Glycerol Kinase Overexpression Induces Metabolic Flux Alterations, Lipid Accumulation, and Increased Glucocorticoid Receptor Transcription Factor Activity In Rat Hepatoma Cells. A Systems Biology Study

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Glycerol kinase (GK) is an important lipogenic enzyme in liver, and exhibits several diverse ("moonlighting") cellular functions. GK overexpression has implications on insulin sensitivity, and GK deficiency is a single gene, yet complex genetic disorder whose complexities cannot be explained simply by lack of GK's biochemical activity. In the light of GK's multifaceted role, it is motivating to examine how perturbations in GK levels affect mammalian metabolic and regulatory networks. Therefore, we investigated metabolic flux, transcriptome, and transcription factor activity alterations caused by GK overexpression in a H4IIE rat hepatoma cell line.

13C labeling-based metabolic flux analysis revealed that GK-overexpressing cells had significantly higher flux through the pentose phosphate pathway (2.04 ± 0.10-fold) as compared to the wild type ($p < 0.01$). Comparable increases were also observed in the mRNA level and enzyme activity of glucose-6-phosphate dehydrogenase, the rate-limiting enzyme of this pathway, thus substantiating the flux analysis. cDNA microarray analysis of the GK-overexpressing and wild type cells revealed that GK overexpression alters the expression of several metabolic genes consistent with the observed metabolic flux changes. Lipid metabolism genes were differentially regulated in GK-overexpressing cells; this finding was corroborated by Oil Red O staining, which revealed larger lipid reserves in the GK-overexpressing cells.

We combined the microarray data with network component analysis to deduce that GK overexpression affected nine transcription factor activities, including that of the glucocorticoid-glucocorticoid receptor complex (GR). This result is particularly interesting because GR transcriptional activity is associated with GK's moonlighting role as ATP-stimulated translocation promoter. We further performed a glucocorticoid dose response experiment, which verified that the transcriptional activity of glucocorticoid receptor was indeed higher in the GK-overexpressing cells.

Thus, by using systems-level analyses, this work elucidates GK's central role in mammalian metabolism and transcription, and provides insights toward better understanding GK deficiency, a complex, yet single-gene inherited disease.