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Biological Science
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SEF Final Reflection Fall 2019

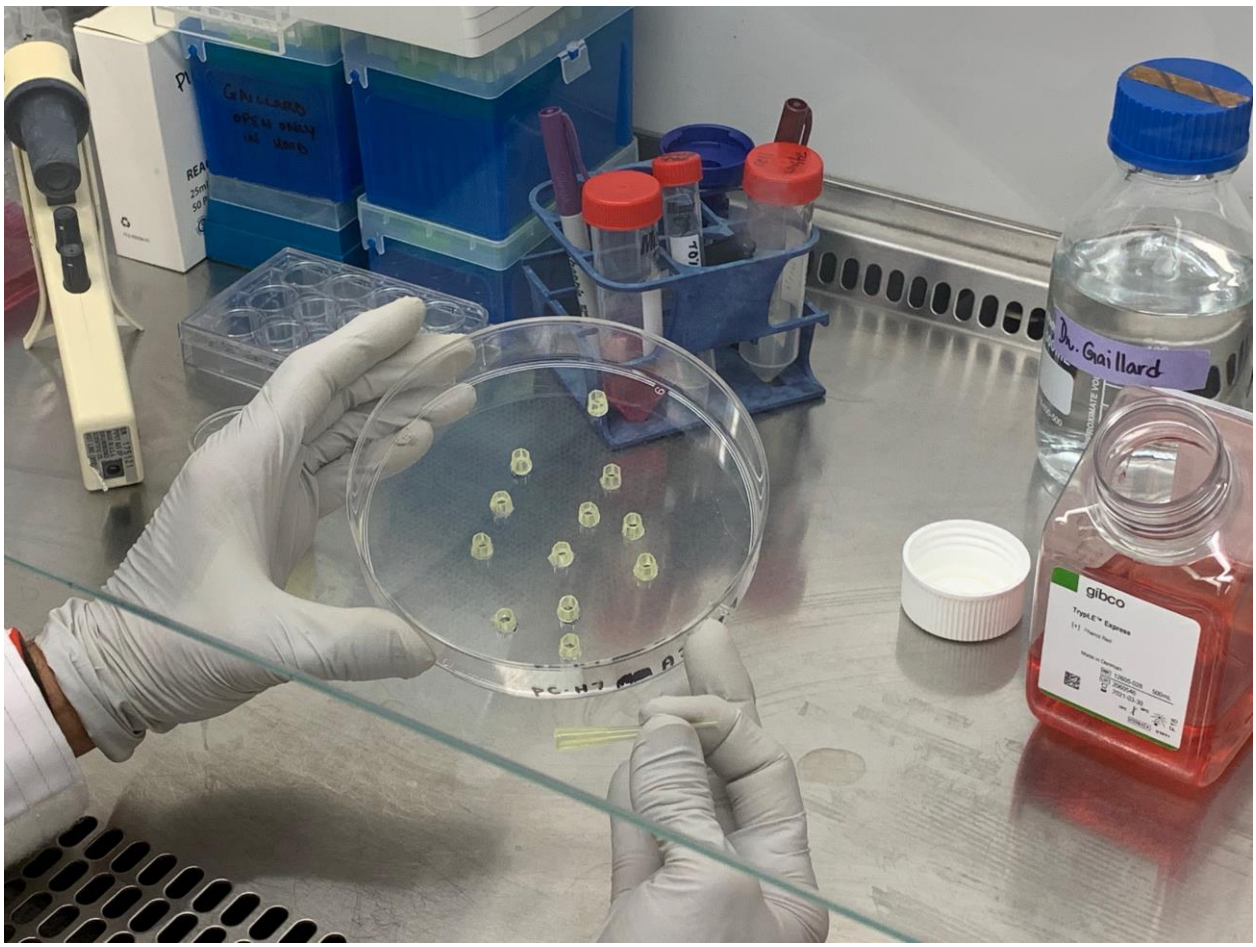
This semester, I was able to begin working on a project involving two types of human hepatocellular carcinoma cells that had been altered with the CRISPER-Cas9 system to eliminate the expression of a specific transport protein, ASCT2. This protein allows for the passage of essential amino acids in and out of the cell, causing it to grow without regulation. ASCT2 has been found to be overexpressed in most types of cancer cell, which is why research is being done on the effect of removing it from cells. Previous projects have found that simply removing the protein forma cell does not prevent the cell from growing, as there are other pathways used by the cell to transport the amino acids that would normally be moved through ASCT2.

My project was focused on isolating populations of cells, and gathering data on which populations were complete knock-outs for ASCT2, and did not show any expression of the protein. This was done by growing out a very small number of Huh7 or Sk-Hep that had been modified by CRISPER-Cas9 tin a very large dish until small colonies of cells began to grow. These colonies were then isolated, removed, and allowed to continue to grow out in separate smaller vessels. Once the cells had been growing for a few weeks, the protein from each harvested and analyzed using a Western Blot analysis, which allowed us to determine which specific cell populations still showed fait amounts of ASCT2 expression.

The next steps for this project will be using the cells that we now know are complete knockouts for the protein in other experiments. If we are sure that ASCT2 is not present, then other tests can be done to see the effects of different potential treatments in addition to the cells being deprived of the transport protein. It has been so interesting to see the steps behind the

scientific methods that go into current research topics, and I have gained a deeper understanding of so many lab techniques. I see so many images and diagrams in textbooks, but by doing research, I have been able to see the work that goes into actually creating those sources of data. I am excited to continue to study this topic and see what more can be done with the work from this semester.

Isolating clonal populations to be moved and grown out



Processing protein samples using sonication

Example Western Blot from Huh7 Isolation