Getting stuck into summer

While studying at university, the thought of what to do after graduating can be daunting. With the changing job market and so many career choices available to bioscience graduates, it can be confusing to know where to start and whether it’s the right path for you. Work experience while at university plays an essential part in making these decisions and bolstering your CV (résumé) to maximize your employability. For those who are keen to pursue a career as a scientist, such as carrying out a PhD and moving on to postdoctoral positions, work experience in a laboratory is essential.

My second home for 8 weeks this summer. Within minutes of being shown my bench, I had donned a white laboratory coat and blue gloves, and launched straight into performing various tasks: setting up PCRs, making up bacterial media, running agarose gels, performing restriction digests, DNA ligations and transformations (to name a few).

The aim of my project was to express a green fluorescent protein (GFP)-tagged version of Maf1, a putative glycosyltransferase in the opportunistic pathogen *Aeromonas caviae*, enabling it to function as a probe in live *A. caviae* cells to follow its subcellular location using fluorescence microscopy. At the start of my project, it was unknown whether Maf1 localized to the cell pole alongside its flagellin substrates or was dispersed throughout the cytoplasm. The prospect of being involved in a new scientific discovery was exciting as exploring the unknown is what science is all about, and is the very reason that I chose to study microbiology at university.

Unfortunately, when cloning in bacteria, you cannot avoid two inevitabilities: first, you need to have a very strong stomach to stand the smell of bacterial culture following breakfast and, secondly, it will not work the first time. For 4 weeks, I tried to clone my GFP–Maf1 fusions into *A. caviae*, repeating what seemed like endless rounds of PCRs, restriction digests, ligations and transformations, all to no avail. Despite the failed experiments, I was never made to feel that it was my fault and very soon came the success. I successfully managed to clone and express a gene that would produce a GFP-tagged version of the flagella basal body protein FliF (FliF–GFP) which was required as a control. I was also asked to come up with a protocol that would enable *A. caviae* to swarm, a type of motility which is difficult to achieve in a laboratory setting.
I had a wonderful time in the laboratory: I got to work both independently and as part of a team, I took part in regular laboratory meetings, went out on laboratory lunches and delivered presentations about my project.

To quote my supervisor, you really do ‘learn more when things go wrong’ and, as a result, I gained extensive experience in performing a range of microbiological and molecular biological techniques. The most rewarding part of my project came on my last day when I visualized FlhF–GFP in A. caviae cells using fluorescence microscopy. Seeing that all my hard work had paid off made it all worthwhile and I felt lucky as I hadn’t been able to perform fluorescence microscopy as part of my undergraduate studies. I strongly urge any student thinking about taking part to go for it! You’ll have a lot of fun, meet like-minded people that’ll give you valuable advice on getting your career started and, as a budding scientist, you won’t find a more inspiring or valuable way to spend your summer. I cannot thank the Biochemical Society enough for funding my experience.

Kristen A. Johnson worked with Dr Greg Miller at the Catholic University of America,

When a protein is first created as a simple amino acid peptide chain, there are multiple steps before it can become a fully fledged highly efficient enzyme. The same is true for any young scientist, like me. Inherently, all of the components are there, but time, guidance, and discipline are first required to mould those naive components into a whole new scientific research-loving undergraduate.

This summer I was awarded a grant from the Biochemical Society that provided me with 8 weeks to deepen my understanding of biochemical research. Under the guidance of Dr Greg Miller, my research advisor at the Catholic University of America in Washington DC, I was introduced to a wide variety of laboratory techniques. I developed discipline through hard work and the daily experiments working to purify and assay the activity of the inositol phosphate phosphatase ipgD. This enzyme is injected into a mammalian cell by Shigella, a genus of pathogenic bacteria that can cause dysentery, and alters host cell Akt signalling by dephosphorylating its inositol phosphate substrates. The end goal of this research was to understand further how this enzyme recognizes a specific set of host substrates. Starting out, I cloned the ipgD gene and transformed this into Escherichia coli, which was our expression system for producing the enzyme. Once sufficient soluble enzyme was observed, a larger-scale bacterial growth and purification protocol was carried out. I isolated ipgD to sufficient purity to allow me to detect and measure the enzyme’s activity using a colorimetric phosphate assay. Much work remains before I understand how ipgD identifies its substrates, and I hope to decode some of the lingering questions regarding substrate specificity this semester with Dr Miller.

The work completed this summer was reasonably successful, but it was not all smooth pipetting. Sometimes experiments did not go quite as planned and they had to be repeated or redesigned. In hindsight, as frustrating as these rough days were, they were usually more educational than the successful days. The difficulties I faced pushed me to develop valuable troubleshooting and experimental design skills. Through this experience I have gained confidence and a passion for learning. As a result, I now know that I want to pursue a PhD in biochemistry. In addition, after my summer vacation studentship with the Biochemical Society in Dr Miller’s laboratory, I want to dedicate myself to learning and research. Finally, I have learned to welcome the integration of experimental and theoretical learning that is catalysed by a research environment. To me, this is the best education any young scientist can have; without the pressures and exams of a normal school year, I could learn solely for the sake of learning. I am so grateful to have had this amazing summer experience with the Biochemical Society, the Catholic University of America and Dr Miller.
Laura Mariotti carried out her placement at Birkbeck College, London, with Dr Cara Vaughan

Laura Mariotti (right) with her supervisor Dr Cara Vaughan

The day before starting my placement, I was so nervous that I had trouble going to sleep. I was scared of disappointing my supervisors with the little I thought I knew. Fortunately, within the first few days, I already felt ‘at home’ (even if I still felt a little bit clumsy), I was comfortable asking questions not only of my supervisor but to other people in the laboratory too.

My project was related to Hsp90, a chaperone. Pp5 is one of its substrates and its activation allows it to dephosphorylate cdc37, an Hsp90 cochaperone. We designed a chimeric protein with the catalytic domain of pp5 on one side of a linker and a cdc37 peptide on the other side. The aims of the project were to clone, express and purify this chimeric protein in order to visualise the cdc37 peptide bound in the catalytic site of pp5.

The first two weeks were physically and mentally tiring, as I had a lot of information to assimilate and running around the laboratory all day turned out to be an effective work-out. Having to write all the experiment details in a lab book helped me to check that I had understood everything. I also had weekly meetings with my supervisor to present my results and discuss the following week’s experiments which were very beneficial for my understanding of all the techniques used.

I purified my protein after 3 weeks but the yield was very low. This disappointing result turned out to be a valuable source of information: I got a chance to see and compare how the yield increased when the fusion tag was changed and therefore further understand the properties of my protein. This studentship was not focussed on getting results; learning was what mattered most.

As the weeks went by, I got more and more committed to the project and it saddened me to leave the laboratory when the studentship was over. You also get to experience many feelings: satisfaction, patience, disappointment, excitement, etc. These are the sort of feelings that you don’t discover in laboratory sessions at university. This internship was much more than work experience; I grew up as a person (my confidence increased dramatically!). I met incredible and friendly scientists at different stages of their careers and I certainly will stay in touch with most of them. I am now sure that I want to pursue a career in research and I am much more prepared for the choices I have to make this year regarding PhD applications, for instance. This placement was the best work experience I’ve had and I strongly recommend all undergraduate students to apply.