WB (IP) Lysis Buffer

Cat #: BMP2020 Size: 100 mL

	WB (IP) Lysis Buffer		
REF	**Cat #:** BMP2020	LOT	**Lot #:** Refer to product label
	Applicable samples: Animal Tissues, Cells		
	Application: SDS-PAGE, WB, IP and Co-IP		
	Storage: Store according to the recommended storage conditions of each component, stable for 12 months		

Assay Principle

WB (IP) Lysis Buffer is a lysate under non-denaturing conditions. It is mainly used to extract soluble proteins from animal cells and tissues. It is mild and can maintain the original protein-protein interaction. The protein sample lysed can be used for conventional SDS-PAGE, WB and other experiments. Stored at 4℃ without thawing. It is a fast and convenient cell tissue lysis solution.

Materials Supplied and Storage Conditions

Kit components	Size	Storage conditions
WB (IP) Lysis Buffer	50 mL×2	4℃
PMSF (100 mM)	1.5 mL	-20℃, protected from light

Materials Required but Not Supplied

- Ice Maker, Refrigerated Centrifuge, Vortex
- Protease Inhibitor Cocktail (100×)
- Phosphatase Inhibitor Cocktail
- Precision Pipettes, Disposable Pipette Tips
- Dounce homogenizer (for Tissue Samples)

Reagent Preparation

WB (IP) Lysis Buffer: Ready to use as supplied. Keep on ice before use. Store at 4℃.

PMSF (100 mM): Ready to use as supplied. Keep on ice before use. Store at -20℃, protected from light.

Working WB (IP) Lysis Buffer: Before use, take an appropriate amount of WB (IP) Lysis Buffer (approximately 50-100 μL per 1×10^6 cells or 100-200 μL per 20 mg tissue sample) and add PMSF (100 mM) at 100:1 (V/V). Add Protease Inhibitor Cocktail (100 ×) and Phosphatase Inhibitor Cocktail as needed.

Assay Procedure

A. For cells

1. Discard the culture medium and wash once with PBS, physiological saline or serum-free culture medium (if the protein in the serum does not interfere, it is not necessary to wash), and centrifuge again to collect the cells.

2. Add Working WB (IP) Lysis Buffer at a ratio of about 50-100 μL per 1×10^6 cells. For example, about 100-200 μL of Working WB (IP) Lysis Buffer should be added to each well of a 6-well plate. Use a pipette several times to make sufficient contact between Working WB (IP) Lysis Buffer and the cells, and lyse on ice for 3-5 min.

3. After fully lysed, centrifuge at 10,000-14,000 g for 3-5 min, and take the supernatant for subsequent SDS-PAGE, WB, IP and Co-IP operations.

B. For tissue samples

1. Cut the tissue into fine pieces.

2. Add Working WB (IP) Lysis Buffer at a ratio of 100-200 μL per 20 mg tissue sample.

3. Homogenize with a homogenizer and lyse on ice for 3-5 min.

Note: 1. If the sample is not sufficiently lysed, the amount of Working WB (IP) Lysis Buffer can be appropriately increased; if a high concentration of protein samples is required, the amount of Working WB (IP) Lysis Buffer can also be appropriately decreased. 2. If the tissue sample is very small, it can be cut properly and directly added to Working WB (IP) Lysis Buffer, and fully lysed by vigorous vortexing.

4. After fully lysed, centrifuge at 10,000-14,000 g for 3-5 min, and take the supernatant for subsequent SDS-PAGE, WB, IP and Co-IP operations.

Precautions

1. All steps of lysing samples should be performed on ice or at 4℃. The lysed protein samples can be aliquoted and stored at -80℃ for a long time.

2. The protein concentration of the cleaved protein sample can be determined with Protein Quantification Kit (BCA Assay) . Due to the high concentration of interfering substances such as Triton X-100, the protein concentration of the sample cannot be determined by the Bradford method.

3. After lysing cells or tissues, there is no very viscous transparent DNA clumps, which can be used for subsequent operations without ultrasonic treatment, and is also suitable for WB detection of phosphorylated proteins. It is recommended to use ExKine™ Pro Total Protein Extraction Kit for Animal Cultured Cells/Tissues to obtain a clean supernatant. If it is found that the target protein cannot be IP down, it means that the strength of WB (IP) Lysis Buffer is too strong, and a milder lysate such as NP-40 lysate can be used.

Recommended Products

Catalog No.	Product Name
KTP3007	ExKine™ Pro Total Protein Extraction Kit for Animal Cultured Cells/Tissues
KTP3008	ExKine™ Pro Total Protein Extraction Kit for Plant Tissues
KTD3001	Protein Quantification Kit (BCA Assay)
BMP1001	Protease Inhibitor Cocktail (100×)
BMU103-CN	SuperKine™ Enhanced Antibody Dilution Buffer
BMU102-CN	SuperKine™ West Femto Maximum Sensitivity Substrate
BMU101-CN	SuperKine™ West Pico PLUS Chemiluminescent Substrate
KTD3003	SDS-PAGE Protein Sample Loading Buffer (5×)

Disclaimer

The reagent is only used in the field of scientific research, not suitable for clinical diagnosis or other purposes.