Section V: Discussion

Using SwissDock, we were able to achieve not only separate iterations of fluorination but also to view the different aspects of each new fluorinated drug's biological nature. Seeing how fluorination affected the drug's lipophilicity and overall binding affinity assured the peculiar nature of fluorine when coupled with other molecules. Through viewing the different iterations of hydroxyurea, we were able to deduce important factors that would make specific iterations more feasible than others.

Analyzing each iteration, the viability of each possible synthesis becomes evident. With respect to normal hydroxyurea, its chemical makeup, as well as its physiochemical makeup allows it to be a fairly viable drug in and of itself. Its small size and its simple synthesis attest to its commonality and monetary feasibility amongst individuals who regularly take the drug. With a low log*P* of -1.09, the drug ranks highly amid all other iterations. Iteration I, with a Log*P* of -0.29, ranks lower than normal hydroxyurea due to its being closer to Log*P* < *5* than normal HU. Iteration II, having a Log*P* of -0.47, ranks higher than the iteration I, yet still lower than normal HU. Iteration III is lower than all three iterations, having a log*P* of -0.04 (much closer to Log*P* < 5). The final iteration, iteration IV proved to rank the lowest of all with a Log*P* of 0.63. Considering all other iterations had Log*P* < 0, iteration IV proves to be the outlier in the data in terms of its closeness to the limit of Log*P*. The reasoning for iteration IV's low ranking thus far is most likely due to the number of fluorine atoms it is comprised of (3). With fluorine being a reactive atom, its stability depends on its bonding with other atoms. Having fluorine added to one nitrogen atom adds minimal confliction the overall lipophilicity of the molecule, however, by adding more fluorine atoms to the molecule, its original physiochemical makeup becomes altered to a substantial amount as compared to iterations where only one fluorine atom is added. This attests to the reasoning as to why it was important to focus on fluorinating hydrogen atoms (less affected by bonding), as most of these atoms were bonded with nitrogen atoms (more stable atoms). Therefore, in terms of lipophilicity, it is expected that normal hydroxyurea performs more optimally due to the lack of fluorine in its chemical makeup.

However, each iteration scored lower respectively in terms of lipophilicity, most likely due to the reason that adding a fluorine atom would alter such. Despite the decrease in the score, all molecules continue to be deemed lipophilic due to their respect Log*P* values being less than 5.

With respect to solubility, hydroxyurea displays optimal results with a Log*S* of 0.81. As previously discussed, due to its lack of added atoms to its chemical nature. Iteration I follow suit with a Log*S* of -0.225, allowing it to be in the *highly soluble* range of solubility, according to SwissDock AME (Log*S* > 0 ~ highly soluble, Log*S* >-2 ~ very soluble). Iteration II offers semi-close with a Log*S* of -0.155, allowing for a *highly soluble* range of solubility, as expected with the change in position of the fluorine atom. Iteration III displays a result of a Log*S* value of -0.185, affirming its position below that of the first iteration and previous iteration, deeming it *very soluble* in the scale of solubility. Iteration IV displays an expected result of Log*S* = -0.61, deeming it the least soluble of all of the iterations, yet is still considered *very soluble* on the solubility scale (see Appendix D). In retrospect, it is most optimal to have drug iterations that fall within the *highly soluble* to the *very soluble range*, as these factors are determinants of whether a drug is viable to synthesize (due to the dependency on the effectiveness of the drug being on its ability to be solved into liquid). All iterations of hydroxyurea met this framework, allowing them all to be possible choices of synthesis.

Analyzing the boiled graphs of each iteration, a pattern is evident in the position of the molecule to the blood-brain barrier. Each time a fluorine atom Is bonded to an atom on the molecule, the overall relationship of the molecule to the blood-brain barrier increases. Based on the results of each molecule, iterations III and IV prove to be the most potentially harmful *in vivo*, as both molecules penetrate the blood-brain barrier potentially causing neurological effects to any individual who was to consume each molecule. In this manner, both iterations can be ruled out when identifying which iteration would be the most optimal to synthesize. With the pharmacokinetics and pharmacodynamics analyzed, it is important to consider the main agenda of the experiment: the hydrogen bonds formed through the protein interaction of each molecule with ribonucleotide reductase.

Analyzing the hydrogen bond formations of each molecule allowed for an in-depth analysis of each molecule. Though the boiled-egg graph allowed for inferences to be made as to what iterations of fluorinated hydroxyurea would be the most optimal if synthesized, this did not provide information about the quantifiable benefactors that would come as a result of each iteration. Normal HU was expected to have a hydrogen bond formation percentage that was higher than most—thus it did, however the hydrogen bond formation percentages of other iterations were higher than that of normal HU, proving that fluorination did in fact play a role in such a factor. Viewing each iteration individually allowed for decisions to be made about the optimality each iteration would have based not only on the results of the boiled-egg graph, but also the bonds that formed via simulation. An increased number of hydrogen bonds indicated that if such a molecule were to be placed in an environment with the protein that it was meant to target (ribonucleotide reductase), then it binding to such a protein would be much stronger—thus allowing the individual taking hydroxyurea to not have to take the drug as frequently as once every day anymore. An increase in hydrogen bonding with respect to a protein and ligand indicates the difficulty it would take for the ligand to be excreted as easily as it was with the original amount of hydrogen bonds present. This goes to show the overall benefit of adding fluorine to molecules as a means to bolster the molecule as whole. However, something that became more evident as the experiment progressed was that hydrogen bond formation either increased, or slightly decreased as a reflection of where the fluorine atom was placed on the molecule.

Each iteration was created in the ideology that every hydrogen atom of hydroxyurea would be replaced with fluorine in order to measure the effects at each position. Although the magnitude at which each position would cause a potential impact was unclear, it was expected that position would play a prominent role in the amount of hydrogen bond formation each molecule would experience.

This information allows for the possibility of more iterations to be made with respect to the iteration that had the most optimal turnout during the experiment; Iteration I. Possibly adding more fluorine to the molecule in addition to the fluorine bonded to the oxygen—as shown in Iteration I—could

possibly allow for an even larger result in the aspect of hydrogen bond formation. This is justifiable from the results displayed Iteration IV, where three fluorine atoms were bonded to different atoms in hydroxyurea, yet its overall h-bond yield, was higher than that of normal HU. This affirms viewing hydrogen bonding as a result of an external atom being bonded to the original molecule from a 'positional' aspect, as the position of binding of such an atom does have an effect on the hydrogen bond yield—as shown.

Conclusion

In retrospect, the question of whether modifying hydroxyurea to increase its binding affinity to ribonucleotide reductase has proven to be a feasible outcome. Seeing that certain iterations harbored flaws in certain areas, yet continued to yield results through simulation, indicates the true value of simulation in the realms of computational biology. Seeing that we were able to view the intrinsic electrostatic interactions of each molecule in order to view its overall outcome with respect to the hydrogen bonds of the molecule allowed us to measure not only the physical properties of each iteration but also its interaction and physiochemical capabilities when placed in environments where such notions are emphasized and tested. Considering that hydrogen bonds formed as result of the position it was placed on hydroxyurea, we are able to deduce not only the feasibility of this method in the realms of other drugs, but the overall affirmation that fluorination does play an impact in the hydrogen bond formation of molecule with the magnitude of impact being a reflection where the atom was bonded on the molecule. Knowing this, the feasibility of fluorination becomes something highly emphasized as fluorination could provide the benefit of increasing the biological nature of a mélange of different molecules. It not only allows for an increase in binding affinity to the protein that the molecule intends to bind to, but it also allows for fluid analysis of how the addition of an atom on a molecule affects the overall nature of the molecule. This may open many doors in the realms of medicine and pharmacology seeing its ability to be

effectively implemented through computational biology. All in all, using computational biology may not only be the key to solving niche issues in the realm of health, but it may be the answer to many questions that human minds may struggle to answer with tangible measures. An emphasized future where computational biology is coupled with experiments *in vivo* is a future where the most powerful and drugs can be made. These findings may transcend to different anemic diseases that infringe on blood cell shape and nature, providing a long-awaited breakthrough for individuals all over the world who suffer in silence.