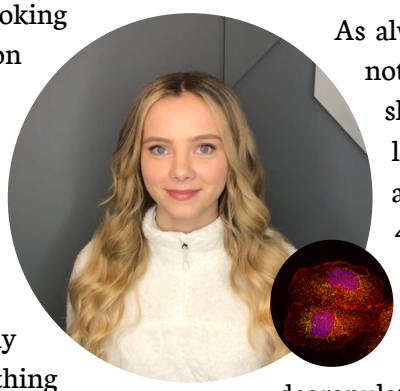
Student Researcher Spotlight – Juliette Eshleman

Juliette is an undergraduate student in the Faculty of Science who is researching the effects of Rho protein activation on mast cells degranulation. Her image titled *Mast Cell Degranulation in Response to Rho Activating Drugs* is featured on the front cover of this issue.

What prompted your interest in research? How did you become involved in research?

I became involved in research starting in my third year of university. By that time, I had already become very interested in cell biology through the prerequisite classes I had to take in my first two years and was looking for opportunities to do more hands-on learning. I attended a research recruitment night, hosted by the faculty of cell biology the semester prior, and through that was able to speak with various principal investigators of labs at the UofA. The passion they demonstrated for their work through the presentation I watched only further convinced me that research was something I wanted early exposure to as an undergraduate degree. I then reached out to several PIs by email, toured a few labs, and found a good fit for my interests! I joined the lab of Dr. Gary Eitzen, studying Rho GTPases in inflammatory processes. There, I've worked on several projects over the last two years and built up a lot of confidence in my lab skills.



What interests you about this field? Why is this research important?

Cell biology has taught me to be fascinated by the intricate and highly regulated functions of cells, and how dysregulations of these controls contribute to human disease, such as allergic responses. Moreover, protein signaling pathways were very interesting to me early on in my undergrad, so the focus my project has on Rho GTPases, a family of proteins which is essential in modulating cytoskeleton dynamics and functioning as downstream signalers in cellular pathways, made it a perfect fit! Pair that with my interest in human health, the immunological aspect of my projects and imagining the clinical applications this research could foreseeably have on allergic disorder treatment, it became all the more intriguing to dive into!

Have there been any unexpected challenges or roadblocks that you have encountered in your research?

As always with research, your hypothesis does not always hold up. I've encountered my fair share of roadblocks, but it's all part of the learning process and pushes you to become a better scientist. Most recently, in my Cell 499 project, none of my final results support my initial hypothesis. I expected the Rho GTPase activating drugs I used on Mast cells to result in elevated levels of degranulation, but what I found was either no significant difference, or even suppressed degranulation! This has pushed me to further develop my critical thinking skills and scientific literacy, as I search for a possible explanation to justify these unexpected findings.

Do you have any ongoing projects? Have you made any interesting findings? What are the implications of your research?

I am currently testing novel Rho activating drugs to determine how these proteins are involved in signaling pathways that govern the release of mast cell pro-inflammatory molecules. Previously published data has shown that drugs which inhibit Rho GTPase activity block degranulation downstream of FcεRI receptor signaling. I use RBL-2H3 cells as model mast cells to examine whether newly developed Rho activating reagents are sufficient to trigger receptor-independent Rho proteins; namely the drugs narciclasine and calpeptin, and deaminases that convert glutamine-63 of RhoA and -61 of Cdc42/Rac1 to glutamate, which block intrinsic GTPase activity and thereby locking Rho proteins in the GTP-bound state. I have performed assays

for granule enzymes in extracellular supernatants after treating RBL-2H3 cells with Rho activating reagents to determine their direct effect on degranulation. We found Rho activating drugs stimulated degranulation while deaminases had little effect in the absence of receptor stimulation. Hence Rho activation is likely not a sufficient degranulation trigger. Downstream effects of FcεRI signaling include binding of IP3 to receptors on the endoplasmic reticulum, resulting in elevation of intracellular $Ca2^{+}$, a required step for degranulation. As such, I used thapsigargin to induce calcium flux which resulted in the recovery degranulation. I also performed immunofluorescence microscopy on Rho activated RBL-2H3 cells to assess morphological changes to the actin cytoskeleton and granule-marker distribution.

Where do you see yourself in 5 years?

I hope to be finishing medical school in five years, give or take! We will see how things turn out as I prepare to graduate this semester. Becoming a doctor is my ultimate goal, but I am still contemplating pursuing a Masters in Cell Biology in the meantime. Either way, I hope clinical research to some capacity will be in my future as a physician.

What have you learned from your research experience? Do you have any words of wisdom for aspiring undergraduate researchers?

These last few years as an undergraduate researcher have really opened my eyes to how much there is to learn! Apart from many practical laboratory skills, this experience has helped me in developing my critical thinking skills, scientific literacy, and pushed me to think creatively in the face of surprising results or many failed experiments. I would urge anyone who is curious about science to push themselves by trying out research in their undergrad.