# CALIFORNIA SCIENCE & ENGINEERING FAIR
# 2018 PROJECT SUMMARY

**Name(s)**

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**Project Number**

38301

**Project Title**

## Identification and Functional Characterization of Circular RNAs in Drosophila

**Abstract**

**Objectives/Goals**

Recent studies have described the important roles of circular RNAs in human disease development, and the unique structure of circRNAs makes their development as diagnostic biomarkers and disease targets possible. The purpose of this project is to investigate the function of circRNAs in Drosophila antibacterial innate immunity as well as to determine whether the circRNAs play any tissue-specific roles in flies (in vivo). Gaining a deeper understanding of these relationships could further elucidate the importance of circRNAs in both Drosophila as well as mammalian systems.

**Methods/Materials**

Stable transfections of Drosophila cells were used to knockdown/overexpress specific circRNAs. A double-stranded RNA transfection to knockdown the IMD pathway protein Relish in stably transfected cells was used to determine whether the circRNAs are dependent on Relish or not. qPCR was used to measure Diptericin mRNA levels and confirm the overexpression/knockdown of circRNAs, as well as Relish knockdown. Furthermore, immunoblot was used to observe Relish cleavage in cells induced/not induced with PGN. Gal4 driver lines were also crossed with shRNA fly lines to observe circRNA functions in vivo.

**Results**

qPCR of stably transfected cells showed that select circRNAs led to decrease in Diptericin mRNA levels when knocked down and increase when overexpressed. Furthermore, dsRNA transfections showed great decrease in Diptericin mRNA in experimental groups when compared to control. Immunoblot revealed increased Relish cleavage in cells with circRNAs overexpressed, and decreased Relish cleavage in cells with circRNAs knocked down. In vivo, similar findings were observed for Diptericin mRNA levels. Furthermore, when certain circRNAs were knocked down in specific tissues (muscle, neuron, female fat body, etc.), phenotypic effects were observed, including lethality and impaired mobility.

**Conclusions/Discussion**

Select circRNAs positively regulate innate immunity in Drosophila, functioning upstream of Relish. Furthermore, they may be required for proper neuron and muscle function, and even for fly survival. This research may shed light on the underlying molecular mechanism of the human innate immune system as well as the function of circular RNAs in humans. The findings of this study also have implications towards neurodegenerative diseases, as the IMD pathway has been shown to be involved in neurodegeneration in Drosophila.

**Summary Statement**

My work has demonstrated that a few novel circular RNAs positively regulate the IMD innate immunity pathway in Drosophila upstream of the protein Relish and play tissue-specific roles in vivo in Drosophila.

**Help Received**

Dr. Rui Zhou for mentoring and training me in basic lab techniques, equipment handling and data analysis as well as in vivo fly work; Dr. Xiao-Peng Xiong for helping me with the use of GraphPad Prism; Mr. Ariel Haas for supporting and mentoring me.