

RIPA Lysis Buffer (10X)

Catalog No. BSRIPA-030

Size: 30mL

Storage: If buffer will be continually used, it is recommended that the 10X buffer be kept at 4°C for 1-2 weeks. For longer periods of time, buffer should be stored at -20°C.

Product Description

RIPA Lysis Buffer is a complete cell lysis solution reagent used for rapid and efficient total cell lysis and solubilization of proteins from both adherent and suspension cultured mammalian cells, effectively extracting cytoplasmic, nuclear and membrane proteins.

Protocol

A. Cell culture:

It is recommended that cells are cultured to 80-90% confluency prior to performing cell lysis. Cells should be washed by PBS and remove PBS prior to addition of RIPA buffer.

B. Cell Lysis:

1. Dilute 10X RIPA buffer to a 1X Solution using ddH₂O.
2. Chill 1X buffer on ice and add protease inhibitors and phosphatase inhibitors (if required) immediately prior to cell lysis.
3. Apply the ice cold RIPA buffer solution to cell pellets. Vortex briefly and incubate on ice for 20 minutes: Use 300µl of RIPA buffer for one to three 10 cm plates of cells. Scale accordingly for other numbers or sizes of plates based on the surface area of the cell culture dish.
4. Centrifuge the cell lysate in a cooled microcentrifuge at full speed for 15 min to partition supernatant and pellet. Collect the supernatant fraction, which contains extracted membrane and cytosolic proteins to new tube for further analysis.

Notes

1. Some protein kinase and other enzymes may be sensitive to the components of the RIPA Lysis Buffer, resulting in their decreased activity. In such cases, prepare a RIPA Lysis Buffer that does not contain sodium deoxycholate and SDS.

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