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Visit Narrative

I spent my summer in the Extavour Lab at Harvard University, working with the cricket *Gryllus bimactulatus*. The cricket stands to be a highly informative emerging model organism for several reasons. Not only is it tractable for laboratory research, but it also is expected to utilize ancestral modes of development which could provide an useful contrast to the highly derived modes of development implemented by *Drosophila melanogaster*.

Specifically, *Gryllus* will be useful for probing the method of germ line development. Previous work performed by the Extavour lab has indicated that *Gryllus* germ cells bypass the germ plasm, instead forming germ cells during mid-embryogenesis. I sought to better characterize this process in and of itself, and as a contrast to *Drosophila*, which utilizes germ plasm for germ cell specification.

Initially, this summer I sought out to utilize CRISPR/Cas technology to generate several new strains of crickets for use in studying germ line development. The gene *vasa* is an established germ cell marker. I intended to generate a *Vasa* knockdown, as well as to insert an attP recombination site into *Vasa* for use as a reporter line (following recombination with a fluorescence marker). So far, all of the offspring of the crickets which I injected have been genotyped as negative for *Vasa* genotype modification. However, there remain additional crickets to rear and breed that might be successfully modified.

As the summer progressed, myself and my mentorship team decided to couple with the high-risk high-reward CRISP/Cas experiments a set of lower-risk lower-reward experiments investigating the expression of Wnt related genes in germ cells. Therefore, I began testing a series of β-catenin antibodies, in order to see if they marked nuclear β-catenin. While one of these antibodies was an excellent cell wall marker, none successfully marked nuclear localization. Therefore, I began cloning Wnt pathway genes, for use in RNAi experiments and in situ hybridizations. Fortunately, the cloning was successful and I was able to begin RNAi experiments by the end of the summer.

While some experiments didn't work, including β-nuclear catenin immunohistochemistry and possibly the genome editing, the cloning did. If I were to go back, I would be inclined to suggest that I pursue more lines of experiments in tandem; however, a major reason I performed some experiments sequentially was that at that time I was still learning them, and wanted to truly focuse on each. Moreover, a major objective for the summer was, as an undergraduate, train in a series of techniques. I certainly learned many techniques, including microinjection, cloning and immunohistochemistry. Additionally, I gained laboratory experience and verified my interested in pursuing a PhD program. I had a tremendously productive summer, and I am very grateful for the support from the EDEN Undergraduate Internship Program which made it possible.