Metabolism of Renal Cells and Osteosarcoma Cells Funded by the NIH Professor Looyenga Emily Roloff, Kylee Rosette, Cami Barnes

This summer I worked on a project concerning the metabolism of cells. The two main classes of cell lines I worked with are the kidney cell lines and the osteosarcoma cell lines. The kidney lines consist of the HK2s, which are normal kidney cells that have been immortalized, and two cancerous lines called Caki2s and SKRC39s. The three different types of osteosarcoma (bone cancer) lines are the 143Bs, MHOS, and U2OS. The different ways I went about looking at metabolism was seeing how each line responds to differing levels of fetal bovine serum (a growth nutrient), varying types of macronutrients, and differing bases of media. Another slightly different project I worked on consists of growing the kidney lines in kifunensine, a drug that inhibits the amount of glucose into the cell, and seeing how they respond.

To obtain results, I performed a variety of different experiments. One is called an EdU assay, which places a fluorescent tag on cells that are dividing, and we use flow cytometry to detect the tag and quantify how many cells are actively dividing, and therefore growing. I also performed resazurin assays, which detect cell growth by using a dye on cells that will change color when more metabolic activity is present. Immunoblots were another tool I used this summer. This consists of separating proteins on a gel by weight, and this allows us to probe for certain proteins by using fluorescence. Lastly, I utilized 2-deoxyglucose (2-DG) uptake assays that track the amount of glucose cells are using by utilizing radioactive glucose. In recap, the EdU and resazurin assays track cell growth and proliferation, immunoblots allow us to look for certain proteins, and 2-DG uptakes show how well glucose is getting into the cell.

From the above methods, we have obtained some useful data. When we alter the amounts of fetal bovine serum used in our growth media on the kidney lines, we notice that the cancer lines (Caki2 and SKRC39) respond better, whereas the HK2s (normal kidney line) are not as sensitive. This is what we would expect because cancer cells will keep growing without restraint, whereas the normal line have precautions in place that tell it when to stop. We have discovered that the Caki2 cells from our kidney lines depend very much so on glucose to survive. We have also found that the HK2s and SKRC39s are not as solely dependent on glucose as the Caki2s, but instead can survive on a variety of medias, which indicate that they use oxidative phosphorylation to survive. When we grow the kidney lines with and without kifunensine, interesting proliferation data emerges. The Caki2s grow better in kifunensine, despite depending on glucose to survive, whereas the SKRC39s that have other means of proliferating grow worse in kifunensine. Both results are the opposite of what we would have expected, and more research needs to be done on why this is.

My research position this summer has brought great benefits to me. For one, it has given me great experience before applying to medical school. Through researching, I have made gains in understanding scientific literature and protocol. For example, I would read an article about a certain experiment, and then I would follow through and do an experiment in the area of the article. It helped to bring everything full circle, which helped comprehension. Overall, research has been an exciting experience that has introduced me to a new perspective of science that it actually doing science, rather than reading about it in class!