

RIPA Lysis Buffer

CAT No. APL-1052

DESCRIPTION

RIPA Lysis Buffer is used for total cell lysis of adherent or non-adherent cells in culture. RIPA Lysis Buffer enables the extraction of cytoplasmic proteins, membrane proteins and nuclear proteins. It is also compatible with many applications, including reporter assays, protein assays, immunoassays and protein purifications. RIPA Lysis Buffer is a more denaturing lysis buffer than NP-40. RIPA Lysis Buffer contains the nonionic detergent NP-40, plus two ionic detergents (Sodium Deoxycholate and SDS). 1 mL of the RIPA Lysis Buffer is sufficient to lyse cells from one 100 mm culture dish or up to 1×10^7 cells of most adherent mammalian cell lines.

CONTENT

	APL-1052
RIPA Lysis Buffer	100 mL

COMPOSITION

	Concentration
Tris	50 mM
NaCl	0.15 M
NP-40	1%
Sodium Deoxycholate	0.5%
SDS	0.1%

STORAGE

This product should be stored at 4°C.

PROTOCOL

Preparation of lysate from cell culture

1. Place the cell culture dish on ice and wash the cells with ice-cold PBS.
2. Aspirate the PBS, then add ice-cold RIPA Lysis Buffer (1 mL per 1×10^7 cells/100 mm dish/150 cm² flask; 0.5 mL per 5×10^6 cells/60 mm dish/75 cm² flask).

NOTES: *If necessary, adding protease and phosphatase inhibitors to RIPA Lysis Buffer and maintain ice-cold status before applying to cells.*

3. Scrape adherent cells off the dish using a cold plastic cell scraper, then gently transfer the cell suspension into a pre-cooled microcentrifuge tube. Alternatively, cells can be trypsinized and washed with PBS prior to

resuspension in RIPA Lysis Buffer in a microcentrifuge tube.

4. Maintain constant agitation for 30 min at 4°C.
5. Centrifuge in a microcentrifuge at 4°C. You may have to vary the centrifugation force and time depending on the cell type; a guideline is 20 min at 12,000 rpm but this must be determined for your experiment (e.g. leukocytes need a very light centrifugation).
6. Gently remove the tubes from the centrifuge and place on ice, aspirate the supernatant and place in a fresh tube kept on ice, and discard the pellets.

PRODUCT USE LIMITATION

Research use only.