

Solubilization and Refolding of Plasmodium BEM-46-Like Protein (PBLP) from Inclusion Bodies

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Plasmodium BEM-46-like protein (PBLP) is found in the *Plasmodium yoelii* parasites that cause malaria in rodents, and knocking out PBLP has been shown to reduce *Plasmodium* infectivity. PBLP contains an α/β hydrolase domain and is predicted to act as a lipid hydrolase. This study investigates the biological function of PBLP and its target substrates, with the potential to inform development of future malaria treatments, given that treatment and cures remain limited. We expressed PBLP in *E. coli* and sought to isolate it to examine for hydrolase activity. PBLP was initially recovered in the insoluble pellet after cell lysis, so a new protocol was developed and tested to solubilize PBLP from inclusion bodies using urea and other reagents, along with a series of centrifugation and dialysis steps. PBLP was successfully solubilized and recovered in the supernatant following the new protocol; however, SDS-PAGE analysis indicated that a relatively low concentration of PBLP was present in the final concentrated protein sample. To identify potential substrates, structural modeling was used to predict the structural interactions between PBLP's catalytic triad and various molecules. Enzyme activity assays demonstrated that the purified PBLP has hydrolase activity toward PNP-acetate and PNP-butyrate. For future work, additional p-nitrophenyl substrates can be tested, and a more efficient protein extraction protocol can be developed since PBLP yields were low. These findings help establish an effective approach to recover PBLP from inclusion bodies and provide evidence of its hydrolase activity, which may aid in the development of PBLP-targeting drugs.