عنوان مقاله:

Recombinant Production and Affinity Purification of the Mature Form of Staphylolysin (LasA) Protein in E.coli

محل انتشار:

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خلاصه مقاله:

Background and Aims: Enzybiotics are probably the future line of weapons against drug resistant bacteria. They lyse bacteria with the new mechanisms with few likelihood of generating resistance. LasA, which is secreted from Pseudomonas strains degrades Staphylococcus aureus (S. aureus) cell wall and has the potential to use against drug resistant S. aureus infections. Materials and Methods: Codon-optimized gene of the mature form of the LasA protein was ordered. The gene was double digested with NcoI and XhoI restriction enzymes and sub-cloned into pET۲۸a(+) digested with the same enzymes. Recombinant construct was introduced into BL۲۱ DE۳ cell. Expression of the gene was induced by ۰.۲ mM isopropyl β-D-۱-thiogalactopyranoside and recombinant protein was affinity purified by Ni-NTA mini-column. The staphylolytic activity of the recombinant LasA protein was evaluated on Methicillin-resistant S. aureus (MRSA) by disk diffusion. Results: Fragment of the LasA gene encoding mature form of the LasA protein was introduced into pET۲۸a(+) expression vector. C-terminal his-tagged recombinant LasA protein was produced in BL۲۱ DE۳ E. coli cells. Over ۵۰% purity has been achieved by affinity purification of the LasA protein from the total cell lysate. The yield of purified protein was ۵.۴ mg.l-۱. Growth of MRSA was completely inhibited by dilutions of recombinant his-tagged LasA. Conclusions: The present study shows that the mature form of the LasA can be expressed in E. coli BL۲۱ DE۳ cells. C-terminal his-tagged form of the mature LasA protein has staphylolytic activity .against MRSA and so it can be a promising therapeutic agent

کلمات کلیدی:
LasA, Methicillin-resistant, Staphylococcus aureus, Staphylolysin