Display of the charged surface of a protein

Purpose: Learn multiple ways to display the surface charge on a protein structure. Pay attention to the advantages and disadvantages of the different methods.

1. What this is not:

Many molecular visualization tools can easily generate a surface representation of your molecule, typically a "solvent-excluded volume". They can frequently color that surface according to the charged residues under it, usually red for negative and blue for positive.

1. Pymol - Use the small buttons in the upper right-hand corner,
   - **all->show->as->surface**
   - With one click, you have the charges represented as you expect, plus hydrophobic surface represented in green.
   - To make a particularly pretty surface display, look here.
2. Chimera - **Tools->Structure Analysis->ResProp** brings up a window in which you can choose:
   - `Color Surface`
   - `Residue Schema = Positive` and `Chosen side chains = blue`
   - `Remaining side chains` to `white` or `No color` (apply), then
   - `Residue Schema = Negative` and `Chosen side chains = red`
     This is simple, and gives you the option of adding additional colorings easily (e.g., you could color perturbed residues yellow).

Important Note: These are over-simplified views of what residues are under the surface. This does not represent the actual charge field in any accurate way. For that, you have to calculate the electrostatics properly. See below.

2. Grasp

This is one of the canonical methods of generating an accurate electrostatic potential for a macromolecule. (along with Delphi, MEAD, Zap, etc.) The user interface takes some work getting used to. **NOTE** It is available **only** on SGI computers, and so we will not be using it for today's workshop. It is mentioned here only for illustration. To learn more about it, visit the Honig lab webpage: http://wiki.c2b2.columbia.edu/honiglab_public/index.php/Software:GRASP

[See also Sybyl, part of the Tripos package, for an integrated commercial tool.]

3. Pymol

There are two ways to do it in Pymol. You can try both or pick one:

1. **(molecule)->A(ctions)->generate->vacuum electrostatics->protein contact potential (local)**
   This has a disclaimer about poor accuracy... but it's a quick first approximation.
2. The more thorough way to do it is to use the menu item:
   Plugin->APBS tools (Adpative Poisson-Boltzmann Solver)
   Click "Set grid", then "Run APBS". Wait for the run to finish. ("ObjectMap: Map Read.")
3. Finally, click on the Visualization tool, update, and click on **Show Molecular Surface**. You may want to select "Solvent accessible surface" and "color by potential on sol. acc. surf." and increase the color ramp range by adjusting the high and low setpoints.

APBS does a more rigorous calculation using correct dielectrics for the protein and solvent, etc. However, it can run into some difficulties if there are e.g. non-standard or missing residues in your pdb file. You may have to edit your pdbfile to get it to work smoothly. If that's not feasible, you can go back to GRASP, which complains less about defective structures, or:

4. APBS standalone + Chimera

In cases where the PyMOL plugin doesn't work well, you may want to have more control- for instance, the ability to manually edit the intermediate files. The following procedure will allow you to do that:

1. Generate input files for APBS by visiting the PDB2PQR server:
    1. Go to this website: http://nbcr-222.ucsd.edu/pdb2pqr_1.9.0/
    2. Upload your pdb file, use whatever forcefield you want (default should be ok), internal naming scheme, check Ensure that new atoms are not rebuilt too close to existing atoms, check Optimize the hydrogen bonding network, check Create an APBS input file.
    3. Click submit. After a few seconds or minutes it'll give you the .pqr and apbs .in files. You should inspect these with a text editor (not a word processor) and customize them if necessary.
    4. Download these into the same directory and then run `apbs file.in` (either /sb/apps/Linux/bin/apbs or /programs/l/apbs/1.2.1/bin/apbs)
    5. In case you don't have the apbs software available, example files have been provided. Look in the sample_files/ directory for files named 1hne.*
2. Display in Chimera
    1. After you have the pot.dx file just load your pdb into chimera, make a surface
    2. then go into **tools -> surface/binding analysis -> electrostatic surface coloring** , load the .dx file and then click color.
        1. The usual way to display is to use three colors red, white, blue with -10, 0, 10 or -15, 0, 15 kT/q