



## Eco™ Buster E Coli Protein Extraction User Manual

Product Name	Cat#	Amount
Eco™ Buster Small Protein Extraction	<b>EB-S100</b>	100 ml
Eco™ Buster Small Protein Extraction	<b>EB-S500</b>	500 ml
Eco™ Buster Large Protein Extraction	<b>EB-L100</b>	100 ml
Eco™ Buster Large Protein Extraction	<b>EB-L500</b>	500 ml

### Storage:

Although stable at room temperature for a long time, Product is recommended to store at 4°C, should stable for 6 month.

### Product Description and Composition:

GenTarget's Eco™ Buster E Coli protein extraction reagent is proprietary formulated for efficient extraction of active protein from E. coli. without mechanical lysis. It is nonionic detergent-based lysis (in Tris-HCl, PH7.5), and much better preserve the activity of the proteins than the harsh mechanical lysis, such as grinding or sonication. It is compatible with most downstream processes and functional assays (His-tag, GST-tag, MBP-tag affinity column, and other in-gel detection, etc), and its performance is not affected by the addition of protease inhibitors, salts or other components (such as EDTA, DTT etc). Compared to other brand lysis reagent, it is more efficient in lysis, and better preserves protein's activities, enabling to obtain higher yield of active, soluble protein. It is suitable for extracting proteins from wide range of bacterial strains.

Eco™ Buster protein extraction reagents are provided in Ready-to-use, single solution configuration. If your protein is greater than 75 kDa, use **Eco™ Buster Large Protein Extraction (Cat# EB-L100 / EB-L500)** which greatly increases the yields for large proteins. For protein smaller than 75 kDa, should use regular **Eco™ Buster Small Protein Extraction (Cat# EB-S100 / EB-500)**. [Note: Eco™ Buster Large Protein Extraction can be used for extracting small proteins, but not recommended since we didn't see any benefits, instead increases the viscosity of the lysate, compared to regular **Eco™ Buster E Coli Protein Extraction.**]



## Product warrantee:

Each batch of Eco™ Buster E Coli protein extraction was tested for ensure its high performance. Below figure showed the typical lysis results compared to leading competitor.

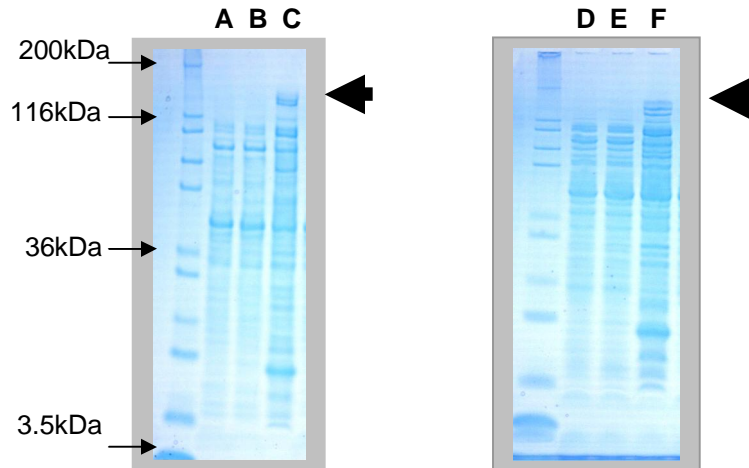


Fig.1: 1ml cells (OD=1.5) were lysated with 100ul of different reagents, then 10ul of each lysate was loaded into SDS protein gel. The left panel: BL21(DE3) cell lysates run in 4-12% Bis-Tris gel, the right panel: BL21(Star) cell lysates run in 10% Bis-Tris gel. Lane A and D: competitor's BB extracting reagent; Lane B and E: Eco™ Buster small protein extract reagent (Cat# EB-S100); Lane C and F: Eco™ Buster large protein extract reagent (Cat# EB-L100).

## Application:

Used for extracting active proteins from bacterial total cell lysate.

## Protein extraction protocols:

1. Grow cell (e.g. after induction) to OD 1~2, Collect cells by centrifugation at 4000g, Discard the supernatant;
2. To the cell pellet, add protein extraction solution (Cat# EB-S100 or Cat# EB-L100) at 50ul/per OD, or 5ul/per mg wet-pellet, or 50ul~100ul / per ml original culture, vortex until the pellet is re-suspended; If desired, add protease inhibitors here.
3. Incubate at RT for 15 minutes with gentle inverting the tube periodically, but no shaking;
4. Centrifuge at 10,000g for 20min at 4°C, transfer the supernatant for downstream purification, such as column fractionation or Ni-NTA affinity column purification, etc.



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5. **Optional:** If protein is expressed in inclusion body, resuspend the pellet from step 4 into appropriate amount of extraction solution and incubate at RT for 30 minutes with shaking, centrifuge to get the supernatant for downstream renaturing process, repeat Step 5 for multiple times until all inclusion body lysis.
6. If desired, load 10ul of lysate into mini-protein gel to check the expressed protein.