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Application of Genetic Engineering in Commercial Enzyme Production: Expression of Recombinant Serine Protease from *Virgibacillus natechei*

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Abstract

Hydrolysis of fish waste protein is an effective and economical strategy for obtaining valuable products with diverse applications. Fish waste, often considered a byproduct, is rich in proteins and bioactive compounds that can be transformed into high-value products such as peptides and amino acids. In recent years, researchers have prioritized green and sustainable technologies for the hydrolysis of fish-derived proteins. These approaches aim to replace traditional chemical methods, which may pose environmental and health risks, with safer and more environmentally friendly alternatives. Among these, proteolytic enzymes have emerged as a promising solution, offering high specificity, efficiency, and mild operational conditions. However, natural enzymes face significant challenges, including high production costs, limiting their large-scale industrial application. To address this limitation, modern biotechnology tools, particularly genetic engineering, are being employed to produce recombinant enzymes with enhanced properties. In the present study, the synthetic gene encoding serine protease, an enzyme with considerable potential for protein hydrolysis, was inserted into the PET28a expression vector. This recombinant construct was then successfully introduced into the *E. coli* BL21(DE3) strain. The accuracy of the gene transfer was confirmed using Colony PCR, while the expression of the recombinant enzyme was evaluated through Real-Time PCR analysis. The successful production of serine protease from *Virgibacillus natechei* highlights its potential as a cost-effective alternative for various applications. If the purified enzyme demonstrates suitable biochemical properties, including

high activity and stability, further studies may establish its viability for commercial use in industries like fisheries, waste management, and protein recovery.

Key words: Gene Synthesis, Recombinant Protease, *E. coli* BL21(DE3), Real-Time PCR