

PROTEIN EXPRESSION AND PURIFICATION OF A MOLECULAR CHAPERON IN ASSOCIATION WITH HEAT STRESS TOLERANCE IN BACILLUS SUBTILIS (D18) ISOLATED FROM HOT SPRING OF INDIA

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ABSTRACT

A thermo tolerant bacterial strain was isolated from hot spring of Sohna, Haryana, India, classified and named as B.subtilisD18 after morphological and 16S rRNA gene sequence analysis. This isolate was used to amplify groEL gene, which encodes molecular chaperon GroEL. The full length gene was 1.6 kb in length encoding a polypeptide of 108amino acid residues. The calculated molecular weight and pI of the protein were nearly 60 kDa and 4.75, respectively. The amino acid sequence of the gene was similar to other groEL proteins and the homologous groEL of different microorganisms. The groEL gene of B. subtilis was successfully expressed in Escherichia coliBL21 (DE3) strain using pET expression systems and purified by polyhistidine tag using Ni-NTA (nitrilotriacetic acid) resin column. Heterologous expression of groEL of B.subtilis in E. coli BL21(DE3) allows the growth of E.coliup to 42°C for 16 h, suggesting that groEL from B.subtilis imparts tolerance to host cells under elevated temperatures.

KEYWORDS: Bacillus Subtilis, Molecular Chaperone, Expression Vector, Heterogonous Expression, His-Tag Purification

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