Role of Circular RNAs in Drosophila Innate Immunity

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Circular RNAs (circRNAs) constitute a class of relatively unstudied RNAs whose function is unknown. Recently, studies have shown that circRNAs are more abundant than previously recorded. There is no current established relationship between circRNAs and the Drosophila IMD innate immunity pathway, which is highly conserved between Drosophila and humans. The goal of this project is to determine if circRNAs play a role in the Drosophila IMD pathway to gain insights on the possible function of circRNAs in humans. Preliminary experiments were conducted to validate circRNA candidates using PCR, gel electrophoresis, and Sanger sequencing. Then, transfections were conducted to knock-down or overexpress individual circRNAs in cells that were PGN-treated and -untreated, and a luciferase reporter assay was used to measure the IMD pathway activity in each group. Finally, qPCR was used to confirm that circRNAs were successfully knocked-down or overexpressed. During the experiment, 14 of 15 original circRNA candidates were validated. Furthermore, through the luciferase reporter assay, two circRNAs (circ_1709 and circ_2465) showed a phenotype when mis-expressed. qPCR confirmed the significant overexpression of circ_1709 in transfected cells and insignificant change in the linear_1709 levels. The principle conclusion reached is that the two novel circular RNAs have an impact on the IMD innate immunity pathway in Drosophila, which may shed light on the underlying molecular mechanism of the human innate immune system.