

INTISARI

Luciferaselike monooxygenase (LLM) adalah *flavine dependent monooxygenase enzyme* (FMO) yang memiliki aktivitas katalitik dan struktur menyerupai BVMO tipe II. Kelompok enzim ini terlibat dalam berbagai fungsi biologis seperti katabolisme, detoksifikasi, dan biosintesis. Pada penelitian sebelumnya, *Luciferase-like monooxygenase 2 (llm2)* telah berhasil dikloning dan diekspresikan pada *Escherichia coli* BL21 (DE3). Hasil ekspresi LLM2 masih dalam bentuk tidak terlarut. Pada penelitian ini, sekresi protein secara ekstraseluler melalui konstruksi fusi dengan *pelB leader sequence* dilakukan guna meningkatkan kelarutan LLM2. *Open reading frame* (ORF) LLM tipe 2 dikloning pada plasmid pET26b (+) yang memiliki *pelB leader sequence*. Plasmid rekombinan kemudian ditransformasikan ke dalam *Escherichia coli* BL21(DE3) dan diamati hasil ekspresi dan kelarutannya menggunakan SDS-PAGE. ORF LLM2 berhasil dikloning dalam pET26b(+) dan menunjukkan presentase similaritas 100% dengan *Priestia megaterium* strain PSA10 *luciferase-like monooxygenase 2*. Analisis ekspresi dan kelarutan menunjukkan bahwa ORF LLM2 yang berfusi dengan *pelB leader sequence* menghasilkan rekombinan LLM2 dengan berat molekul $\pm 40,9$ kDa dan peningkatan kelarutan protein rekombinan sebesar 14%.

Kata kunci : Ekspresi, Kloning, Kelarutan, LLM2, *pelB leader sequence*

ABSTRACT

Luciferase-like monooxygenase (LLM) is a flavin-dependent monooxygenase enzyme (FMO) that have exhibits catalytic activity and a structure resembling type II BVMOs. This enzyme group is involved in various biological functions, including catabolism, detoxification, and biosynthesis. In previous research, *Luciferase-like monooxygenase 2* (LLM2) was successfully cloned and expressed in *Escherichia coli* BL21 (DE3). The results of LLM2 expression are still in insoluble form. In this study, extracellular secretion of the protein was achieved through fusion construction with the pelB leader sequence to enhance the solubility of LLM2. Open reading frame (ORF) of LLM2 was cloned into the pET26b (+) plasmid which have pelB leader sequence. Recombinant plasmid was then transformed into *Escherichia coli* BL21(DE3). The expression and solubility were analyzed using SDS-PAGE. The ORF successfully cloned into pET26b(+) and showed have 100% similarity to *Priestia megaterium* strain PSA10 *luciferase-like monooxygenase 2*. Expression and solubility analyses demonstrated that the ORF of LLM2 fused with pelB leader sequence produced recombinant LLM2 with a molecular weight $\pm 40,9$ kDa and increase 14% of solubility.

Key words: Expression, Cloning, Solubility, LLM2, pelB leader sequence