The identification and visualization of homologous early embryonic patterning genes in *Sciara coprophila*
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Many scientists have explored the question of how an embryo "knows" where to grow different body structures as it develops. They have found that specific genes expressed during embryonic development dictate the body plan. Generally, these genes code for proteins that function as transcription factors, which have the ability to activate or repress other genes that contribute to developing body structures. These transcription factors are expressed in specific patterns throughout the embryo, often in striped patterns. The genes involved in early embryonic development have been characterized in great deal in *Drosophila melanogaster*, the common fruit fly. The question then follows of how broadly applicable is *Drosophila's* developmental pathway to other related organisms? For example, *Drosophila* has a well-studied gene called *bicoid*, which helps organize body structures along the anterior/posterior axis. Interestingly, the gene *bicoid* is *Drosophila*-specific. No other organism has evolved to express it. How do other organisms that have different genetic makeups organize their body structures during development?

My research explored which early embryonic genes are involved in embryo patterning in *Sciara coprophila*, a species of fungal gnat. I chose *Sciara* because it is fairly closely related to *Drosophila,* but unlike *Drosophila*, it lacks *bicoid*, and may also have significant differences in the patterns and functions of its developmental genes. I was interested in characterizing the similarities and differences between *Sciara* and *Drosophila* and how they pattern their embryos through reconstructing developmental pathways in *Sciara*.

I identified fifteen potential homologs of known early embryonic patterning genes in *Drosophila* in the *Sciara* genome. To visualize the expression patterns of these genes, I utilized mRNA in-situ hybridization. Much of the summer was spent troubleshooting and developing an in-situ hybridization protocol to utilize in *Sciara*.

One aspect of this troubleshooting included the use of reverse transcription PCR (RT-PCR) to confirm the presence or absence of gene expression. While this technique does not provide any information on where a gene is being expressed, it is a sound method to establish whether there is indeed any expression of a gene. I performed RT-PCRs of the pair-rule gene *even-skipped* and the gap gene *giant* on 2-, 12-, 24-, and 48-hour old *Sciara* embryos. My results showed evidence of expression of *even-skipped* in the 12-hour old embryos and expression of *giant* in all time-points.

Another aspect of my troubleshooting included exploring the morphology of *Sciara* embryos at different time points to better understand when the stage cellularization occurs. In *Drosophila*, many early embryonic patterning genes are expressed just before cellularization. There are conflicting answers in the literature regarding the timing of cellularization in *Sciara*. I collected and fixed *Sciara* embryos at various time points and stained them with the nuclear stain DAPI. While I did gain a better understanding of *Sciara* morphology, I have yet to pinpoint the exact timing of cellularization.

I performed a successful in-situ hybridization of the gene *giant* in *Sciara* embryos (Fig 1). The expression patterns looked similar to those seen in *Drosophila* with two bands of expression in the younger embryos and expression concentrated in the anterior head region in the older embryos (Fig 1J, K, F, L). Both of these embryos are from the 27 hours after egg lay sample, which speaks to the wide range in embryo ages within a sample. This has contributed to the difficulty in pinpointing the timing of cellularization.

I look forward to visualizing the expression patterns of more early embryonic patterning genes, as well as contributing to a better understanding of *Sciara* embryo morphology during this upcoming year.

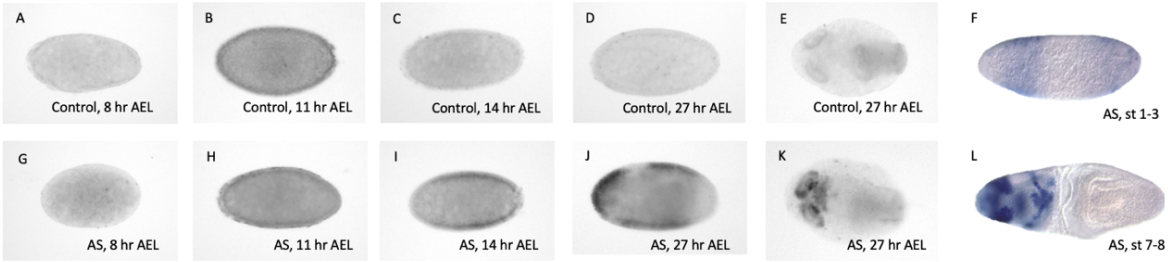

Figure 1. Visualization of *giant* expression in *S. coprophila* and *D. melanogaster* (F & L) embryos through mRNA in situ hybridization.

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