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1X Protein Lysis Buffer

Store at -20°C

Cat. No.	Description	Quantity
G032	1X Protein Lysis Buffer	5.0 mL

Description
abm's 1X Protein Lysis Buffer is optimized to release proteins of interest from cells and tissues for use in Western Blot experiments. The buffer includes anionic denaturing detergent sodium dodecyl sulfate (SDS) and bromophenol blue, which allows quick preparation of denatured and reduced protein samples from cells. A brief sonication and heating at 100°C for 5 minute will allow the samples from direct loading onto SDS-PAGE gell. **abm**'s 1X Protein Lysis Buffer provides an easy and efficient way for SDS-PAGE and Western Blot sample preparation.

Composition
SDS, Tris-HCl, glycerol, bromophenol blue, DTT.

Storage Condition
Store at -20°C upon receiving.

Protocol

Pre-thaw and thoroughly mix the 1X Protein Lysis Buffer before use.

For Adherent Cells

1. Aspirate media from cells that are 80% to 100% confluent at the time of lysis.
2. Wash the cells with ice-cold 1X PBS briefly and aspirate PBS.
3. Add the 1X Protein Lysis Buffer to the cells (Refer to **Table 1** for volume of G032 needed for different culture vessels). Swirl the container gently to distribute the buffer to all corners.
4. Pipet the cells in buffer up and down a few times. Scrape adherent cells off the vessel surface using a cell scraper if necessary. The cell lysate will be viscous if lysis is complete.
5. Transfer the lysate into a suitable clean tube based on the volume.
6. Sonication: Based on the model of the sonicator and volume of the cell lysate, use a low amplitude setting, and sonicate the cell lysates 1-2 times with one-minute rest on ice between each sonication pulse. Keep sonicating until the lysate is not viscous.
7. Heat the lysate at 100°C for 5 mins.

The lysate is now ready to load directly onto SDS-PAGE gel for protein analysis.

For Suspension Cells

1. Transfer the cells into a suitable conical tube based on the volume. Centrifuge the suspension cells at 1500 rpm for 3 mins to pellet the cells.
2. Aspirate the media supernatant from the cell pellet.
3. Add the 1X Protein Lysis Buffer to the tube (Refer to **Table 1** for volume of G032 needed for different cell numbers).
4. Resuspend thoroughly by pipetting up and down a few times. The cell lysate will be viscous if lysis is complete.
5. Sonication: Based on the model of the sonicator and volume of cell lysate, use a low amplitude setting, and sonicate the cell lysates 1-2 times with one-minute rest on ice between each sonication pulse. Keep sonicating until the lysate is not viscous.
6. Heat the lysate at 100°C for 5 mins.

The lysate is now ready to load directly onto SDS-PAGE gel for protein analysis.

Table 1: Buffer Volume for Different Culture Vessels and Cell Numbers

Culture Vessel	Surface Area (cm^2)	Cell Number at Confluency*	Buffer volume
6-well Culture Plate	9	1.2×10^6	200 µL
35 mm Dish	9	1.2×10^6	200 µL
60 mm Dish	21	3.2×10^6	400 µL
100 mm Dish	55	8.8×10^6	1 mL
150 mm Dish	152	20.0×10^6	3 mL
T-25 Flask	25	2.8×10^6	400 - 500 µL
T-75 Flask	75	8.4×10^6	1-2 mL

* This cell number at confluency in Table 1 was estimated using HeLa cells and will vary with cell type.