RIPA Lysis Buffer (Strong)

Cat. No: E-BC-R327 **Size: 20 mL/ 50 mL/ 100 mL**

Product Content

Cat	Products	20 mL	50 mL	100 mL	Storage
E-BC-R327	RIPA Lysis Buffer (Strong)	20 mL	50 mL	100 mL	-20 ℃
E-BC-R287	100 mM PMSF	200 μL	500 μL	1 mL	-20 ℃
E-BC-R250	100 mM Na_3VO_4	200 μL	500 μL	1 mL	-20 ℃
Manual			**1 copy**		

Introduction

RIPA Lysis Buffer is a traditional rapid cell tissue lysate used as the preferred lysate for protein extraction from tissues or cells in the Western Blot assay.

Instructions

1. For Tissue Samples

a. Take the samples, wash the tissue thoroughly with pre-cooled PBS (0.01M, pH7.4) to remove the surface blood and internal debris.

b. Weigh and smash the tissue, add an appropriate ratio of RIPA Lysis Buffer (add 10 μL PMSF and 10 μL Na_3VO_4 to 1 mL RIPA Lysis) and homogenizely lyse the tissue.
It is recommended to homogenize according to the ratio of tissue weight: RIPA volume = 3:10. For example, add 1 mL RIPA Lysis Buffer to 0.3 g tissue sample, the specific volume can be adjusted according to experimental requirements.

c. Shake and lyse on the ice for 30 min after homogenization. Blow the sample repeatedly with a Pipette gun for about 50 times (under ice water bath conditions) to Make sure the DNA strand is broken and reduce the viscosity of sample.

d. Centrifuge at 12,000 rpm for 10 min at 4 ℃.

e. Take the supernatant and measure the protein concentration.

2. For Cell Sample

a. Collect the cells, wash them thoroughly with pre-cooled PBS (0.01 M, pH7.4) to remove the medium off (it is generally recommended to wash 3 times).

b. Add an appropriate ratio of RIPA Lysis Buffer (10 μL PMSF and 10 μL Na_3VO_4 in 1 mL RIPA Lysis) and lyse on the ice for 30 min.
It is recommended to add 0.1 mL of RIPA Lysis Buffer to each well of a 6-well plates (the protein content in different cells may vary, and the volume of the lysate added can be appropriately adjusted).

c. Blow the sample repeatedly with a Pipette gun for about 50 times (under ice water bath conditions) to Make sure the DNA strand is broken and reduce the viscosity of sample.

d. Centrifuge at 12,000 rpm for 10 min at 4 ℃.

e. Take the supernatant and measure the protein concentration.

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RIPA Lysis Buffer components

50 mM Tris (pH7.4), 150 mM NaCl, 1% Triton X-100, 1% $C_{24}H_{39}O_4Na$, 1 mM EDTA, 0.1% SDS, 10 mM NaF, 1 mM Na_3VO_4, 1 mM PMSF.

Storage

Store at -20 ℃ for 12 months.

Cautions

1. All steps to lyse samples should be performed on ice or at 4 ℃.
2. It is recommended to add 10 μL PMSF and 10 μL Na_3VO_4 to 1 mL RAPI Lysis before use. If RIPA Lysis is precipitated, please dissolve at room temperature or in a warm water bath.
3. It is normal for a clear gelatinous substance to appear in the lysate of RIPA lysate. This transparent gel is a complex containing genomic DNA. Repeatedly blowing the sample with a pipette about 50 times can effectively reduce the viscosity of the sample. After centrifugation, the supernatant can be taken for subsequent experiments.
4. The tissue or cell sample lysed by RIPA Lysis Buffer cannot be measured by the Bradford method because of the high concentration of detergent in the lysis. It is recommended to measure the protein concentration by the BCA method.
5. For your safety and health, please wear the lab coat and disposable gloves before the experiments.