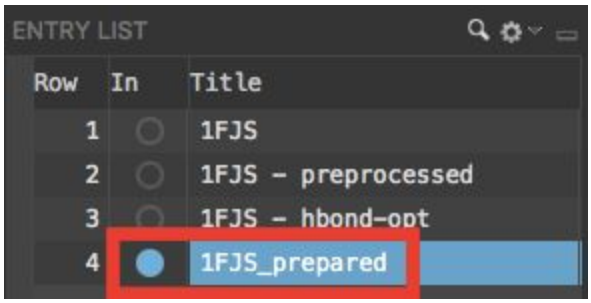


Figure 2-7. Change entry title.

4. In the Entry List, double-click **1FJS - minimized**
5. Type **1FJS_prepared** to rename the entry

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

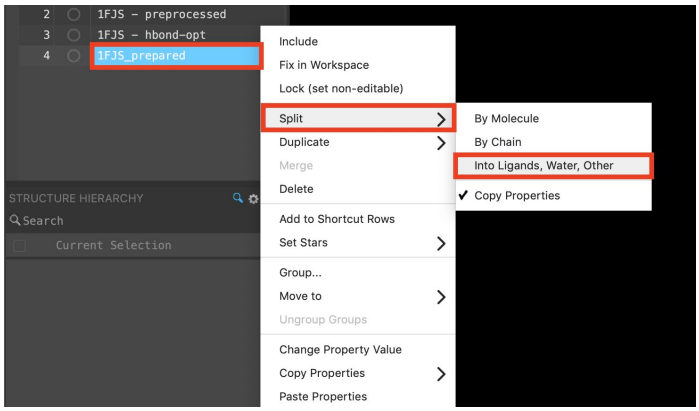


Figure 3-1. Right-click to split an entry into different components.

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

3.2 Run LigPrep

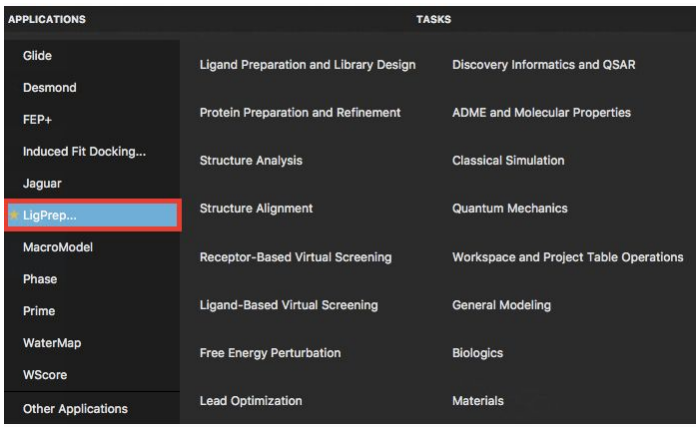


Figure 3-2. LigPrep application in the Task toolbar.

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

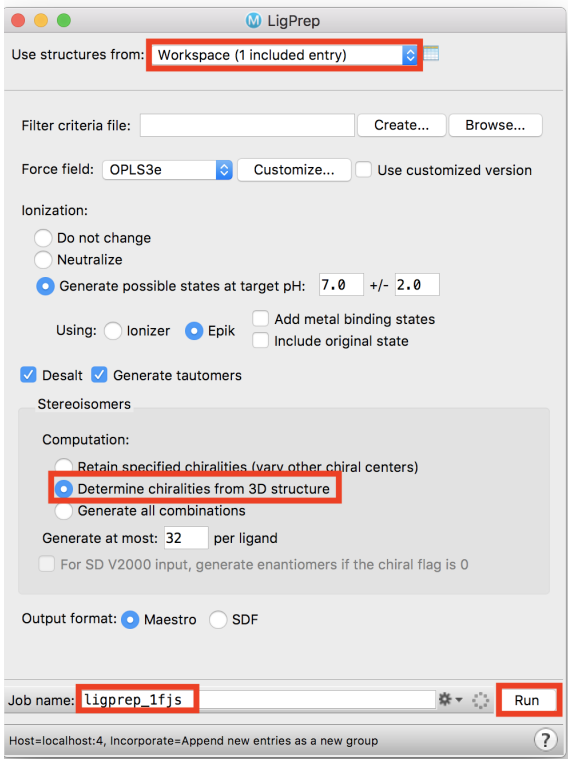


Figure 3-3. The LigPrep panel.

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.

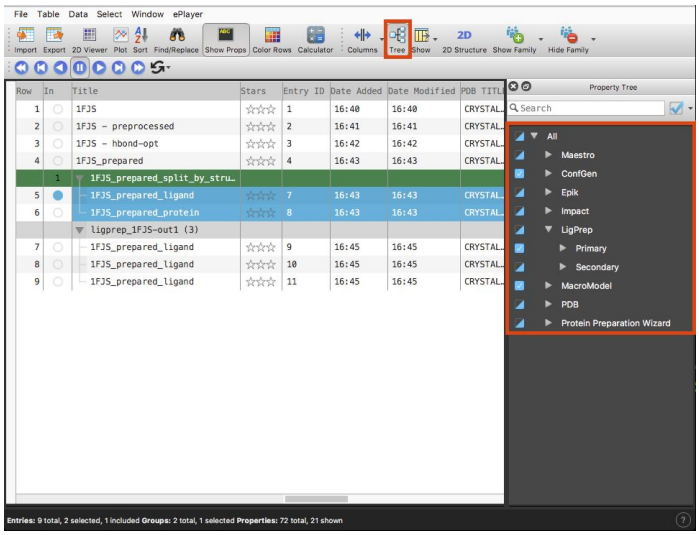


Figure 3-4. The Project Table with the Property Tree open.

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

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, and generate a custom set for some binding residues of interest, Finally, we will visualize the surface of the binding pocket and I 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

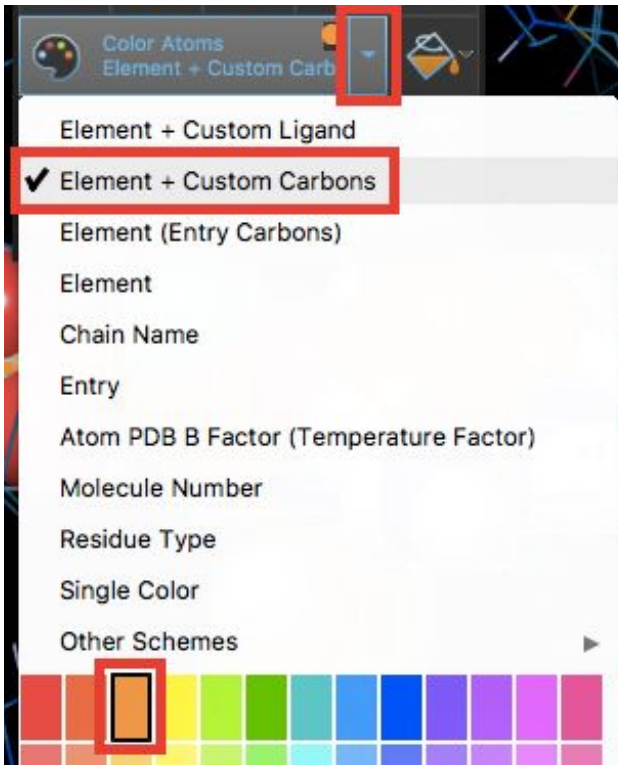


Figure 4-2. The Color Atoms menu.

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

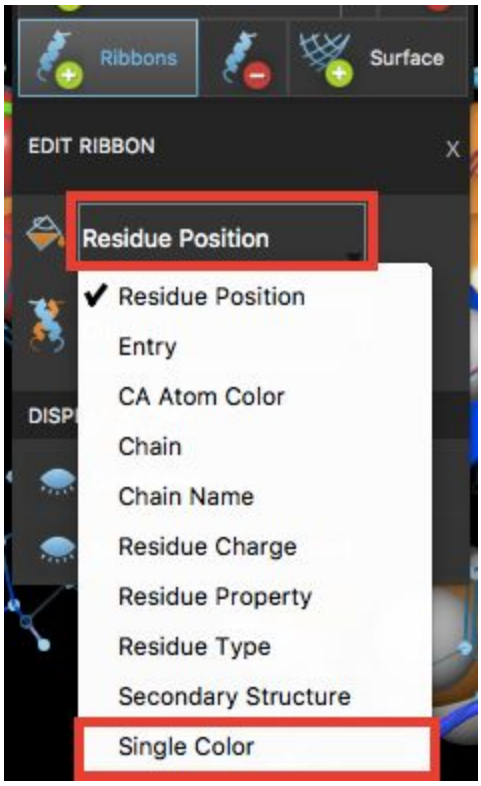


Figure 4-3. The Edit Ribbon panel.

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

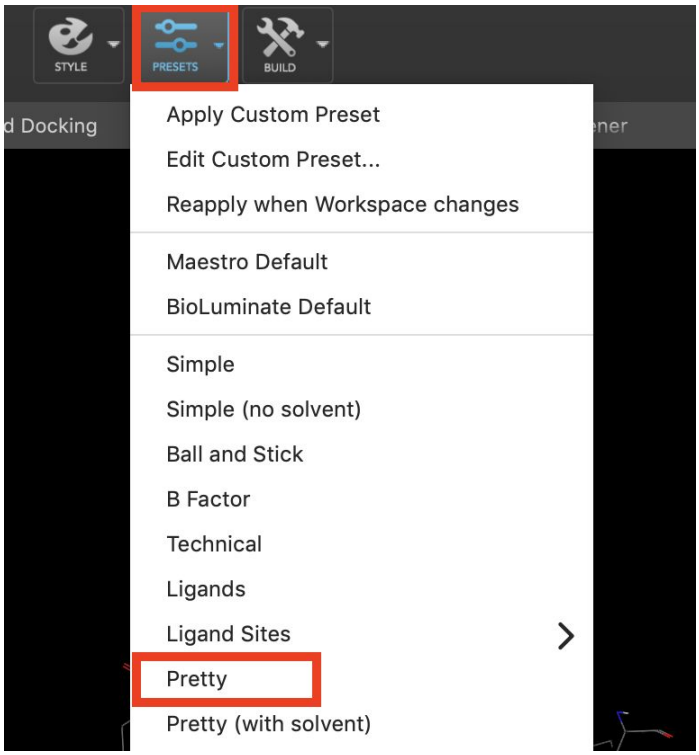


Figure 4-4. The Presets menu.

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

4.3 Visualize Interactions

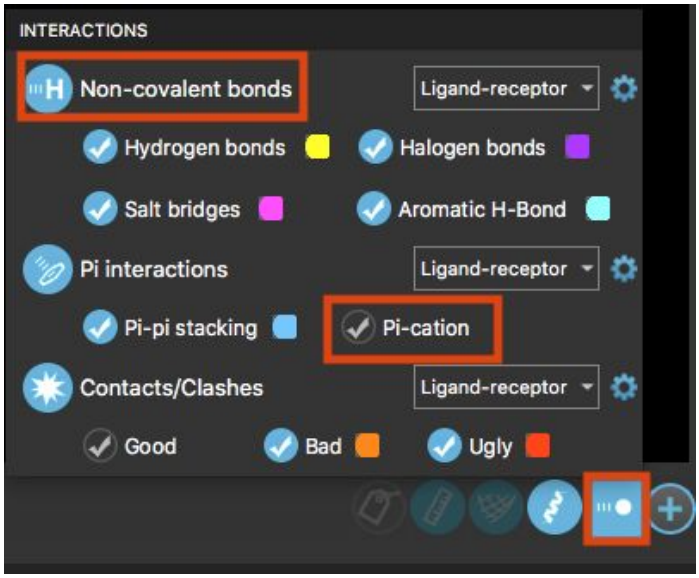


Figure 4-5. The Interactions panel in the Workspace Configuration Toolbar.

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

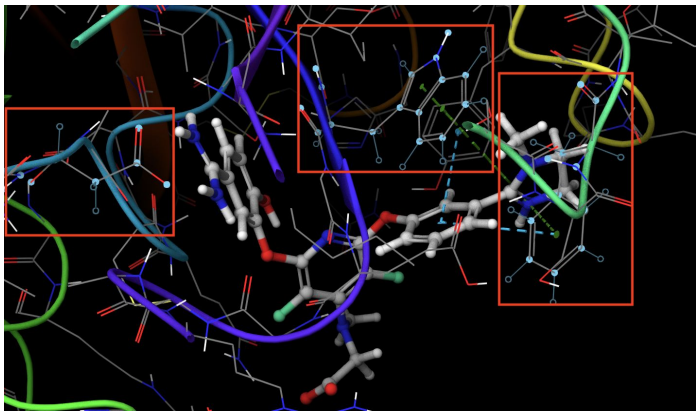


Figure 4-6. Select PHE 174, TRP 215 and ASP 189.

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

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

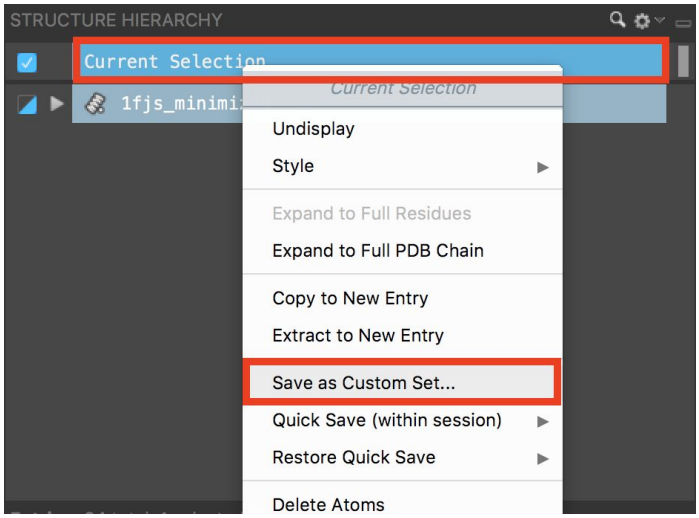


Figure 4-7. Open Save as Custom Set panel.

3. In the Structure Hierarchy, right-click on **Current Selection**
4. Choose **Save as Custom Set**
   - The Save Selection as Custom Set panel opens

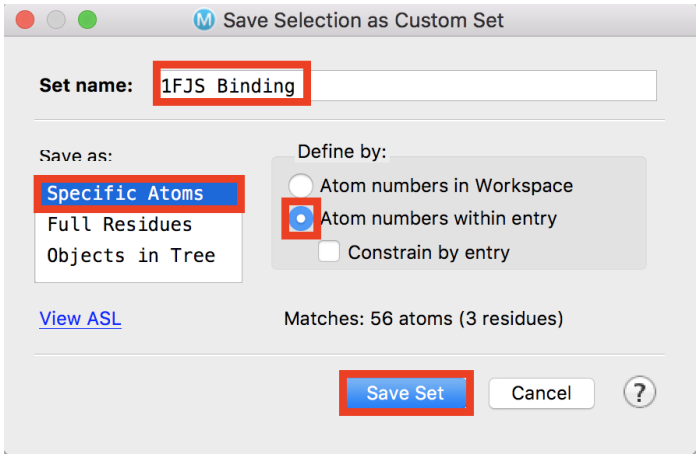


Figure 4-8. Save Custom Set.

5. For Set Name, type **1FJS Binding**
6. For Save as, choose **Specific Atoms**
7. For Define by, choose **Atom numbers within entry**
   - This will ensure that the selection will remain consistent when multiple entries are in the Workspace
8. Click **Save Set**

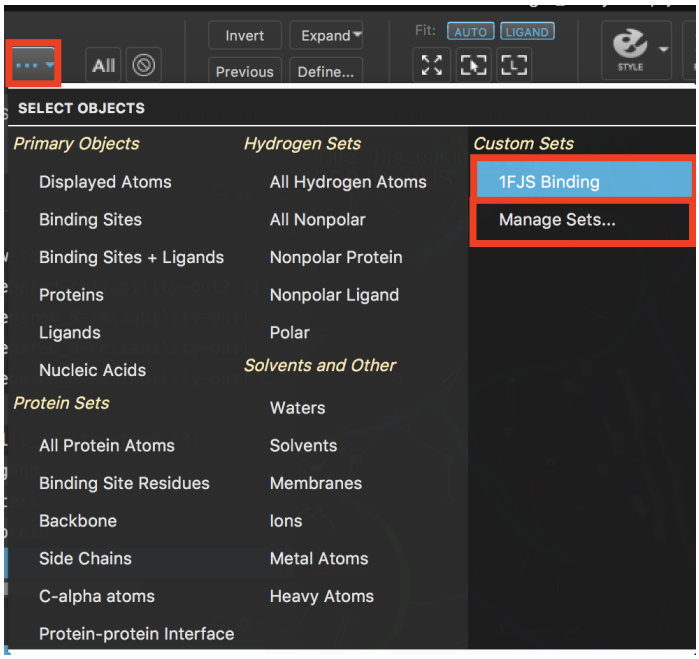


Figure 4-9. Custom Sets section in the Selection Toolbar dropdown.

Note: Custom Sets can be accessed and edited through the Custom Sets section in the dropdown in the Selection Toolbar

4.5 Generate and manipulate a surface

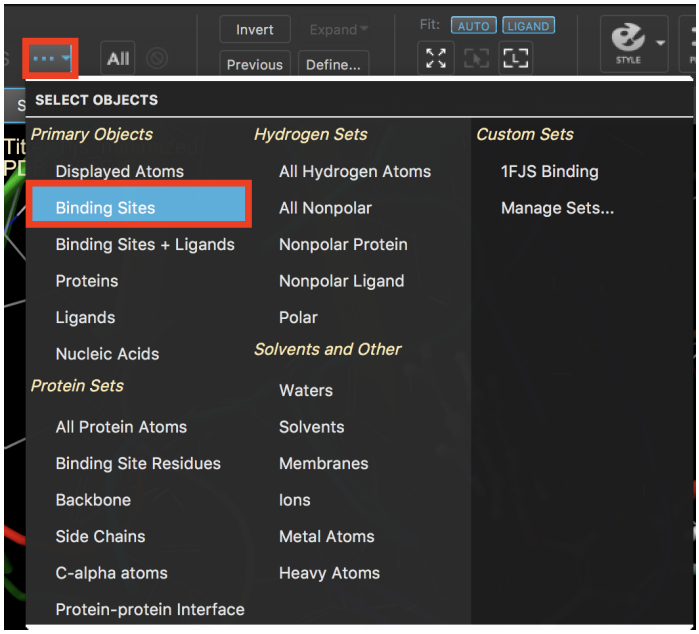


Figure 4-10. More options in Quick Select.

1. Under Quick Select, click **...** and choose **Binding Sites**
2. 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

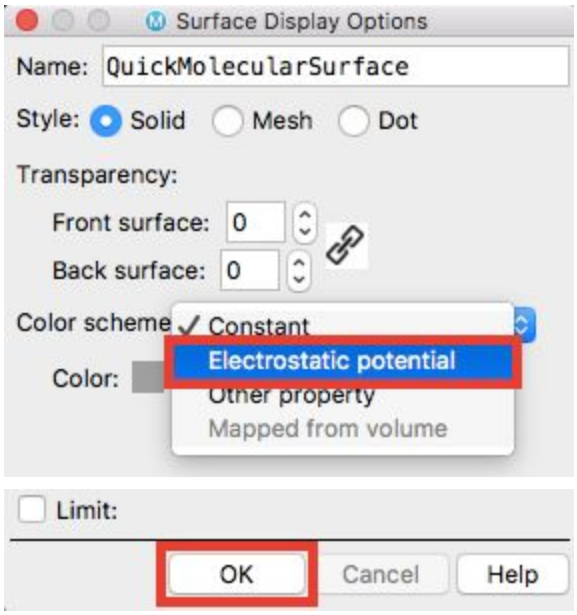


Figure 4-11. The Surface Display Options panel.

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

4.5 Generate a 2D interaction diagram

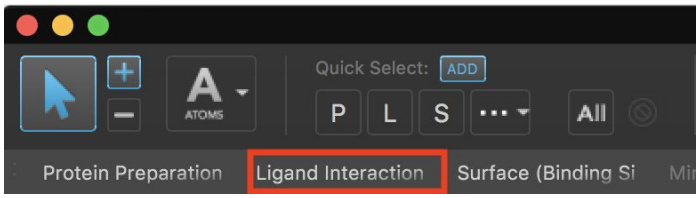


Figure 4-12. Ligand Interaction Diagram in the Favorites toolbar.

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

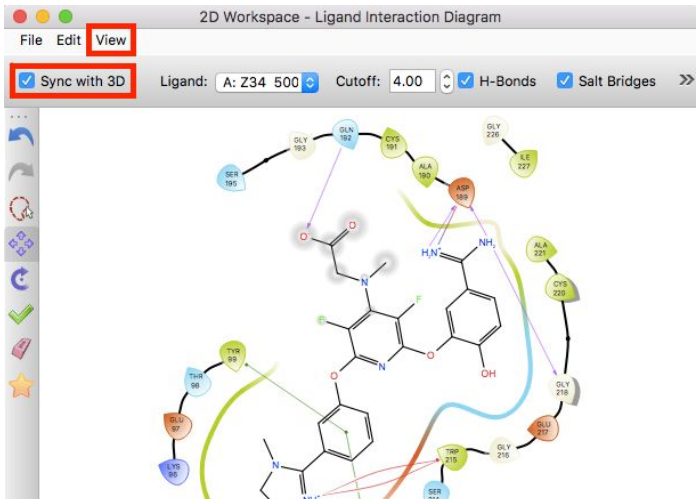


Figure 4-13. The Ligand Interaction Diagram with Sync with 3D turned on and LID legend open.

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

4.6 Save an image of the Workspace

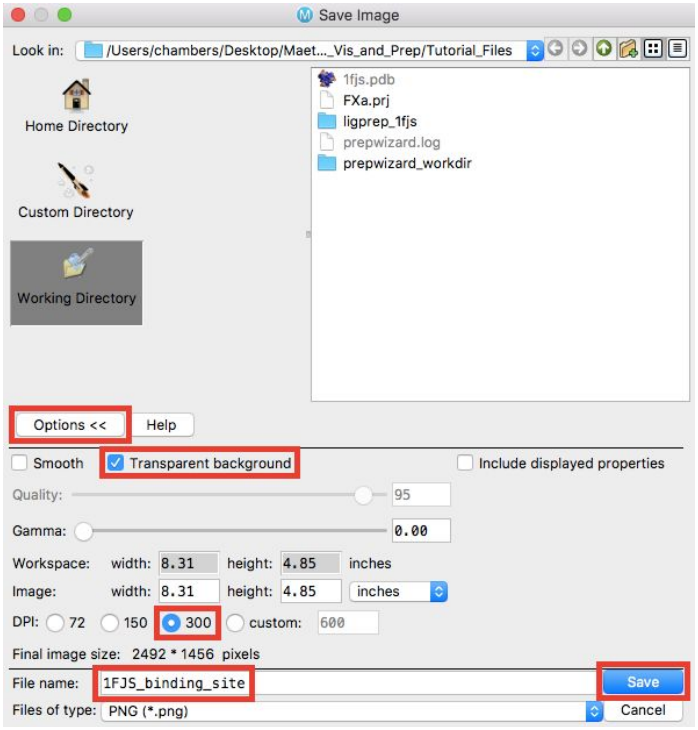


Figure 4-14. Save Image panel with expanded Options shown.

1. Go to **Workspace > Save Image As**
   - The Save Image panel opens
2. Click **Options >>**
3. Check **Transparent background** and select **300** DPI
4. Change File name to **1FJS_binding_site**
5. 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 is saved with the selection highlights

Note: Go to **Tasks > Browse > Workspace and Project Table Operations** for more image options, including **Ray Trace**

5. 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, including docking (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

6. 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