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Electrophoretic assessment of recombinant λ -exonuclease production in different *E. coli* strains

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Viral exonucleases play role in many processes essential for genome maintenance, including DNA repair and recombination. Lambda exonuclease (λ -exo), isolated from lambda bacteriophage, hydrolases double-stranded DNA (dsDNA) in the highly processive manner in 5'→3' direction, yielding mononucleotides and single -stranded DNA (ssDNA). This unique enzymatic properties offer several promising biotechnological applications, such as highly sensitive quantification of DNA modifications and single -molecule sequencing. Hence, optimization of the expression conditions is a prerequisite to achieve high-level production of λ -exo. Here we have tested λ -exo expression in five different *E. coli* strains under various temperature regimes in order to establish the optimal conditions for efficient production of recombinant λ -exo. The N-terminally His-tagged λ -exo was successfully expressed in *E. coli* BL21(AI), SHuffle T7, C41(DE3) and C43(DE3) strains in LB broth. Collected aliquots were analysed by SDS-PAGE, followed by CBB staining. Relative yield of target protein bands was determined by densitometry in total cell lysate, as well as in soluble and insoluble cytoplasmatic fractions. We identified *E. coli* BL21(AI), SHuffle T7 and C41(DE3) as good producers of recombinant λ -exo, and upon scaling up, λ -exo was purified from crude cell lysates by metal affinity chromatography in satisfactory yield. Our data suggest that densitometric analysis could serve as a powerful low-cost screening platform for improving recombinant protein expression strategies.

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