

The Loss of Polyamines from Cells is a Marker of Toxicity

BTS Vacation Scholarship 2017

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This 8-week project gave me an excellent insight into working on the fascinating field of toxicology. It provided me with great opportunities to deepen my current medical science knowledge, and with a better understanding of the numerous and diverse biochemical pathways underlying uncountable mechanisms of toxicology.

From the first day, after a warm welcome from Professor Wallace and her colleagues, I had extensive opportunities to plan, set up and carry out experiments that I hadn't encountered before. Throughout the whole project, I developed effective processes to find, read, and interpret appropriate peer-reviewed papers and reviews. This greatly improved my critical and analytical thinking about the results of each of our experiments and how they contributed to our overall conclusion.

During the first two weeks, the recovery and characterisation of cell lines was our focus. Cell lines of interest were HCT116 and WiDr (both human colon cancer cell lines) as these were found to be suitable for further experiments to investigate our hypothesis, which is that the loss of specific polyamines from cells will be an early marker of toxicity and that the pattern of loss will distinguish inhibition of cell growth from major toxic. Cells were cultured under the required conditions and normal growth curves were prepared by applying techniques used for total and viable cell counting, such as the trypan blue exclusion. Here I learned the importance of accurate and precise laboratory record keeping and the proper way to maintain my laboratory notebook.

To determine the cytotoxic effect of etoposide and paraquat on HCT116 and WiDr, the MTT assay method was used. The application of this method was very challenging and required dexterity, focus and perseverance.

With the appropriate concentrations of drugs, the two cell lines were exposed to growth inhibiting and cytotoxic conditions. For the protein content analyses of the treated and control cells, the Lowry protein assay was used, while for the polyamine content analysis LC-MS was applied. Results were analysed using Prism. Learning all these techniques further improved my precision, attention to detail and my ability to analyse data.

The results and summarised data of the project seems to support our hypothesis, however further development of experiment designs and the testing of other cell lines, such as healthy

human colon cells, could additionally add to our current findings or provide a new perspective on them.

This project greatly contributed to my scientific development by giving me confidence in working in laboratory, planning and applying different experimental designs and analysing data. Attending weekly lab meetings and presenting the latest findings and status of the project, I acquired good presentation, communication and brainstorming skills.

Being given this excellent opportunity has allowed me to become more determined and motivated to progress my studies in toxicology after graduating. The extensive support, supervision and feedback of Professor Wallace and her colleagues on my work and professional development during the project improved my scientific perspectives tremendously.

I would also like to thank the British Toxicology Society for funding this project and giving me the privilege to join to their internationally acknowledged organisation.