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EFFECTIVE EXPRESSION OF RECOMBINANT STAPHYLOCOCCUS AUREUS CLUMPING FACTOR IN E.COLI CELLS AND IT IMMUNOBIOLOGICAL ACTIVITY

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Fragment of the *clfA* gene encoding the ligand-binding domain of S.aureus clumping factor was cloned into the expression plasmid pQE30 (to generate the construct pCF41) in E.coli XLI-Blue cells. We demonstrated instability of expression construct, the elimination of the plasmid and the reduction of the strain efficiency during the cultivation of a producer. To raise stability of plasmid pCF41 and increase the yield of recombinant protein cultivation conditions were selected. The approaches were suggested to maintain considerable amount of plasmid-containing cells in population: (1) preliminary screening of highly productive clones, which produce not less than 18-20 percent of recombinant protein and (2) their cultivation in the presence of high levels of ampicillin (200 „g/ml) that added each two hours during growth of cells (but an hour later after IPTG induction). The cultivation of strain on growth medium containing buffered basis M9 with glucose (1%), yeast extract (0,5%) and casein hydrolysate (1%) increases the level of he recombinant polypeptide biosynthesis and it reached to 30% of the otal cell proteins 4h after induction (we found the yield of recombinant rotein by scanning of SDS-PAGE gels and analysing them with PC rogram Densitometer). The clumping factor, which is expressed in .coli XLI-Blue, was localized either in the soluble (22% of the total cell roteins) or in the unsoluble (8% of the total cell proteins) cell fractions. rotein contains an N-terminal extension of six histidine residues (His ıg), which allows purification on IDA-Sepharose-6B with high yield 24mg from 1l of culture). Recombinant protein binds to anti-ClfA ıtibodies, to fibrinogen immobilized in an ELISA plates. We used combinant antigen for analysis of the serum antibody responses jainst native clumping factor in patients with staphylococcal spiratory tract infections by enzyme immunoassay (EIA). The study s shown that antibodies against Clf are present in various extents in e healthy individuals serum and that increased levels are observed ring infection in many cases.