Research in Pharmaceutical Sciences, 2012;7(5)
School of Pharmacy and Pharmaceutical Sciences
Isfahan University of Medical Sciences

Investigation smta mutant gene expression in *E. coli* host

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Background and Aims: Heavy metal pollution is one of the most important environmental problems today. Various industries produce and discharge wastes containing different heavy metals into the environment. Metallothioneins (MTs) are small, cysteine rich proteins, which have been isolated from a wide range of eukaryotes, are involved in homeostasis of essential metal ions such as Zn2+, Fe2+ and Cu2+ and also in the detoxification of toxic metals like cadmium, copper, silver and mercury. This proteins bind metals, typically in metal-thiolate clusters and can neutralize an excess of heavy metals in the cell and protects cell structures against non-specific interactions with heavy metals. Today's, bacteria, plants and their protein production use in order to heavy metal removal. Bacterial metallothioneins (BmtA), exemplified by its prototype, SmtA from the cyanobacterium Synechococcus PCC 7942 is target of this study.
Methods: The plasmid pET15b containing the smtA mutant gene was transformed into competent E. coli BL21(DE3) cells according to standard procedures. A single colony was used to inoculate 10 ml LB medium containing Ampicilin (500 g/ml) at 37 C overnight.2 ml of this culture were added to 100 ml of fresh LB containing Ampicilin. Bacteria were grown at 37 C to OD600 = 0.5. Induction was achieved by addition of 1 mM IPTG. The expression of smtA mutant gene was verified by 12% SDS–PAGE of supernatant and pellet. After fractionation by SDS–PAGE, protein were transferred to membrane and the recombinant SmtA was characterized by immunoblot.
Results: SDS-PAGE illustrated that protein production is higher than control sample and immunoblot analysis was verified this.

Keywords: Metallothionein; smtA; Expression; IPTG