Total Protein Extraction Kit

Cat. No. KITG-074 **Lot. No.** (See product label)

SPECIFICATION

Product Overview	This kit is intended for the extraction of total protein from mammalian tissues and cells. The Lysis Buffer contains Protease Inhibitors and Phosphatase Inhibitors, enabling rapid recovery of total protein. The extracted protein is suitable for Western blotting and other basic research experiments.
Species	Mammalian
Description	Pack Size: 50 tests or 100 tests. For Research Use Only. Not for Use in Diagnostic Procedures.
Form	Kit
Applications	Extraction of total protein from mammalian tissues and cells. The extracted protein is suitable for Western blotting and other basic research experiments.
Notes	Instructions for Use 1. Extraction of Total Protein from Tissue 1) Preparation of Extraction Solution: Pipette 1 mL Lysis Buffer into a centrifuge tube. Add 10 µL Phosphatase Inhibitors, 10 µL Protease Inhibitors, and 10 µL PMSF. Mix thoroughly and place on ice. 2) Weigh 0.1 g fresh tissue and place it at the inlet buffer area of a glass homogenizer. Use ophthalmic scissors to cut the tissue pieces as finely as possible. Add 0.5-1 mL Extraction Solution and grind until no visible tissue pieces remain. Perform the operation on ice. The resulting material is the tissue homogenate. 3) Transfer the tissue homogenate to a 1.5 mL centrifuge tube. Centrifuge at 12,000 × g for 30 min at 4°C. Collect the supernatant as the protein

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	extract. It may be used for subsequent protein quantification or denatured for subsequent protein experiments. 2. Extraction of Total Protein from Cells 1) Adherent Cells: Discard the culture medium. Wash twice with pre-cooled PBS and discard the PBS. Add 1 mL Lysis Buffer per 10^7 cells. Scrape the cells off on ice using a cell scraper. Transfer the detached cell lysate to a centrifuge tube and lyse by inversion for 20-30 min. Centrifuge at 12,000 × g for 30 min at 4°C. Collect the supernatant as the protein extract. It may be used for subsequent protein quantification or denatured for subsequent protein experiments. 2) Suspension Cells: Collect the cells by centrifugation at 400 × g at 4°C. Wash twice with pre-cooled PBS. Add 1 mL Lysis Buffer per 10^7 cells. Vortex for 10 s, lyse on ice for 10 min, and repeat 3-4 times. Centrifuge at 12,000 × g for 30 min at 4°C. Collect the supernatant as the protein extract. It may be used for subsequent protein quantification or denatured for subsequent protein experiments. Note: Store the extracted protein at -80°C. Take it only when needed, and avoid repeated freeze-thaw cycles and prolonged storage. Precautions 1) During the experiment, all reagents should be pre-cooled or freshly thawed to maintain a low-temperature environment throughout the procedure. 2) PMSF is toxic and degrades rapidly in aqueous solution. Add it immediately before use and take appropriate protective measures. 3) The Lysis Buffer contains Protease Inhibitors. After extraction, the protein cannot be used for protein kinase or phosphokinase studies. 4) To ensure accurate results and avoid reagent loss, carefully read the instructions before the assay, confirm that reagent storage and preparation are adequate, and ensure that the operating steps are clear. Use 2-3 samples expected to show substantial differences for a preliminary test. If any problem occurs during the procedure, contact the relevant technical personnel promptly.
Storage	Kit Components and Storage: Lysis Buffer: 50 mL for 50 tests or 100 mL for 100 tests; store at 4°C. Protease Inhibitors: 0.5 mL for 50 tests or 1 mL for 100 tests; store at -20°C. Phosphatase Inhibitors: 0.5 mL for 50 tests or 1 mL for 100 tests; store at -

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	20°C. PMSF: 0.5 mL for 50 tests or 1 mL for 100 tests; store at -20°C. Store extracted protein at -80°C. Use as needed and avoid repeated freeze-thaw cycles and prolonged storage.
Reconstitution	Preparation of Extraction Solution: Pipette 1 mL Lysis Buffer into a centrifuge tube. Add 10 µL Phosphatase Inhibitors, 10 µL Protease Inhibitors, and 10 µL PMSF. Mix thoroughly and place on ice.
Usage	For extraction of total protein from mammalian tissues and cells.

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