Take-home exercise

Study the interaction of an antifungal drug (fluconazol) with its target enzyme (lanosterol 14α-demethylase) at the atomic level

Key to instructions in this take-home:

- ***M** (means **Ctrl + M**) opens a window (*RasMol command editor*) for typing in a command (you get the same by clicking on "Command" in the "Edit" drop-down menu). Alternatively you can use the "RasMol Command Line" window to type on commands (check "Command Line" in the "Window" drop down menu; you may want to place it below the main window you are working in)
- **"select x"** is a command "x" to be typed in the window
- ***LMB*** or ***RMB*** are left or right mouse botton, respectively, when specified (otherwise use ***LMB***)
- "click on" - use ***LMB***

a. Preparation

1. Download pdb file **1EA1** (see C. above) - fluconazole bound to a bacterial demethylase
2. open it in **RasTop** (as in D. above)
3. under "Edit", click on "Command" or (**Ctrl + M**)
4. type in the window: "select hetero" and click OK
5. on the upper toolbar, click on  (ball&stick)
6. get familiar with the structure (move, enlarge, etc); experiment using the mouse bottons - use "Help" to get guidance

b. Structure analysis

After you completed the Preparation **E.** 1. – 5. above, do the following:

1. ***M "select solvent"**
2. click on  on the upper tool bar (removes the water molecules)

(You can get to this stage by typing "select ligand" instead of "hetero" in **E.** 4. above; no water molecules will show up)

3. enlarge the image holding **Shift + *LMB*** and moving the mouse
4. click on  (center)
5. click on the **Fe** (orange ball in the center of the heme); click on  again close 'center'

When you rotate the molecule, it will rotate around the **Fe** as a center; use the ***RMB*** to position the **Fe** in the middle of the screen, it will keep the molecule in place

6. ***M "select within (3.0,ligand)"** (*no* space after *within!)*
7. click on  (ball&stick)

Observe the new balls - these are the closest atoms of the surrounding aa residues and water oxygens (within 3.0 Angstroms distance from the drug and the heme)
8. ***M "restrict within (3.0,ligand)"** to get rid of the rest of the protein (displayed in wireframe)
9. make sure that in the "Window" drop-down menu the "Main Toolbar", "Selection Toolbar", the "Command Line", the "Command Panel" and the "Status Bar" are all checked
10. If you place the cursor on any atom, you can identify the residue (aa, drug, heme, water) and its number in the "Res" window of the status bar on the bottom. **Write down the names (3 letters) and numbers for every aa residue, water (HOH), and ligand!**
11. click on  (color picker), then click on  (atoms) and then click on each of the water molecules you identified, followed by clicking on bright green on the color palette to make the water molecules look different
12. click on  (residues)
13. click on all the new atoms (single balls) with the cursor (exept the waters), one at a time, followed by clicking on  (ball&stick) each time; this way the rest of the structures of the aa residues will be added on

Now you have the **heme (HEM 460)**, the **drug (TPF 470)** and the closest (within 3 A) amino acid residues and water molecules on the screen, and ready to work on the take-home exam below.