

Bioworld Technology CO., Ltd. RIPA Total Protein Extraction Lysis Buffer

Cat No.: BD0031

Introduction

RIPA total protein extraction lysis buffer is a traditional lysate of cell and tissue protein extraction. The extracted protein samples can be used for conventional WB, IP, as well as protein purification. The main ingredients of the buffer are Triton-X100, sodium deoxycholate, and SDS. The lysis buffer can extract total proteins both of cells and tissues effectively.

Note: 1). Please prepare protease or phosphatase inhibitors and inject into lysis buffer before use.

2). BCA Protein Assay method is recommended to quantify the protein concentrations of extracts.

Reagents

RIPA total protein extraction lysis buffer: 100ml

Application

Total protein extraction

Storage & Shelf life

Stored at -20° C. It's recommended to aliquot before stored at -20° C.

Procedure

1 Lysis of cultured Mammalian Cells

1) For adherent mammalian cells(1-10*106 cells): the cells were harvested with trypsin-EDTA, then centrifuged at 1000 rpm for 3min, 4° C; For suspension mammalian cells(1-10*106 cells): the cells were centrifuged at 1000 rpm for 3min.

2) Resuspend and wash the cells with PBS in 1.5 ml tube. Then centrifuge the cells at 1000 rpm for 3min, remove and discard the supernatant as much as possible.

3) Add 800-1000 μ l lysis buffer (for 1-10*106 cells) to the precipitate and resuspend the precipitate, vortex for 15s.

4) Place the tube on ice for 20min. During this period, vortex the tube for 15s each 5min.

5) Centrifuge the tube at 12,500 rpm for 15min, 4°C. Transfer the supernatant (cell lysate) to a new tube. Store at 4°C (for short term) or -80°C (for long term).

2、Lysis of animal tissues

1) Cut the tissues into small fragments, resuspend and wash the cells with PBS. Then centrifuge the cells at 3000 rpm for 3min, remove and discard the supernatant as much as possible.

2) Add 500 μ l lysis buffer (for 100mg tissues) to the precipitate and resuspend. (According to the actual requirements , the quantity of lysis buffer could be added or reduced.)

3) Use the homogenizer to grind the tissue fully on ice. Place the mixture on ice for 30min. During this period, vortex the tube for 15s to resuspend the precipitate each 5min.

4) Centrifuge the tube at 12,500 rpm for 15min, 4°C. Transfer the supernatant (cell lysate) to a new tube. Store at 4°C (for short term) or -80°C (for long term).

NOTE

1) This product is only used for mammalian cells and animal tissues.

2) Avoid freeze-thaw cycles. Recommend to aliquot before stored at -20°C or -80°C. Period of validity is 12 months (-20°C).

3) To increase yields, sonicated the pellet with 50% pulse for 30 seconds.

4) All steps on ice.

Research Use

For research use only, not for use in diagnostic procedures.

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