

B-PER[®] Bacterial Protein Extraction Reagent

78243 78248 78260 90078 90079 90084

0736.4

Number	Description
90078	B-PER Bacterial Protein Extraction Reagent with Enzymes Kit Contents: B-PER Bacterial Protein Extraction Reagent, 250mL Lysozyme, 0.5mL supplied at 50mg/mL DNase I, 0.5mL supplied at 2500U/mL
90079	B-PER Bacterial Protein Extraction Reagent with Enzymes Kit Contents: B-PER Bacterial Protein Extraction Reagent, 500mL Lysozyme, 2 × 0.5mL supplied at 50mg/mL DNase I, 2 × 0.5mL supplied at 2500U/mL
78243	B-PER Bacterial Protein Extraction Reagent, 165mL
90084	B-PER Bacterial Protein Extraction Reagent, 250mL
78248	B-PER Bacterial Protein Extraction Reagent, 500mL
78260	B-PER II Bacterial Protein Extraction Reagent, 250mL

Composition: Solution contains a proprietary, nonionic detergent in 20mM Tris•HCl; pH 7.5

Storage: Upon receipt store B-PER Reagent at ambient temperature and Lysozyme and DNase I at -20°C. Product #90078 and 90079 are shipped with ice packs. All others are shipped at ambient temperature.

Introduction

The Thermo Scientific B-PER Bacterial Protein Extraction Reagent enables mild extraction of proteins from bacteria (*E. coli*) without the need for mechanical disruption. The reagent may be used for soluble protein extraction and inclusion body purification from bacterial cell lysates. The B-PER Reagent with Enzymes is supplied with lysozyme and DNase I to improve the extraction efficiency of large (> 70kDa) molecular weight proteins and proteins expressed in inclusion bodies. Depending on the particular application, additional components, such as protease inhibitors, salts, reducing agents and chelating agents may be added to the reagent (add chelating agents only when using the B-PER Reagent without enzymes). The B-PER II Reagent is twice as concentrated as the B-PER Reagent and designed for small volumes of lysate so the protein remains concentrated.

Important Product Information

- The B-PER Reagent extracts proteins from recently prepared cells and frozen cells. Extraction is typically more effective with frozen cells. The amount of reagent required depends on the amount of cell paste.
- If desired, add EDTA-free protease inhibitors (Product No. 78425, 78437 or 78439), salts, and reducing agents directly to the reagent. Chelating agents such as EDTA can be used with B-PER Reagent without enzymes.

- Add lysozyme and DNase I to B-PER reagent for the most efficient extraction; however, the extraction of some over-expressed proteins does not require the addition of lysozyme. If the addition of lysozyme and DNase I might interfere with the downstream application, the use of these enzymes should be omitted.
- Some over-expressed proteins can be insoluble, misfolded or expressed in inclusion bodies. The best way to obtain soluble protein is to adjust the expression conditions; however, if this is not possible, the Thermo Scientific Inclusion Body Solubilization Reagent (Product No. 78115) can be used.
- B-PER Reagent effectively extracts soluble proteins from several common bacterial host strains and is especially suitable for the protease-defective expression host BL21 strains. If lysis is inefficient for a particular bacterial strain, freeze cells before extraction.
- Whole cell lysates prepared with the B-PER Reagent are compatible with the Thermo Scientific Pierce BCA and Coomassie (Bradford) Protein Assays (Product No. 23225 and 23236, respectively).

Procedure for Extracting Protein from Bacteria

1. Pellet bacterial cells by centrifugation at $5000 \times g$ for 10 minutes.
2. Optional: Add 2 μ L of lysozyme and 2 μ L of DNase I per 1mL of B-PER Reagent. Add EDTA-free protease inhibitors.
3. Add 4mL of B-PER Reagent per gram of cell pellet. Pipette the suspension up and down until it is homogeneous.
Note: If using the B-PER II Reagent, 2mL of reagent per gram of cell pellet may be used to achieve a more concentrated protein solution.
4. Incubate 10-15 minutes at room temperature.
5. Centrifuge lysate at $15,000 \times g$ for 5 minutes to separate soluble proteins from the insoluble proteins.
Note: If a large percentage of over-expressed protein remains in the pellet, the protein of interest might be expressed in inclusion bodies. Either use the Inclusion Body Solubilization Reagent (Product No. 78115) or alter the expression conditions to minimize inclusion body formation.

Troubleshooting

Problem	Possible Cause	Solution
Protein of interest is not solubilized	Protein of interest is expressed in inclusion bodies	Adjust expression conditions or use an inclusion body solubilization reagent
		Add lysozyme and DNase I to the B-PER Reagent
Bacteria are not lysed upon enzyme addition	Temperature is too low	Warm culture and other reagents to room temperature
	Lysozyme has low activity or is inactive	Add more lysozyme to the B-PER Reagent or purchase new enzyme
Viscosity of extract is high	DNase I has low activity or is inactive	Add more DNase I to the B-PER Reagent or purchase new enzyme
		Add Mg ²⁺ ions to 2mM final concentration
Excessive over-expressed protein remains in the pellet	Protein is too large for B-PER Reagent alone	Add lysozyme and DNase I to the B-PER Reagent
B-PER Reagent is cloudy after shipping	Reagent is cold	Warm reagent to room temperature

Additional Information from Our Website

- Frequently Asked Questions
- Tech Tip #19: Remove detergent from protein samples
- Tech Tip #43: Protein stability and storage
- Tech Tip #6: Extinction coefficients guide

Related Thermo Scientific Products

90081	B-PER Direct Bacterial Protein Extraction Kit
89965	HisPur™ Cobalt Resin, 100mL
88222	HisPur Ni-NTA Resin, 100mL
87730	Slide-A-Lyzer® G2 Dialysis Cassettes, 10K MWCO/3mL, 10/pkg.
87731	Slide-A-Lyzer G2 Dialysis Cassettes, 10K MWCO/15mL, 8/pkg.
87786	Halt™ Protease Inhibitor Cocktail (100X), 1mL
87785	Halt Protease Inhibitor Cocktail, EDTA-Free (100X), 1mL
22660	Pierce® 660nm Protein Assay Reagent, 750mL
23227	Pierce BCA Protein Assay Kit
24594	GelCode® Blue Safe Protein Stain, 1L
24590	GelCode Blue Stain Reagent, 500mL, (coomassie G-250)
78990	Y-PER® Yeast Protein Extraction Reagent, 500mL
89802	I-PER® Insect Cell Protein Extraction Reagent, 250mL
90082	Lysozyme, 0.5mL supplied at 50mg/mL
90083	DNase I, 0.5mL supplied at 2500U/mL
78115	Inclusion Body Purification Reagent, 100mL
15160	Immobilized Glutathione Resin, 10mL
24615	Imperial™ Protein Stain, 1L, (coomassie R-250)

This product ("Product") is warranted to operate or perform substantially in conformance with published Product specifications in effect at the time of sale, as set forth in the Product documentation, specifications and/or accompanying package inserts ("Documentation") and to be free from defects in material and workmanship. Unless otherwise expressly authorized in writing, Products are supplied for research use only. No claim of suitability for use in applications regulated by FDA is made. The warranty provided herein is valid only when used by properly trained individuals. Unless otherwise stated in the Documentation, this warranty is limited to one year from date of shipment when the Product is subjected to normal, proper and intended usage. This warranty does not extend to anyone other than the original purchaser of the Product ("Buyer").

No other warranties, express or implied, are granted, including without limitation, implied warranties of merchantability, fitness for any particular purpose, or non infringement. Buyer's exclusive remedy for non-conforming Products during the warranty period is limited to replacement of or refund for the non-conforming Product(s).

There is no obligation to replace Products as the result of (i) accident, disaster or event of force majeure, (ii) misuse, fault or negligence of or by Buyer, (iii) use of the Products in a manner for which they were not designed, or (iv) improper storage and handling of the Products.

Current product instructions are available at www.thermoscientific.com/pierce. For a faxed copy, call 800-874-3723 or contact your local distributor.

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