Cell Lysis Buffer

Catalog number: AR0103

Boster’s Cell Lysis Buffer is a ready-to-use Western blot related reagent solution used for efficient extraction of total soluble protein in nondenatured state from mammalian cells.

This package insert must be read in its entirety before using this product. For research use only. Not for use in diagnostic procedures.
# Cell Lysis Buffer

**Catalog Number:** AR0103

## Overview

<table>
<thead>
<tr>
<th>Physical State</th>
<th>Liquid</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pack Size</td>
<td>100 mL (The kit contains sufficient lysis buffer for 200 cell pellet fractions containing $5 \times 10^6$ cells each)</td>
</tr>
<tr>
<td>Description</td>
<td>Boster's Cell Lysis Buffer is a ready-to-use Western blot related reagent solution used for efficient extraction of total soluble protein in non-denatured state from mammalian cells.</td>
</tr>
<tr>
<td>Storage &amp; Expiration</td>
<td>Upon receipt store at 4°C. Cell Lysis Buffer is stable for one year. Product is shipped on ice.</td>
</tr>
<tr>
<td>Equivalent</td>
<td>Thermofisher (Product No. 78501, 78503, 78505); Millipore Sigma (Product No. C2978)</td>
</tr>
<tr>
<td>Application</td>
<td>Immunoassays: WB, ELISA, RIA, immunoprecipitation; reporter assays; protein kinase assays; protein purifications</td>
</tr>
<tr>
<td></td>
<td>*Our <a href="#">Boster Guarantee</a> covers the use of this product in the above tested applications.</td>
</tr>
<tr>
<td>Enzymatic Activity</td>
<td>None; use additives to add function</td>
</tr>
<tr>
<td>Additional components compatibility</td>
<td>Compatible with additives, e.g. protease inhibitors (PMSF), reducing agents, chelating agents, salts</td>
</tr>
<tr>
<td>Cite This Product</td>
<td>Cell Lysis Buffer (Boster Biological Technology, Pleasanton CA, USA, Catalog # AR0103)</td>
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Description

This Cell Lysis Buffer is a ready-to-use Western blot related reagent solution used for efficient extraction of total soluble protein in nondenatured state from mammalian cells. Cell lysis can be finished in 30 minutes. The cell lysis buffer has been validated for use with COS-7, NIH3T3, Hepa 1-6, 293T, CHO and other cell types. It is able to lyse plated cells and pelleted cells from suspension mammalian cultures. The produced cell lysates are directly compatible with reporter gene expression assays (luciferase, beta-galactosidase, chloramphenicol acetyl transferase, CAT, alkaline phosphatase), protein kinase assays (PKA, PKC, tyrosine kinase), phosphatase assays (general phosphatases, tyrosine phosphatases), immunoassays (Western blots, ELISAs, RIAs, immunoprecipitation), Coomassie-Blue and silver staining, BCA protein assay, protein purification procedures, electrophoresis, and many other downstream applications.

Additional Materials Required

- Protease inhibitor (Product No. AR1182) and phosphatase inhibitor (Product No. AR1183)
- 2 ml microcentrifuge tubes
- Microcentrifuge capable of spinning at 10000 x g
- Cell Scraper

Procedure for Lysis of Monolayer-cultured Adherent Mammalian Cells

**Note:** Pre-chill an appropriate volume of Cell Lysis Buffer at 4°C. If desired, add protease inhibitor and phosphatase inhibitor to the lysis buffer immediately before use.

1. In a microcentrifuge tube, resuspend $5 \times 10^6$ cells in the growth media by scraping the cells off the surface of the plate with a cell scraper. Centrifuge harvested cell suspension at 600xg for 5 min, then carefully remove and discard the supernatant.
2. Resuspend the cells in chilled PBS. Centrifuge at 600xg for 5 min, then carefully remove and discard the supernatant.
3. Add 0.5 mL of chilled cell lysis buffer to the cell pellet. Vortex briefly. Incubate on ice for 30 minutes.
4. Centrifuge samples at 10000xg for 10 minutes.
5. Transfer supernatant to a new tube for further analysis.

**Note:** Cell lysis buffer can be added directly to the flask containing cells. Please see the following procedures.

1. Carefully remove culture medium from adherent cells.
2. Wash cells with chilled PBS. Carefully remove PBS.
3. Add chilled Cell lysis buffer to the cells. Vortex briefly. Incubate on ice for 30 minutes. (For the volume of the lysis buffer, follow the instructions listed below)

<table>
<thead>
<tr>
<th>SIZE of the plate/surface area</th>
<th>Volume of the lysis buffer</th>
</tr>
</thead>
<tbody>
<tr>
<td>100mm</td>
<td>500-1000μL</td>
</tr>
<tr>
<td>60mm</td>
<td>250-500μL</td>
</tr>
<tr>
<td>6-well plate</td>
<td>200-400μL per well</td>
</tr>
<tr>
<td>24-well plate</td>
<td>100-200μL per well</td>
</tr>
<tr>
<td>96-well plate</td>
<td>50-100μL per well</td>
</tr>
</tbody>
</table>
4. Centrifuge samples at 10000xg for 10 minutes.
5. Transfer supernatant to a new tube for further analysis.

**Procedure for Lysis of Suspension-cultured Mammalian Cells**

*Note: Pre-chill an appropriate volume of Cell Lysis Buffer at 4°C. If desired, add protease inhibitor and phosphatase inhibitor to the lysis buffer immediately before use.*

1. In a microcentrifuge tube, harvest 5×10^6 cells by centrifugation at 600xg for 5min. Carefully remove and discard the supernatant.
2. Resuspend the cells in chilled PBS. Centrifuge at 600xg for 5min, then carefully remove and discard the supernatant.
3. Add 0.5 mL of chilled Cell lysis buffer to the cell pellet. Vortex briefly. Incubate on ice for 30 minutes.
4. Centrifuge samples at 10000xg for 10 minutes.
5. Transfer supernatant to a new tube for further analysis.

**Precautions**

- All steps of protein lysis should be operated on ice or at 4 °C.
- Use BCA Protein Assay kit (Product No. AR0146) to quantify lysed proteins. Bradford Protein Assay kit is not recommended.

**Example Data using Cell Lysis Buffer**

![Graph showing protein yield](image)

**Figure 1. Cell lysis protein yield with Cell Lysis Buffer**

Proteins were extracted from different cells following the Cell Lysis Buffer protocol. The protein concentration of each lysate was determined by BCA Protein Assay Kit (Product No. AR0146) to determine protein yield per milligram of starting cells.
Figure 2. Protein extraction from various cells using Cell Lysis Buffer
Proteins were extracted from various cells following the Cell Lysis Buffer protocol. Lysates (20ug) were separated by SDS-PAGE and transferred to a nitrocellulose membrane. Incubate with primary antibodies. Images were generated using Boster Hypersensitive ECL chemiluminiscence substrate (Product No. AR1170).

Troubleshooting

<table>
<thead>
<tr>
<th>Problem</th>
<th>Possible Cause</th>
<th>Solution</th>
</tr>
</thead>
<tbody>
<tr>
<td>Low total protein yield</td>
<td>Some cells are more resistant to lysis than others</td>
<td>Make sure the cell pellet is thoroughly suspended in Cell Lysis Buffer</td>
</tr>
<tr>
<td></td>
<td></td>
<td>and incubate for longer with occasional swirling – sonicate the pellet to increase yield</td>
</tr>
<tr>
<td>Low concentration of proteins</td>
<td>Excess buffer used</td>
<td>Use less buffer</td>
</tr>
</tbody>
</table>

Related Boster Products

AR1182  Broad Spectrum Protease Inhibitor Cocktail
AR1183  Broad Spectrum Phosphatase Inhibitor Cocktail
AR0146  BCA Protein Assay Kit
AR0138  SDS-PAGE Gel Preparation Kit
AR1170  Hypersensitive ECL Chemiluminiscence Substrate