

## CONSTITUTIVE EXPRESSION OF RECOMBINANT CLUMPING FACTOR OF *STAPHYLOCOCCUS AUREUS* IN *ESCHERICHIA COLI*

Ivanova V.D., Pozur V.K.

National Taras Shevchenko University of Kiev, Ukraine  
e-mail: victdzani@yahoo.com

The choice of an expression system for the high-level production of recombinant proteins depends on many factors, such as cell growth characteristics, expression levels, it includes a cost breakdown of process. The aim of our study was the construction of the bacterial strain constitutively producing staphylococcal clumping factor (Clf) and determination of the Clf levels ratio in constitutive and inductive expression systems.

The earlier obtained fragment of the *clfA* gene encoding the Clf ligand-binding domain was cloned into the expression plasmid pQE30 (Qiagen) to generate construct pCF41. The introducing of the recombinant plasmid pCF41 into *E.coli* BL21(DE3)Gold cells, not carrying lac-repressor gene (*lacI<sup>r</sup>*) in the bacterial chromosome, provided constitutive expression of the protein of interest. The presupposition of the high-level production of the recombinant protein in cells of recipient strain is their *lon<sup>-</sup>* and *ompT<sup>-</sup>* characteristics.

The significant level of the recombinant protein constitutive biosynthesis was observed in BL21(DE3)Gold (pCF41) cells and it reached more than 20% of the total cell protein that comparable with its yield after IPTG induction in *E.coli* XLI-Blue (pCF41) cells. The advantage of constitutive expression system (its fermentation simplicity and cost-effectiveness) determines its priority utilization for high-level production of the recombinant Clf. The Clf was localized either in the soluble or in the insoluble cell fractions in the proportion of approximately 5:1. Recombinant protein binds to anti-Clf antibodies and to fibrinogen. The high specificity of the protein provides enabling its use in ELISA to detect the anti-Clf-antibodies in patients with staphylococcal infections.