

ABSTRACT

Yosa Prawiratama, 24020220140022, **Production of PET Plastic Degrading Enzyme (PET Hydrolase) under the Control of Respective T7 and P2069 Mutant Promoter on Defined Medium and Auto Induction Medium.** Under the guidance of Anto Budiharjo and Is Helianti.

One frequently used plastic that is difficult to degrade is PET (polyethylene terephthalate) which is difficult to degrade. PET can be degraded by the enzyme PETase (PET hydrolase). Various engineering to increase the production of PETase has been done. One engineering that needs to be tried is the development of recombinant PETase production by using the right gene promoter in an economically feasible medium. The P2069 mutant promoter derived from Bacillus pumilus is a candidate that needs to be tried to express PETase, because of its ability to encourage foreign recombinant genes to reach the highest expression. The objectives of this study include examining the function of the P2069 mutant promoter in expressing PETase in Escherichia coli. In addition, analysis of PETase expression under the control of the T7 promoter in Escherichia coli in media without IPTG inducer (defined medium and autoinduction medium). Then, the PETase activity and protein concentration under the control of P2069 mutant promoter with T7 promoter in various medium were compared. The gene expressed under the control of P2069 Mutant is IsPETase, while those expressed under T7 are IsPETase Wild Type (WT) and ICCG. The study was conducted on defined medium (LB and TB) and autoinduction (AIM and ZYM-5052) in Escherichia coli. Observations were made at 0, 4, 8, 12, 16, 20, and 24 hours including optical density, protein concentration by Bradford test, SDS PAGE visualization, and enzyme activity by esterase test. The results of this study showed that the P2069 Mutant promoter functioned in expressing PETase both in defined medium and autoinduction medium with the highest activity seen in AIM autoinduction medium at the 20th hour of 3.44 U/mL. PETase activity under T7 promoter control was highest in IsPETase WT samples in TB medium at 24 hours with 37.85 U/mL, while ICCG was seen in ZYM-5052 autoinduction medium at 20 hours with 27.47 U/mL. PETase enzyme activity was higher under the T7 promoter control compared to the P2069 Mutant promoter in all media tested.

Keywords : *Expression, Escherichia coli, ICCG, IsPETase Wild Type, PETase.*