RIPA Buffer

Descriptions:
RIPA Buffer enables efficient cell lysis and protein solubilization while avoiding protein degradation and interference with the proteins' immunoreactivity and biological activity. RIPA Buffer also results in low background in immunoprecipitation and molecular pull-down assays.

Compositions:
Ready-to-use solution containing 150 mM NaCl, 1.0% NP-40, 0.5% sodium deoxycholate, 0.1% SDS, 50 mM Tris, pH 7.5, 1mM PMSF, 10 g/ml Leupeptin.

Usage:
Use 1 ml of cold RIPA Buffer for every 5×10^6 of cells (~20 l of packed cells, which is equivalent to ~40 mg of cells).

Procedure for Lysis of Monolayer-cultured Mammalian Cells

1. Carefully remove (decant) culture medium from adherent cells.
2. Wash cells twice with cold PBS.
3. Add cold RIPA Buffer to the cells. Use 1 ml of buffer per 75 cm^2 flask containing 5×10^6 cells. Keep on ice for 5 minutes, swirling the plate occasionally for uniform spreading.
4. Gather the lysate to one side using a cell scraper, collect the lysate and transfer to a microcentrifuge tube. Centrifuge samples at ~14,000 × *g* for 15 minutes to pellet the cell debris.
5. Transfer supernatant to a new tube for further analysis.

Procedure for Lysis of Suspension-cultured Mammalian Cells

1. Pellet the cells by centrifugation at 2,500 × *g* for 5 minutes. Discard the supernatant.
2. Wash cells twice in cold PBS. Pellet cells by centrifugation at 2,500 × *g* for 5 minutes.
3. Add RIPA Buffer to the cell pellet. Use 1 ml of RIPA buffer for 40 mg (~5×10^6 of HeLa cells) of wet cell pellet. Pipette the mixture up and down to suspend the pellet.
4. Shake mixture gently for 15 minutes on ice. Centrifuge mixture at ~14,000 × *g* for 15 minutes to pellet the cell debris.
5. Transfer supernatant to a new tube for further analysis.

Storage: 2-8°C

BIONOVAS Biotechnology Co., Ltd.
647-808-8236
No. 194 kingsdale Ave, Toronto. Ontario. M2N3W9