ABSTRACT

TITLE:	Cloning the Triose Phosphate Isomerase Gene from A. Cap to E. Coli
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ABSTRACT: The gene tpiA was chosen from Acetobacter capulatum to be cloned in Escherichia coli. The genomic DNA was removed from the bacteria and purified using a Quigen DNeasy 96 kit. The desired gene was found on KEGG, the Kyoto Encyclopedia of Genes and Genomes. Using the DNA sequence provided by KEGG foreward and reverse primers were designed for PCR. Through polymerase chain reaction, tpiA was replicated to usably concentrations. Gel electrophoresis confirmed the presence of amplicons approximately the desired size. The amplicons were then attached to a plasmid vector containing a gene for ampicillin resistance. The resulting plasmid was "fed" to the E. coli. We selected E. coli with the plasmid to grow colonies on ampicillin-containing agar. Colonies were taken from the agar and grown in LB broth. The E. coli were tested for the tpiA gene by removal of the DNA and isolating the plasmid for use in another gel electrophoresis. After the presence of tpiA was confirmed, expression of triose phosphate isomerase was tested for. Rapid expression of the gene was induced. TPI will be isolated and tested using various protein and kinetics assays. This will hopefully show beyond doubt that the gene tpiA was successfully taken from A. cap and not only cloned into E. coli, but expressed.