ExKine™ Total Protein Extraction Kit

Cat #: KTP3006 Size: 50 T/200 T

	Total Protein Extraction Kit		
REF	**Cat #:** KTP3006	LOT	**Lot #:** Refer to product label
	Applicable samples: Tissues, Cells		
	Storage: Stored at -20℃ for 12 months		

Assay Principle

Extraction of cellular proteins requires efficient cell lysis and protein solubilization, while avoiding protein degradation and/or interference with protein immunoreactivity and biological activity. ExKine™ Total Protein Extraction Kit is composed of rapid and efficient solutions for lysis of mammalian adherent cells, nonadherent cells, and tissues, which is compatible with many different applications, such as reporter assays, protein purification, Western blot, immunoprecipitation.

Materials Supplied and Storage Conditions

Kit components	Size		Storage conditions
	50 T	200 T	
Protein Extraction Reagent (2×)	25 mL	100 mL	-20℃
Protease Inhibitor (100×)	0.5 mL	2 mL	-20℃

Materials Required but Not Supplied

- Vortexer, centrifuge tube
- Cell scraper
- Precision Pipettes, Disposable Pipette Tips
- Phosphate buffered saline (PBS) , Deionized Water
- Dounce homogenizer(for Tissue Samples)

Reagent Preparation

Working Protein Extraction Reagent (1×): Before use, dilute with deionized water to 1×Protein Extraction Reagent, immediately add 10 μL protease inhibitors to 1 mL 1×Protein Extraction Reagent, store on ice; store at -20℃.

Protease Inhibitor (100×): Ready to use as supplied. Place on ice before use; store at -20℃. The remaining working solution can be stored at -20℃ after aliquoting to avoid repeated freezing and thawing.

Note: 1. If you continue to use the Protein Extraction Reagent (2×), it is recommended to keep the Protein Extraction

Reagent (2×) at 4℃ for 1-2 weeks. For long-term storage, it is recommended to aliquot and store at -20℃; 2. If necessary, add the phosphatase inhibitor to the buffer immediately before use.

Assay Procedure

Note: Perform all steps at 2-8℃. Use precooled buffers and equipment. Ensure all the solutions are defrosted and homogeneous.

I For adherent cells

1.Remove the growth media from the cells and wash the cells twice with PBS.

2.Add 1 mL 1×Working Protein Extraction Reagent to 10 cm dish.

Note: Cells grown in 10 cm plates typically contain 10^7 cells (50 mg).

3.Incubate the plate on ice for 5 min, swirling occasionally for uniform spreading.

4.Scrape and collect the lysate to a centrifuge tube.

Note: To increase the protein yields, sonicate the lysate briefly.

5.Centrifuge samples at 14,000 g for 15 min to pellet the cell debris and transfer the supernatant to a new tube for further analysis.

II For non-adherent cells

1.Pellet the cells by centrifugation at 700 g for 10 min.

2.Discard the supernatant and wash with cold PBS twice.

3.Add 1 mL 1×Working Protein Extraction Reagent to 0.5-1×10^7 cells. Pipette the mixture up and down to suspend the pellet.

4.Shake mixture gently for 15 min on ice. To increase the protein yields, sonicate the lysate briefly.

5.Centrifuge samples at 14,000 g for 15 min to pellet the cell debris and transfer the supernatant to a new tube for further analysis.

III For Tissue Preparation

1.Cut 5-20 mg of tissue into small pieces and place in a centrifuge tube.

2.Wash tissue with PBS. Centrifuge tissue at 500 g for 5 min and discard the PBS.

3.Resuspend the tissue gently in 1mL cold 1×Working Protein Extraction Reagent.

4.Homogenize tissue using a homogenizer or a tissue grinder until the cells are completely lysed.

5.Centrifuge samples at 14,000 g for 15 min to pellet the cell debris and transfer the supernatant to a new tube for further analysis.

Recommended Products

Catalog No.	Product Name
KTP3001	ExKine™ Nuclear and Cytoplasmic Protein Extraction Kit
KTP3002	ExKine™ Nuclear Protein Extraction Kit
KTP3003	ExKine™ Cytoplasmic Protein Extraction Kit
KTP3004	ExKine™ Total Membrane Protein Extraction Kit
KTP3005	ExKine™ Membrane and Cytoplasmic Protein Extraction kit

Disclaimer

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