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Funding Source: Life Sciences Undergraduate Research Opportunity Program

Yeast two-hybrid screen of PGAM5

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PGAM5 is a member of the phosphoglycerate mutase family. It has been identified as a substrate for BTB-Kelch protein Keap1, a substrate adaptor for a cullin-based ubiquitin ligase. Keap1 plays an important role in cellular redox homeostasis through negative regulation of transcription factor Nrf2. Recent evidence suggests that PGAM5, Keap1 and Nrf2 may form a ternary complex that contributes to the regulation of anti-oxidant gene expression. In order to understand the role of PGAM5, we will identify its interacting partners through yeast-two hybrid screening. Fusions of the binding domain and four constructs of PGAM5 were cloned and subsequently transformed into yeast cells. These fusions were tested to ensure that they do not autonomously activate reporter genes, which suggests protein interaction. The yeast containing the fusions will be mated with a library of proteins fused to the activation domain. The progeny that activate reporter genes will suggest novel interacting protein partners for PGAM5.