

RIPA Lysis Buffer (Strong, with Inhibitors)

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1. Product Description

1.1 Product Introduction

RIPA Lysis Buffer (Strong) exhibits a potent lytic effect on animal cell membranes, cytoplasmic, and nuclear components. It is a classic, rapid lysis buffer for cells and tissues, enabling efficient lysis and protein extraction from cultured mammalian cells (including adherent and suspension cells). The extracted proteins are suitable for routine applications such as PAGE, Western blot, and IP.

1.2 Product Advantages

- 1) Convenience: Ready-to-use solution, no need for self-preparation.
- 2) Flexibility: Suitable for routine PAGE, Western blot, IP, Western blot for phosphorylated proteins, etc.
- 3) Versatility: Exhibits strong lysis effects on animal cell membrane, cytoplasmic, and nuclear components.

1.3 Reagent Components

Reagent Name	Size
RIPA Lysis Buffer (Strong)	100ml
100× PMSF	1ml

2. Instructions for Use

2.1 Adherent Cells:

- 1) Aspirate and discard the culture medium. Wash the cells once with PBS, physiological saline, or serum-free culture medium (this step can be omitted if serum proteins cause no interference).
- 2) Add RIPA Lysis Buffer (Strong) at a ratio of 150-250 μ L per well of a 6-well plate, along with 1.5-2.5 μ L of 100× PMSF. Pipette several times to ensure the lysis buffer fully contacts the cells. Typically, animal cells are lysed within 1-2 seconds after contacting the RIPA Lysis Buffer (Strong).

2.2 Suspension Cells:

- 1) Collect cultured cells by centrifugation at $800 \times g$ for 5 min at 4°C. Estimate the pellet volume (e.g., 10^6 cells \approx 20 μ L, 10^7 cells \approx 100 μ L).
- 2) Add 250-500 μ L of RIPA Lysis Buffer (Strong) and 1.5-2.5 μ L of 100× PMSF per 50-100 μ L of cell pellet. Incubate on ice for 10 min, vortexing for 30 sec every 5 min.
- 3) Note: If the total protein lysate is too viscous, it can be heated at 95°C for 5 min, rapidly cooled on ice for 5 min, and then proceed to the next step.
- 4) Centrifuge at $12,000 \times g$ for 10 min at 4°C. Transfer the supernatant to a new tube to obtain the total cellular protein lysate.

2.3 Tissue Samples:

- 1) Mince the tissue into fine pieces. Wash twice with pre-chilled PBS, centrifuging to discard the PBS after each wash.
- 2) Add RIPA Lysis Buffer (Strong) at a ratio of 150-250 μ L per 20 mg of tissue, along with 1.5-2.5 μ L of 100× PMSF. If lysis is insufficient, increase the volume of RIPA Lysis Buffer (Strong). For higher protein concentration, use a smaller volume of lysis buffer relative to tissue weight.
- 3) Homogenize the tissue thoroughly. Alternatively, flash-freeze the tissue sample in liquid nitrogen, grind it thoroughly, and then add RIPA Lysis Buffer (Strong) for lysis.
- 4) After complete lysis, centrifuge at $12,000 \times g$ for 10 min at 4°C. Collect the supernatant for subsequent operations such as PAGE, Western blot, and IP.

3. Precautions

3.1 All steps for lysing samples should be performed on ice or at 4°C.

3.2 Selecting the appropriate lysis buffer may require optimization through preliminary experiments to determine the best conditions for your specific application.

3.3 This product contains high concentrations of detergents and is not suitable for protein concentration determination using the Bradford method. Please use the BCA method (e.g., BK0001-01, BK0001-02) instead.

4. Related Products

Product	Cat.No.	Size
RIPA Lysis Buffer (Strong, with Inhibitors)	BK0060-01	100ml