Instructions for Using Molecular Models

Aim: To understand that protein structure can impact protein function, using the bioinformatics tool Molecule World to visualize molecules.

Instructions: Write the answers to your questions on Student Handout—Using Molecular Models Worksheet, in your lab notebook, or on a separate sheet of paper, as instructed by your teacher.

PART I: Viewing DNA Structure

1. Open the Molecule World iPad app.

2. Tap the list icon in the top left-hand corner to view the list of pre-loaded structures.

3. If your structures have already been loaded, locate 1NAJ in the list of structures and touch the title to view it in the structure window.

If 1NAJ is not in the list, tap the + icon to open the search menu. Select the MMDB database, type “1NAJ” in the search area, and touch the Search button to locate the structure in the NCBI’s Molecular Modeling Database. Touch the title of the structure when it appears to view it in the structure window.

We will begin our investigation of 3D structure by looking at a molecule you are already familiar with: DNA. “1NAJ” is the accession number for a file that contains structure information for a small piece of double-stranded DNA. An accession number is like a catalog number or bar code; it bears no resemblance to the product itself, but allows you to access information in databases such as the MMDB, PDB, or PubChem.

4. View 1NAJ, a DNA structure. Drag the structure with one finger to turn it around.

5. Drag the structure with two fingers to move the structure without turning it.

6. Pinch your fingers together to make the structure smaller or spread them apart to make the structure larger.

7. Experiment with controlling the movement and viewing the DNA molecule from multiple angles.

8. Touch the molecule icon to view possible drawing and coloring styles.

Explore each of the options for drawing the DNA molecule.

Ball & stick                   Spacefill                      Tubes

Figure 1: Touch the title of the structure to download and view the file.
Imagine you are teaching a class about DNA. Which drawing styles would you use to teach students about DNA structure? Explain the reasons for your decision.

9. Touch the molecule icon again to view different visualization styles. There are many different styles for coloring molecules. Try each of the coloring styles listed below. Answer each of the following questions.

   a. **Charge**: How did the charge coloring style change the color of the DNA? Use the palette icon to view the charge color key. What does the color key tell you about the way DNA is charged?

   b. **Element**: How did the element coloring style change the color of the DNA? Use the palette icon to view the element color key. How many elements are in DNA? What elements are in DNA?

   c. **Rainbow**: ‘Rainbow’ uses the color red at the start (5’ end) and continues through the rainbow. Why are two regions in the DNA colored red?

   d. Explore a few combinations of drawing and coloring styles. Which coloring options do you find most useful? Why?

10. Touch the “Show sequence” button to open the **Sequence viewer**.

Touch a letter in the DNA sequence to see what happens. Each letter represents a base in the DNA.

Note that there are two DNA sequences: *1NAJ-A* and *1NAJ-B*. Each sequence corresponds to a different strand of the DNA.

![Sequence viewer](image)

*Figure 2:* The Sequence viewer shows the sequences for nucleic acids or proteins.

Credit: Digital World Biology, 2015

11. Touch the molecule icon and the “Reset Appearance” button to restore the DNA to its original state.

12. In the sequence *1NAJ-A*, touch the first “G” to select it. Next, touch the first “G” in sequence *1NAJ-B*.

Where are these two guanines located in the DNA molecule? Explain why.

13. Touch the molecule icon. Select the “Molecule” coloring style. Note the correspondence between the colors in the sequence and the colors in the structure.
a. What did changing the coloring style to Molecule do? Explain. How many molecules are in DNA?

b. Move the DNA so that the ring structure(s) of the selected nucleotides bases can be seen clearly. What do you see?

14. Open the Selection menu in the bottom right-hand corner. Choose “Clear Selection.”

15. Touch any base in either DNA sequence to select it.

16. Open the Show/Hide menu and choose “Hide Unselected Residues.” Answer the following questions:
   a. What do you see? Zoom in to view the base more easily.
   b. Challenge question: Is the base you selected a purine or a pyrimidine, and how do you know?
   c. In the Sequence viewer, touch the name of the chain that contains your base to see the complete strand.

PART II: Viewing the BRCA1 Protein

Now that you are familiar with Molecule World, we will view part of the BRCA1 protein and part of a second protein that it interacts with.

17. If your structures have been pre-loaded, locate 1Y98 in the structure list. Touch the title to view the structure.

If the 1Y98 structure is not in the list, tap the + icon to open the search menu. Select MMDB as the database, type “1Y98” in the search area, and click the Search button to locate the structure. Touch the title of the structure after it appears to download the structure and view it in the structure window.

18. Turn the structure around to identify regions where a single chain shows multiple loops (alpha helices).

19. Locate regions where chains are organized in parallel lines (beta sheets). Both alpha helices and beta sheets are examples of secondary structure.

   Draw an alpha helix and a beta sheet.

20. The structure contains two BRCT domains. To see the domains more clearly:

   A. Touch the molecule icon and choose the “Rainbow” coloring style.
   B. Touch the Show sequence button to view the amino acid sequence.
   C. Then, touch 1Y98-A (the name of the top protein chain) to select it.
   D. Touch the Show/Hide button and choose “Hide unselected” to hide other objects.

The rainbow coloring style will help you visualize the two BRCT domains. BRCT stands for Breast cancer C-Terminal domain. This domain is involved in protein-protein interactions. One domain will be colored in red, yellow, and green. The other will be colored in shades of blue and purple. If you turn the structure, you’ll see a space between the two domains.
21. Touch the molecule icon and choose “Reset appearance.”

22. Open the Sequence viewer and touch the name of the second protein chain, “1Y98-B,” to highlight the CtIP protein. This protein interacts with BRCA1 when damaged DNA is repaired. A phosphate group has been added to this protein.

23. Touch the chain name, 1Y98-B, again to deselect it.

24. Scroll through the amino acid sequences and look for the letter “M” in the top chain, “1Y98-A.” M stands for the amino acid methionine. Touch the M to select it and look at the PDB position to see where this amino acid would be located in the complete protein chain.

25. Touch the letter M again to deselect it.
26. Scroll through the sequence, selecting and deselecting M’s, until you find the M at PDB position 1775.

Amino acid position PDB 1775 is the location of the M1775R mutation in BRCA1 that affects the Lawler family. When the M (methionine) at PDB position “1775” has been selected, it will appear slightly brighter in the protein structure. The structure you are looking at does not have the mutation. (Recall that the mutation converts M to R.)

27. Touch the Show/Hide button and choose “All atoms in residue” to see the entire side chain.

28. Touch the molecule icon and choose “Ball & stick” to see the methionine more clearly.

29. Touch the Selection button and choose “Select nearby” to select other molecules within a radius of six Angstroms. [Note: An Angstrom, Å, is 1x10⁻¹⁰ meters or 0.1 nanometer.]

30. Touch the Show/Hide button and choose “All atoms in residue,” then touch the molecule icon again and choose Ball & stick.

Notice how close the side chains in the CtIP residues are to the methionine at the mutation site in BRCA1. You can imagine how a change in BRCA1 might impact its ability to interact with the CtIP protein which is also required for DNA repair.

31. Record your results by capturing an image of the interaction site between the BRCA1 and CtIP proteins:
   a. Touch the camera icon and choose “Save image” to save an image of the two proteins in Photos.
   b. Open a photo annotation program like Skitch and add an arrow or a circle to show the mutation site. Alternatively, you can add circles later in Word.
   c. Open the Word® document you created in Lesson Four. It should be labeled with your LASTNAME_BRCA1_NCBI.
   d. Insert the labeled image into your document.
   e. Type these instructions and questions at the bottom of your Word® document:
      1. Circle the location of the mutation in the picture of the BLAST alignment.
      2. Explain what the picture of the protein structure and the picture of the alignment represent.
      3. Explain how the pictures are connected to each other.
   f. Follow the instructions you typed above (1-3) and answer the questions in your Word® document. If you haven’t added arrows or circles to identify the mutation site already, you can use the drawing tools in Word® to draw circles around the mutation in the BLAST sequence window and the protein image.
   g. Save and close this document.

32. Challenge Question: What can you see now that you could not see before you annotated this structure? Does this help you understand the consequences of the Lawlers’ M1775R mutation?

33. Optional: If time permits, you may wish to experiment with protein structures 1JNX and 1N5O. Make sure that you enter capital O and not a zero for “1N5O.”

1JNX: This is the accession number for the BRCT domains of the non-mutated version of the BRCA1 protein.

1N5O: This is the accession number for the BRCT domains of the M1775R mutation of the BRCA1 protein.