Bioinformatics 2

Exercise 2--
make a small protein from scratch
due Feb 8
We’re going to make a small protein whose sequence is all alanines, 24 A’s to be exact. We will then fold the protein into a classic beta-alpha-beta unit. Be sure that you make the unit right-handed.

We will learn and use the following MOE tools. If you need additional help with any tool, use the MOE tutorials.

Step by step instruction follow. However, your results may vary.
Protein Builder

• Edit-->Build-->Protein
  – select “extended” and click ALA 8 times.
  – You now have a 8-residue beta strand.
  – select “helix”. Click ALA 8 more times.
  – select “extended” again and hit ALA 8 more times.
  – Now you have a beta-alpha-beta unit, but it is not “folded”.
Freeze the helix ... for the duration of this exercise

- We will fold the two strands around the helix, keeping the helix fixed.
- `cntl-q` (goes to Sequence Editor)
- Display-->single letter residues
- Display-->Actual secondary structure
- Select the 8 residues of helix (under red bar)
- Turn on “Synchronize” (Selection-->Synchronize)
- In Moe window: Edit-->Potential-->Fix
  - Fixed atoms cannot be moved, neither by using the meta-key nor by Minimizing.
- Select-->invert
  - Selects the un-selected. Un-selects the selected.
- Edit-->Potential-->Unfix
  - Makes sure the rest of the molecule can move, in case you have done any Fixing on your own.
Fold the strands to make a beta sheet

• In Sequence editor: select residues 1-8
• In Moe window: Use meta-key to rotate the strand parallel to the helix axis.
• In Sequence editor: select residues 17-24
• In Moe window: Use meta-key to rotate the strand parallel to the helix axis. Make it a right-handed $\beta\alpha\beta$ unit.
• Try to rotate the strands so the the sidechains point in/out, and the oxygens/nitrogens point to each other, as they will when they make H-bonds.
• Start GizMOE-->Minimizer
  – Watch the stretched bonds relax to their ideal lengths.

In this view, strand 1 is on the right, connected to the helix (behind) at the top. This is right-handed.
Use restraints to make H-bonds

- Edit-->Potential-->Restrain
  - Restrain menu bar appears. Asks you to “pick first atom”, but wait.
  - Select “distance”. Set target distances to 1.8 and 1.8. These are the minimum and maximum allowed distances. We want perfect H-bonds, so we want the distance from hydrogen to oxygen to be 1.8Å
  - Set weight to 100
  - Select O of residue 1
    - the Restrain window now says “pick second atom”
  - Select HN of residue 19
    - This is the hydrogen attached to the backbone nitrogen.
    - the Restrain window now says “Press create”
  - Press create
    - Watch the red distance monitor go to 1.8Å (if Minimizer is turned on)
Make the rest of the beta-sheet H-bonds

- Keep the same Restrain settings and continue selecting first and second atoms and pressing create. We are making a regular, parallel beta-sheet H-bonding pattern.
- First atom (O)  Second atom (HN)
  1  19
  19 3
  3 21
  21 5
- Cancel-->Restrain
- Now select blocks of atoms, pull and push gently (using the meta-key) to help the sheet relax into place. The selected atoms relax only after they are unselected.
To remove a restraint

- If you clicked on the wrong atoms and want to remove the restraint.
- Edit-->Potential setup...
  - A window appears. Select Restraints. Find the one you want to delete by looking at the atom names. Select. Delete.
A beta-sheet has its sidechains alternating sides, up-down-up-down, when viewed from the side. To make a canonical parallel beta sheet:

• Drag the sidechains of 2, 4, 19, and 21 to the outside of the sheet (if they are not there already).
• Drag the sidechains of 1, 3, 5, 18, 20 and 22 to the inside of the sheet (the side that packs against the helix).
• Let the Minimizer relax them into place as you move them.
Knobs in holes packing

- Select the sidechains
  - Selection --> Protein --> sidechain atoms
- and render them as
  - Render --> spacefilling
    - so you can see them clearly.
- *Push* and *pull* atoms so that sidechain (knobs) fill the spaces between sidechains (holes).

Helix sidechains (green) pack in the spaces between strand sidechains.
Save .moe file

Make sidechains ball-and-stick.
Add a cartoon backbone.
Save as a moe file.
   File-->Save
Upload it to

http://www.bioinfo.rpi.edu/~bystrc/courses/biol4550/homework.html