

RIPA Cell Lysis Buffer (5X)

RP05-10/RP05-100

V2.0

Store at 2-8 °C For Research Use Only

Introduction

RIPA Cell Lysis Buffer (5X) is used to lyse mammalian cells from adherent or suspension cells in culture. It enables the extraction of cytoplasmic, membrane and nuclear proteins and is compatible with many applications, including reporter assays, protein assays, immunoassays and protein purification. RIPA Cell Lysis Buffer (5X) supplied as a ready to use solution and could minimize non-specific protein-binding interactions to keep background low, while allowing most specific interactions to occur, enabling studies of relevant protein-protein interactions. When diluted to a 1X working solution, it contains 25 mM Tris•HCl pH 7.6, 150 mM NaCl, 1% NP-40, 1% sodium deoxycholate, 0.1% SDS.

Product Components

RIPA Cell Lysis Buffer (5X) (RP05-10)

RIPA Cell Lysis Buffer (5X) 10 mL 1 bottle

User's manual

RIPA Cell Lysis Buffer (5X) (RP05-100)

RIPA Cell Lysis Buffer (5X) 100 mL 1 bottle

User's manual

Safety Information

Please wear gloves, lab coat and goggles while operating. Prevent contact product directly. In case of contacting, wash with large amount of water.

Storage

RIPA Cell Lysis Buffer (5X) should be shipped at room temperature and be stored at 2-8 °C. Expiration date is labeled on the bottle or box.



Materials needed but not provided

- 1. Centrifuge
- 2. Microcentrifuge tubes
- 3. PBS wash buffer: 10 mM KH₂PO₄, 150 mM NaCl, pH 7.4
- 4. Protease Inhibitor Cocktails (if desired)
- 5. Phosphatase Inhibitor Cocktails (if desired)

Instruction

NOTE: Before use, dilute **RIPA Cell Lysis Buffer (5X)** to 1X with ddH₂O. **RIPA Cell Lysis Buffer (5X)** does not contain any protease or phosphatase inhibitors. If desired, add cocktails to the 1X buffer immediately before applying to cells.

A. Sample preparation

- Adherent Cells
- 1. Remove growth medium from the cells by decantation or aspiration.
- Carefully wash cells twice with a volume of ice cold PBS equal to that of the culture media removed.
- Suspension Cells
- 1. Pellet the cells by centrifugation at 2,500 x g for 5 minutes. Discard the supernatant.
- Carefully wash cells twice with a volume of ice cold PBS equal to that of the culture media removed.

B. Sample lysis

- 1. After removal of the final wash solution from the cells, add an appropriate volume of RIPA Buffer (1 mL for 0.5 to 5 x 10⁷ cells). Incubate on ice or in a refrigerator (2-8 °C) for 5-15 minutes.
- 2. Use cell scraper to scrape off cells. Pass the cell lysate through pipette several times to form a homogeneous lysate and transfer lysate to a 1.5 mL microcentrifuge tube on ice.
- 3. Centrifuge the lysate at 14,000 x g for 15 minutes at 4 °C to pellet the cell debris.

NOTE: If a mucoid aggregate of denatured nucleic acids is present, carefully remove it with a micropipette before centrifugation.

- 4. Transfer supernatant to a new tube for further analysis.
- 5. If necessary, aliquot the protein samples for long-term storage at -20 °C. Repeated freeze and thaw cycles cause protein degradation and should be avoided.

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