When I applied for the vacation studentship, I was halfway through the second semester of my second year of pharmacology and drug discovery. I needed to gain lab experience, and due to my interest and enjoyment of my degree, I wanted to continue to learn throughout the summer. Dr Leanne Stokes advertised the BPS vacation studentship, with the suggestion that any interested students could apply with her. It was exactly what I was looking for, as well as the bonus of being up to 10-weeks, which would be much closer to the length of a real research project than others offered at the time. We discussed the project and weighed up different potential topics focused on pharmacology. Once we came to a decision, we discussed a research plan and wrote a project outline. Fortunately, the process to apply for the studentship was easy and didn't take much time at all.

The project would be an exploration into the pharmacology of P2X7, an ionotropic purinergic receptor, activated by extracellular ATP- typically thought of as an intracellular energy molecule, but well known to produce signalling through purinergic receptors. P2X7 is involved in inflammation and cancer amongst other diseases such as psychiatric disorders, and antagonists of the receptor are currently in clinical trials for depression. Much is still unknown about the receptor, including the binding sites of many negative and positive allosteric modulators. The goal of the research was to characterise both positive and negative allosteric modulators and attempt to provide evidence for their sites of action. To do this, we used HEK-293 cells with stable expression of human wild-type and mutant P2X7 receptors.

Using various techniques, the cells would be exposed to varying concentrations of compounds and agonists (either ATP or BzATP). The resultant response from P2X7 activation would be measured mostly via intracellular calcium measurements using the indicator dye Fura-2AM. The Fluorescence would then be recorded via flexstation. The experimental data would then be used to generate agonist dose response curves for negative and positive modulators. It was also used to create dose inhibition curves to derive the IC50 values of P2X7 antagonists. We explored the impact of key mutations in the known antagonist binding pocket at P2X7 on the effect of a series of potential antagonists.

Initially, I found it very challenging to get used to splitting cells, maintaining sterile technique, and accurately pipetting. I found myself making mistakes and working slowly to avoid them happening again. With practice, help and advice from those in the lab, I managed to improve over time, growing in both skill and confidence. This confidence manifested itself in me being able to get more involved in the research; I did more reading and used the questions it arose to suggest potential experiments. During the project, I began to take interest in computational docking, to which Dr Stokes taught me the best methods to go about it and gave me documents and files I could use to do it. Over time we confirmed and discovered new information about the receptor's pharmacological properties, and I attempted to come up with ideas to explain them during discussions with Dr Stokes, which really helped push my knowledge of the field.

I was also witnessing techniques from other members of stokes lab that I was not doing myself, such as flow cytometry, transient transfection, and various types of cellular assays. I was also introduced to their research during regular lab meetings, where we would discuss our findings as well as other important papers.

The project was a success in more ways than one. We achieved what we to set out to: we characterised the pharmacology of the NAM mutants and found many interesting pieces of evidence to follow up in the future. It was also personally a success- as I had achieved everything that I had set out to do with the studentship and more. I have improved confidence, and improved technical ability, and as a result am now much more prepared for my third year and beyond. I am also now better at planning experiments, reading papers, and directing my focus. I have found a topic to peruse in the future, as I look to see the questions raised before and during this studentship be answered.

For all this I have gained from this experience, I would like to thank the British Pharmacological Society for funding this opportunity. I would also like to thank Dr Leanne Stokes for not just allowing me to work in her lab, but in guiding me and giving me her time and knowledge to improve as a scientist, as well as the freedom to pursue some of my ideas and make my own mistakes.