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## Genetic Annotation and Evolutionary Rate of CHICO in D. rhopaloa and D. miranda

The insulin signaling pathway plays a major role in the body, through helping insulin regulate homeostasis in blood glucose levels, as well as in metabolism. This pathway involves a variety of proteins that act at various steps during the signal transduction process, with our research focusing on the evolution of these various proteins in relation to their positions along the pathway. The fruit fly species *Drosophila melanogaster* was used as the reference species since the entire genome had already been annotated. A comparative study was done in analyzing the evolution of the CHICO gene, a gene that acts primarily in the beginning of the insulin signaling pathway. Three different *Drosophila* species were studied, including *D. melanogaster* (the reference), D. rhopaloa and D. miranda. Evolutionary rates are directly impacted by the location of the gene in the insulin signaling pathway, and the phylogenetic distance of the Drosophila species with each other. Our first experimental goal was to annotate CHICO in these target species, then align the encoded amino acid sequences and analyze synteny (the colocalization of genes on the same chromosome in different species) and genomic neighborhoods. Lastly, compare these sequences with D. melanogaster using the online program MEGA X to perform sequence alignments. This analysis showed that after annotating these genes and determining their encoding sequences, we were able to retrieve the amino acid sequence of CHICO in these species and measure their evolutionary rates through comparing the number of amino acid substitutions with each other. D. rhopaloa had a lower evolutionary rate and had direct matches (in genomic neighborhoods) to D. melanogaster, whereas D. miranda had a higher evolutionary rate, which supports the hypothesis that the closer two fruit fly species are to each other, the smaller their evolutionary rates will be.