

ENGINEERING YEAST SURFACE DISPLAY SYSTEM FOR EXPRESSION OF ESTERASE FROM *Geobacillus stearothermophilus*

Viktorija Kurpickaja¹, Arūnė Verbickaitė¹, Rasa Petraitytė-Burneikienė¹

¹Department of Eukaryote Gene Engineering, Institute of Biotechnology, Life Sciences Center, Vilnius University, Lithuania
viktorija.kurpickaja@gmc.stud.vu.lt

Yeast surface display system is a widely used protein engineering technology that enables the anchoring of recombinant proteins to the yeast cell wall. Our technique uses the α -agglutinin system, where the Aga1 and Aga2 subunits form disulfide bridges. By fusing the target protein with the Aga2 subunit, it can be effectively exposed on the yeast surface [1]. This approach has broad applications in biotechnology, including enzyme immobilization, biocatalysis, and industrial processes. It simplifies substrate accessibility and allows continuous enzymatic reactions. Moreover, this system reduces the risk of enzyme contamination in the final product, as the enzymes remain immobilized on yeast cells and are not released into the reaction mixture.

This study aimed to express and display the extremoenzyme esterase from *Geobacillus stearothermophilus* [2] on the surface of *Saccharomyces cerevisiae* using five distinct yeast expression vectors. Previously developed with different linker peptides between Aga2 and the target protein [3], these vectors were designed to optimize protein synthesis and enzymatic activity. Previous studies have demonstrated that lipase from mesophilic bacteria was successfully displayed on the yeast surface and had enzymatic activity [4]. These findings suggest that the yeast surface display system could be an effective platform for esterase displaying as well, since lipases and esterases belong to the same enzyme class. The constructed plasmids will be tested in further research to evaluate their enzyme display and activity efficiency. This study contributes to expanding the application of yeast display in biocatalysis and industrial enzyme production.

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