

METABOLIC ENGINEERED *ESCHERICHIA COLI* TO ENHANCE POLYHYDROXYALKANOATES PRODUCTION

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Abstract: Polyhydroxyalkanoates (PHAs) are linear polyesters produced through fermentation of sugar or lipid. Biosynthesis of PHA consists of three enzymes which are acetyl-CoA acetyltransferase (*phaA*), acetoacetyl-CoA reductase (*phaB*) and PHA synthase (*phaC*). *Comamonas* sp. is one of the strains commonly used for PHA production. Under growth conditions PHA is synthesized by excess of carbon sources and other essential nutrients. In order to develop higher PHA production from bacterial respond strategy, PHA biosynthesis operon of *Comamonas* sp. EB172 was introduced into *Escherichia coli* BW25113 through pGEM-T vector. *E. coli* was chosen due to the complete genome information and the absence of depolymerization gene, *phaZ*. The presence of PHA operon in *E. coli* has yielded PHA about 46% (w/w) with glucose as the carbon source. Therefore, the aim of this study is to improve the PHA production by screening of specific genes related to metabolic pathway of PHA in *E. coli*. Single gene deletion strains of keio collection harboring PHA biosynthesis operon of *Comamonas* sp. EB172 were used. Six genes *pgi*, *frdC*, *fdnG*, *gltA*, *pta*, and *poxB* were found to be associated with PHA metabolism activity. Second genes knockouts were introduced in through P1 transduction in order to improve PHA production and *E. coli* BW25113 *frdC gltA::kan* was shown an improvement of PHA production 53% compared to the wild type.

Keywords: Polyhydroxyalkanoates, *Comamonas* sp. EB172, *E. coli* BW25113, genes knockouts