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Data Article

Recombinant protein production data after expression in the bacterium *Escherichia coli*



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ABSTRACT

Fusion proteins have become essential for the expression and purification of recombinant proteins in *Escherichia coli*. The metalbinding protein CusF has shown several features that make it an attractive fusion protein and affinity tag: "Expression and purification of recombinant proteins in *Escherichia coli* tagged with the metalbinding protein CusF" (Cantu-Bustos et al., 2016 [1]). Here we present accompanying data from protein expression experiments; we tested different protein tags, temperatures, expression times, cellular compartments, and concentrations of inducer in order to obtain soluble protein and low formation of inclusion bodies. Additionally, we present data from the purification of the green fluorescent protein (GFP) tagged with CusF, using Ag(1) metal affinity chromatography.

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Specifications table

Subject areaMolecular BiologyMore specific
subject areaProtein expression and purification

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Type of data How data was acquired	Pictures. Images of gel electrophoresis. Sodium dodecyl sulfate polyacrylamide gel electrophoresis (SDS-PAGE)
Data format	Raw
Experimental factors	Fusion proteins: GST, CusF, and SmbP. Inducer concentration, time and tem- perature during protein expression. Metal affinity chromatography with silver ions Ag(I). Periplasmic or cytoplasmic protein expression.
Experimental features	Production levels of soluble protein and inclusion bodies, protein purity.
Data source location	San Nicolas de los Garza, Nuevo Leon, Mexico
Data accessibility	Data is within this article

Value of the data

- The data shows that the production of soluble recombinant proteins in the bacterium *Escherichia coli* can be improved by varying the fusion protein, time, temperature, and concentration of inducer during protein expression.
- The data indicates that recombinant proteins, exemplified here with the red fluorescent protein (RFP), can be expressed in the cellular periplasm when tagged with full-length CusF and SmbP.
- Protein purification data shows that CusF-tagged proteins may be purified using immobilized metal affinity chromatography with Ag(I) ions instead of the most common Cu(II) or Ni(II).

1. Data

Fig. 1 shows the SDS-PAGE analysis comparing soluble and insoluble protein content for the proteins LovR and SHY2 tagged with two different protein tags and expressed at different conditions. Additionally, the electrophoretic analysis in Fig. 2 compares the content of inclusion bodies.

Recombinant proteins tagged with the protein tags CusFp and SmbPp, containing their signal sequences, are exported to the cell's periplasm. Fig. 3 shows an image of *E. coli* BL21(DE3) cells after RFP expression and the electrophoretic analysis of the periplasmic lysates.

Fig. 4 shows pictures of the synthesized silver chromatographic media before and after incubation with the *E. coli* lysate expressing green fluorescent protein tagged with CusF. Fig. 5 shows the SDS-PAGE analysis of the purification steps, it shows the protein content in the flow-through (the lysate after incubation with the Ag(I) resin), and two elutions steps with 160 mM methionine.

2. Experimental design, materials and methods

2.1. DNA constructs

Full-length CusF (CusFp, for periplasmic expression) was amplified with primers 5'-AGTCAGTCA-CATATGAAAAAAGCACTGCAAGTCG-3' (NdeI, forward) and 5'-ATGCATGCAGGTACCCTGGCTGACTT-TAATATCCTGTAA-3' (KpnI, reverse). The 50 μ L reaction comprised 10 ng of template DNA, 60 pmol of each primer, 1.5 μ L of 10 mM dNTPs mix, and 2 units of Vent DNA polymerase (New England Biolabs) in 1 × ThermoPol reaction buffer. The thermocycler conditions were 95 °C for 2 min; 30 cycles of 95 °C-1 min, 59 °C-1 min, 72 °C-1 min; and a final extension at 72 °C for 10 min. Amplification of CusF lacking the signal sequence (for cytoplasmic expression) was done with forward primer 5'-AGTCAGTCACATATGGCTAACGAACATCATCATGAAAC-3' (NdeI) and the same reverse primer and thermocycler conditions as before. pET30a vector was linearized with NdeI and KpnI, and the CusF

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