Tasks on the Computer:

Many proteins interact with other proteins, triggering series of events called pathways. In cases involved in disease we often want to stop a certain pathway from taking place — thus we look for steps along the way that can be inhibited.

With this exercise you will cement the concepts on the previous exercise, and add knowledge about peptide molecules as drug precursors and drug candidates.

We will focus on the MDM2 interaction with p53, and the homologous interaction MDMX with p53. P53 is the guardian of the genome, protecting us from incorrect cellular behavior by triggering apoptosis. In many cancers, p53 is not available to perform its duties. One of the pathways that elliminates p53 from our cells is through its interaction with MDM2 and MDMX, which mark p53 for degradation. Pharma has been interested for decades in the design of dual inhibitors of the MDM2/p53 and MDMX/p53 interactions.

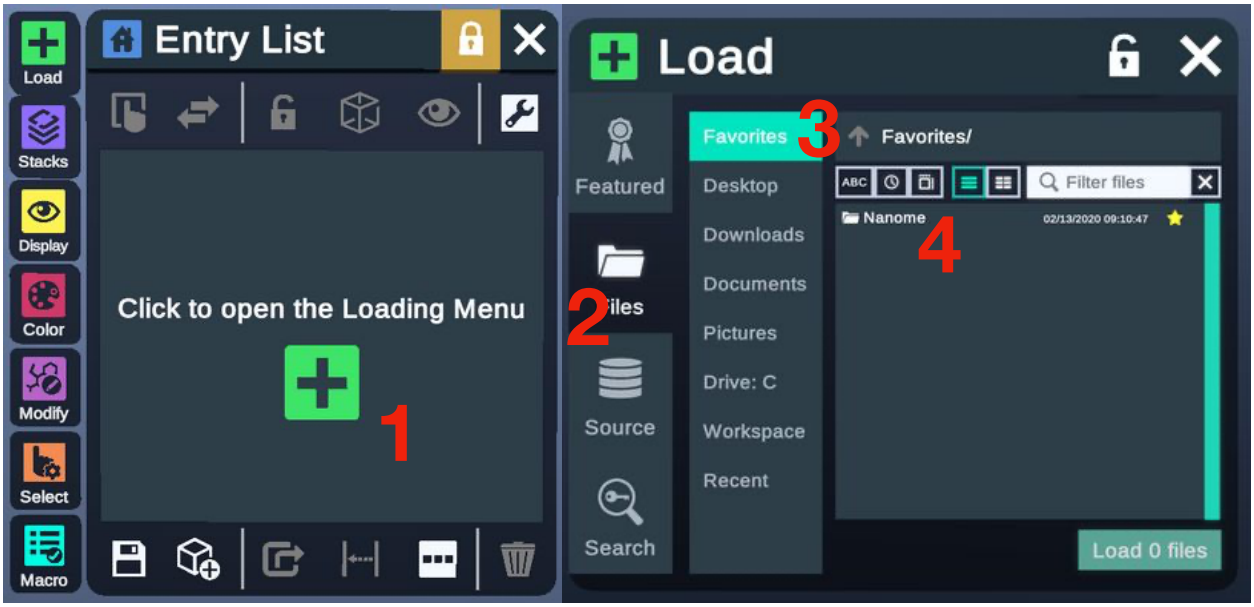
In this case, a short fragment of p53– a peptide epitope— folds into a small alpha helix to interact with MDM2 and MDMX. First go ahead and search for the structures of MDM2 and MDMX, and possibly find a pdb with p53 bound.

Report the PDB ID of one or more complexes you find.

You can now identify what are the main driving forces for binding:

Tasks in VR:

1. Open Nanome and load the `MDM2-X.pdb` file from `favorites/Nanome/Task-X/` by using the load module. In this file, chain A corresponds to MDM2 and chain B corresponds to MDMX. This file is adapted from PDB codes 1YCR and 4N5T.

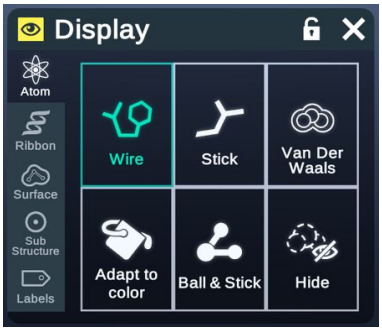


2. How similar/different are MDM2 and MDMX?

- Answer from the point of view of structural topology (secondary structure elements, looking at a cartoon representation).
- And then answer from the point of view of a ligand binding: show the surface representation, is the cavity equally large on both proteins?

One capability of Nanome that you can use to compare to similar structures is structural alignment. In the Entry List screen, select the two chains and click on the button to align them. This will help you see major differences between two similar systems.

3. Now use the trash bin button to delete the structures and load the `MDM2-p53.pdb` file from the same directory. Inspect the structure. This file is adapted from PDB ID 1YCR.
4. Use the *Display* panel and the *Entry List* to set the representation of different parts of the system so that you can answer the following questions:



5. Does p53 bind in a cavity?
6. List at least five interactions that you think are favorable for inhibition of MDM2 by this peptide. Explain each item in detail, including which atoms are at stake and why the interaction is favorable. Such interactions can include hydrogen bonding, stacking, hydrophobic interactions, salt bridges, etc.
7. Label the atoms participating in each interaction with atom number.
8. From the *Tools menu* choose the *Measurement tool*, use the *Distance* option to measure the distance of your potential interactions. Keep the distance labels for your list of five interactions.
9. Use the camera module to take a picture of your interface with the labels for all five interactions visible.

There are several structures of ligands and peptides bound to mdm2/mdmx. Most notably pdiq and ATSP peptides.

10. Now load `MDM2-pDIQ.pdb` from the same directory as previous files and drag it side by side with `MDM2-p53`. This file is adapted from PDB code `3JZS`
11. What are the differences in the PDIq peptide structure with respect to p53? Are the anchoring residues the same? Are there more/less?
12. Finally, select the pDIQ peptide from the Entry List and duplicate it to generate a copy of the peptide ligand, modify it (mutate it) and create a new peptide that is a better candidate to bind MDM2/X.
13. While small molecule drugs inhibitors to MDMX remain unsuccessful (low binding affinity), there are several small molecules (in the PDB) binding MDM2. What features do they have that make them similar to the original peptides?

One of the disadvantages of peptides is that they cannot be taken as pills, since they rapidly degrade (are digested) — some of the famous peptide drugs have to be injected (think about insulin). Thus, pharmaceutical companies will work hard to develop small molecules that can mimic peptides — rather than using the catalog of millions of known small molecules. The technique is called peptidomimetic design. Search for relevant bibliography on the subject and:

1) identify what would be the steps you would take to apply those principles to your system
2) can you derive a small molecule based on these principles?