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Production of Recombinant Blue Fluorescent Protein (mTagBFP) for Further Researchs in Bioimaging

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Abstract

mTagBFP is a monomeric blue fluorescent protein generated by sitespecific and random mutagenesis of TagRFP [Subach et al. 2008]. TagBFP possesses bright blue fluorescence with excitation/emission maxima at 402 and 457 nm, characterized by high photostability and extremely high pH-stability. This work aimed to expression of mTagBFP in bioreactor in high yield using *E.coli* expression system and purification of it.

Transformed *E.coli* cells with pBAD- mTagBFP recombinant plasmid were cultured in 3 liters LB triple medium supplemented with 100 ug/ml ampicillin at 37 °C in bioreactor. When the optical density at 600 nm was 1.5, L- arabinose was added to a final concentration of %0.04 in order to express mTagBFP. After 5 hours, the cells were harvested by centrifugation. Cell pellets were suspended in lysis buffer and disrupted by sonication. Soluble protein was collected using ultra-centrifugation. 6xHis-tag on the N-terminus protein used for purification of recombinant mTagBFP. The expression levels of mTagBFP was assessed using 10% (w/v) SDS-PAGE and UV spectroscopy.

One of the most efficient expression systems for producing recombinant proteins in *E.coli* is a pBAD- system. It is observed that at optimized arabinose concentration (0.04 %) for 5 hours induction resulted high levels of fluorescent protein expression. The method relies on induced expression in the BL21-AI strain of *E.coli* and yields large amounts (20 mg/L) of fluorescent protein from a 3 liters culture. This method provides a quick, high-yield production and can be used to produce any fluorescent protein that is needed in biomedical research especially bioimaging.

Keywords: mTagBFP, Recombinant Protein, Blue Protein

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Reference: Subach OM et al. (2008) Conversion of Red Fluorescent Protein into a Bright Blue Probe. Chemistry & Biology 15(10): 1116–1124.