MM-GBSA routine to explore the contribution of interface residues to the overall stability of the tetramer. Compared to the wildtype controls, Q11N had the least impact on the various tetramer interfaces while I15Q appeared to significantly strengthen the A-B interface (deltaGo = −176.1+/−1.7 kCals/mol vs wildtype = −165.2+/−1.5 kCals/mol) but had the weakest A-D interface. Overall, these results support our hypothesis concerning the importance of the local polarity around the conserved E18 residue in *Plasmodium falciparum* MDH, and form the basis for future studies aimed at exploring possible targets on *Plasmodium falciparum* MDH for potential drug design as well as providing basic information about the mechanisms of subunit interactions in oligomeric proteins.

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Biochemical characterization of protein 3r8e as a novel glucose kinase

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The Protein Data Bank (PDB) contains over 198,000 experimentally determined protein structures, approximately 4300 of which have not been assigned a specific function. One such protein is PDB ID 3r8e from *Cytophaga hutchisonii*, a potential kinase with a solved structure but no confirmed function. Utilizing modules from the Biochemistry Authentic Scientific Inquiry Laboratory (BASIL) consortium, we first used a range of in silico tools to analyze 3r8e. These online tools included BLASTp, Pfam, and DALI. Together the in silico results indicated that 3r8e could be a glucose kinase. Next, molecular docking was used to explore whether glucose was an appropriate substrate for 3r8e. Glucose and five other sugars were docked into 3r8e along with ATP. Although all the sugars could be docked, glucose had the best fit for the active site. The 3r8e protein was then overexpressed in *E. coli* and purified using nickel affinity chromatography. Protein purity was assessed using SDS PAGE analysis. The purified protein was subsequently used in coupled kinase assays to determine the specific activity of 3r8e for the six different sugar substrates. A high specific activity was seen for 3r8e with glucose as the kinase substrate. Little to no activity was seen using fructose, galactose, lactose, ribose, or sucrose. In summary, online tools, molecular docking, and coupled kinase assays using purified protein demonstrate that 3r8e is a glucose kinase.

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