Biochemical studentship report
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Supervisor: Taufiq Rahman

Background:

The project was aimed to design novel peptides targeting membrane proteins using an ion channel (Nav1.7) and a G-protein coupled receptor (the GLP-1 receptor, GLP-1R). Both these proteins are valid therapeutic targets (https://doi.org/10.1517/14728222.2016.1162295 and https://doi.org/10.1016/j.molmet.2020.101102), with some small-molecules and peptide modulators (including venom-derived toxins) already existing. Exploiting the known critical hot spots on the ligand-binding sites of these proteins, I designed some novel potential peptide binders utilising computational tools such as Pymol, AlphaFold and RFDiffusion in this process.

Method and Results:

As the project began, I was introduced to various molecular modelling and viewing software such as Pymol and ChimeraX, of which I had no prior experience. I spent the first few days familiarising myself with the different tools available in each software learning to manipulate and visualize protein structures effectively.

Following this, I was introduced to protein structure modelling using AlphaFold 3.0. and I ran some validation modelling exercises to predict binding modes of known peptide ligands of the GLP-1 receptor and the Nav1.7 channel. During this time, I also evaluated he performance of few protein-protein docking approaches (e.g. using ClusPro, ZDOCK, ROSIE-2, PyDock). I settled on using PyMol to view my protein structures and a combination of AlphaFold 3.0 and ClusPro to predict the protein-protein docking as this seemed to give me the best results.

After getting comfortable with these tools, I then trialled various existing peptides to see how well they docked to the truncated versions of the GLP-1 receptor and Nav1.7 ion channel and after finding the best poses, I submitted these for computational alanine scanning using BUDE alanine scan (https://pragmaticproteindesign.bio.ed.ac.uk/balas/) to confirm the binding hotspots of GLP-1 and Nav1.7 and to identify the key residues in the peptides that are responsible for binding to GLP-1 and Nav1.7. Using a similar process, I trialled a few sequences that my supervisor, Dr Rahman, designed by grafting a section of a venom-derived toxin (Huwentoxin) into a sunflower peptide STFI-1 backbone and flagged the ones that gave the best results.

Using RFDiffusion (https://pubmed.ncbi.nlm.nih.gov/37433327/), recently introduced by the David Baker group, I was able to design some novel peptide binders against the Nav1.7 ion channel and using both AlphaFold 2.0 ClusPro I identified the peptide binders that are likely to bind in the ideal position on the ion channel.

Future Directions:

The next step is to custom-synthesise the top-performing peptides and experimentally evaluate their binding affinities and functionalities through assays. This could lead to the discovery of more selective peptide modulators with potential therapeutic applications.

Departures from the original project plan:

We had planned to use RFDiffusion to design novel peptides for GLP-1 and we had also planned to look at the Adenosine A1 receptor however due to time constraints we were, unfortunately, not able to do this.

Value of Studentship to the Student and the Research group:

Student:
This internship helped me gain advanced computational skills that are widely being utilised to understand protein structure and function, such as structural prediction using AlphaFold, peptide design with RFDiffusion, and molecular docking simulations. I also enhanced my project management and communication skills through presentations and lab discussions. This experience has further motivated me to pursue a research career in rational drug design, aiming to develop novel therapeutics. This research experience will no doubt help open up future opportunities for research in my career since I am very keen to work in rational drug design.

Research group:
This funded studentship allowed Aleena to pursue some studies that Rahman group had been thinking for a while. Aleena did excellent and has come up with several interesting peptide designs that will be taken forward and experimentally validated in the near future. This would certainly help in generating the proof-of-the-principle for new research avenues for the group.