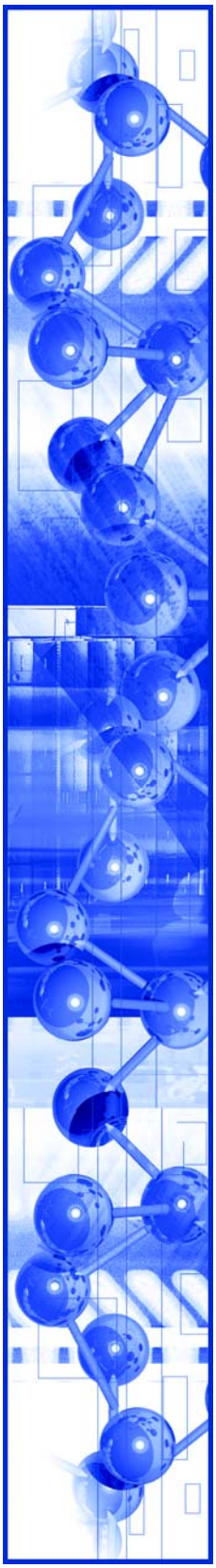

Total Protein Cell Lysis Buffer

Code	Description	Size
M329-10ML	Total Cell Lysis Buffer *For Mammalian Tissue Culture Cells* *Buffer is sufficient for 20 protein extractions of $1x10^6$ cells*	10 mls

General Information:

AMRESCO's Total Protein Cell Lysis Buffer is a non-ionic detergent lysis buffer optimized for the isolation of total proteins from adherent or non adherent cells in culture. The Total Protein Cell Lysis Buffer enables efficient solubilization of the plasma and intracellular membranes, breaks weak intermolecular bonds, and solubilizes most of the commonly studied protein antigens. Total Protein Cell Lysis Buffer reduces protein denaturation, protein complex disruption and loss of enzymatic activity since it contains less detergent than a typical RIPA lysis buffer.

Storage/Stability:

Store product cold, 2-8°C

Application Disclaimer

For Research Use Only.
Not for Therapeutic or Diagnostic Use.

Procedure:

Materials not supplied:

Protease Inhibitors

Phosphate Buffered Saline (PBS) (ice cold)

Note: All procedures should be performed on ice in a cold room with ice cold reagents to reduce proteolysis, dephosphorylation and denaturation.

Add the protease inhibitor cocktail to AMRESCO's Total Protein Cell Lysis Buffer so that the final concentration of protease inhibitors is 1X.

Protocol below is designed for approximately $1x10^6$ cells

Cell Collection:

1. Transfer cells from tissue culture flask to an appropriately sized centrifuge tube.
2. Centrifuge at 2,000rpm for 5 minutes at 4˚C.
3. Remove the media, and resuspend the cell pellet in 10ml cold 1X PBS.
4. Centrifuge at 2,000rpm for 5 minutes at 4˚C.
5. Remove the supernatant and resuspend the pellet in 1ml cold 1X PBS. Transfer this resuspended pellet in PBS to a 1.5ml microcentrifuge tube.
6. Spin 1 minute at 2,000 rpm at 4˚C.

Cell Lysis:

1. Discard the supernatant, and resuspend the pellet in 20X the packed cell volume with Total Protein Cell Lysis Buffer containing protease inhibitors. Vortex vigorously for 10 seconds.
2. Incubate the cell suspension on ice with shaking for 30 minutes with periodic vortexing.
3. Vortex vigorously for 30 seconds.
4. Centrifuge at 14,000xg for 20 minutes at 4˚C.
5. Transfer the supernatant containing total cell protein into a separate microcentrifuge tube.
6. The total cell protein should be stored frozen at 20˚C until needed.

Related Products

Code	Product
M221-1ml	Protease Inhibitor Cocktail 100X, General Use
M222-1ml	Protease Inhibitor Cocktail 100X, General Use with EDTA
M250-1ml	Protease Inhibitor Cocktail Mammalian
E504-100ml E504-500ml	Phosphate Buffered Saline (PBS), 1X Sterile Solution
N655-50ml N655-6x5ml	SeraFree™ Cryopreservation Media (RPMI)
N673-50ml N673-6x5ml	SeraFree™ DMEM Cryopreservation Media
0260-25g 0260-50g	Trypsin 1:300
N182-5x10ml	DMSO, Ultra Pure Grade
K952-100ml	Penicillin/Streptomycin, 100X *Tissue Culture Grade*

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