Reproductive Morphology In Adult Nematode Species and their Corresponding Reproductive and Cuticle Gene Expression

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Abstract- *Caenorhabditis elegans* has a hermaphroditic reproductive system which differs greatly from the [1]*Dioecious* systems of *Steinernema carpocapse* and *Steinernema feltiae*. The objective of our research was to compare the reproductive morphology of all three species utilizing [2]*Dapi Staining* then attempt to explain the findings by analyzing the relative expression levels of reproduction-regulating genes which are [3]*Ortholog* in all three species. To explain this phenomenon, the expression of cuticle-regulating orthologs was analyzed in the same manner as the reproduction-regulating orthologs. The analysis found that multiple orthologs exhibited expression levels in adult *S. feltiae,* which differed substantially from the other two species; this could be a possible cause of the differences in cuticular structure and permeability to the DAPI stain. The analysis of the reproduction-regulating orthologs demonstrated multiple differences in expression levels between all three species. However, *C.elegans* demonstrated the most drastic differences when compared to the *Steinernema* species.

[1]*Dioecious*- having the male and female organs in separate and distinct individuals [2]*Dapi Staining*- a fluorescent stain that binds strongly A-T rich regions in DNA. It is used often fluorescence microscopy [3]*Ortholog*- gene found in two or more species that can be traced to a common ancestor

I.INTRODUCTION

The reproductive system is one of the major areas in which *C. elegans d*iffers from *Steinernema carpocapsae* and *Steinernema feltiae. Steinernema* nematodes either have a male or female reproductive system, while *C. elegan*s are all hermaphrodites. The original objective of our research was to compare the reproductive morphology of *S. carpocapsae, S. feltiae,* and *C. elegans* by DAPI staining then use the differential expression data we obtained from RNA sequencing to explain the morphological differences we observed through DAPI staining. However, when DAPI staining was performed, we found the species demonstrated various levels of permeability in response to the DAPI stain. We attempted various times to crack the nematode cuticle, and experienced success. To determine the cause of the difference in response to DAPI staining, we analyzed the expression of cuticle-regulating orthologs in all three species, to identify which differentially expressed genes might be the cause of the differences in cuticle structure.

II.ANALYSIS

Although our analysis did not identify the specific orthologs or clusters that cause the reproductive and cuticular morphological differences, the possible orthologs that could cause such differences have been narrowed down. The RNA sequencing demonstrated that roughly 50% of the orthologs demonstrated differences in their expression patterns throughout the life cycle stages

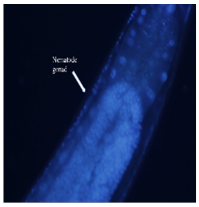

Fig 1:S.Feltiae (cracked) - 40x

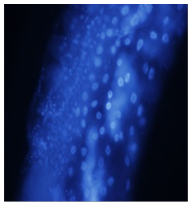
Fig 2: S. Carpocapsae (cracked) - 400x

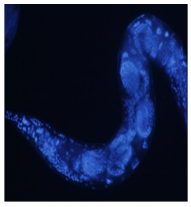
Fig 3: C. Elegans (cracked) - 40x

Orthologs that demonstrated different expression patterns in *C. elegans* than the *Steinernema* species (especially in the adult stage) may possibly be responsible for the hermaphroditic reproductive system in *C. elegans*. Similarly, orthologs that showed different expression patterns in *S. feltiae* than the other two species could be the cause of the differences in cuticular structure and permeability to the DAPI stain. Of the 288 reproductive orthologs and 31 cuticular orthologs, a significant number demonstrated variations in differential expression patterns between the three species, which could have caused the morphological differences. Due to the variance of multiple genes, we were unable to construct experiments isolating each varying gene and see if the morphological difference still persisted.

III.CONCLUSION

For future research to identify these orthologs, one could discard the ortholog that showed no significant change in their gene expression patterns between the three species. From the remaining orthologs, one could then select the orthologs that exhibited the greatest differences in expression and alter their expression levels, to see to how their expression levels affect the morphology of the nematode. Furthermore, one could mutate or completely remove the gene and observe the subsequent effect on the nematode. Such a study would provide greater insight into the genetics of *Steinernema* species and *C. elegans*, as well as the relationships between the two species.

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