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Produktinformation

- Forschungsprodukte & Biochemikalien
- Zellkultur & Verbrauchsmaterial
- Diagnostik & molekulare Diagnostik
- Laborgeräte & Service

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See the following pages for more information!

Lieferung & Zahlungsart

siehe unsere Liefer- und Versandbedingungen

Zuschläge

- Mindermengenzuschlag
- Trockeneiszuschlag
- Gefahrgutzuschlag
- Expressversand

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（FOR RESEARCH USE ONLY. DO NOT USE IT IN CLINICAL DIAGNOSIS !）

Catalog No：E-BC-E002

Specification：50 Assays / 100 Assays

Elabscience®Total Protein Extraction Kit

This manual must be read attentively and completely before using this product.
If you have any problem, please contact our Technical Service Center for help：

Toll-free: 1-888-852-8623
Tel: 1-832-243-6086
Fax: 1-832-243-6017
Email: techsupport@elabscience.com
Website: www.elabscience.com

Please kindly provide us the lot number (on the outside of the box) of the kit for more efficient service.

Intended use

This kit can be used to extract total protein from animal tissues and cells, and the obtained protein can be used for subsequent studies such as Western Blot and co-immunoprecipitation.

Detection principle

Cell and tissue samples are treated with lysates containing protease inhibitors and phosphatase inhibitors to prevent the enzymes in the sample from being released to hydrolyze or dephpsphprylate protein due to the disruption of the membrane system.

Kit components & storage

Item	Component	Size 1 (50 assays)	Size 2 (100 assays)	Storage
Reagent 1	Lysis Buffer	60 mL ×1 vial	60 mL ×2 vials	2-8℃, 12 months
Reagent 2	Phosphatase Inhibitor	0.6 mL ×1 vial	1.2 mL ×1 vial	-20℃, 12 months, shading light
Reagent 3	Protease Inhibitor	0.6 mL ×1 vial	1.2 mL ×1 vial	-20℃, 12 months, shading light
Reagent 4	Phenylmethylsulfonyl Fluoride	0.6 mL ×1 vial	1.2 mL ×1 vial	-20℃, 12 months, shading light

Note: The reagents must be stored strictly according to the preservation conditions in the above table. The reagents in different kits cannot be mixed with each other. For a small volume of reagents, please centrifuge before use, so as not to obtain sufficient amount of reagents.

Materials prepared by users

Instruments:

High-speed freezing centrifuge, 5mL Glass homogenizer

Reagents:

Double distilled water, PBS(0.01 M，pH=7.4)

Reagent preparation

① Place all reagents in ice water for pre-cooling for at least 15 min until it has returned to the solution state before use.

② The preparation of lysis working solution:

Mix lysis buffer, phosphatase inhibitor and protease inhibitor with a ratio of 1000:10:10 and preserved on ice with shading light. Prepare the fresh needed amount before use and the prepared solution should be used within 20 min..

Operating steps

1. Total Protein Extraction of Tissue

① Take 0.1g of fresh tissue, wash with PBS（0.01 M，pH 7.4） at 2-8℃ to remove blood. Blot the water with absorbent paper.

② Cut the tissue into pieces with scissors and place them into a pre-cooled 5 mL glass homogenizer.

③ Add 1 mL of pre-cooled lysis working solution and 10 μL of precooled phenylmethylsulfonyl fluoride.

④ Grind the tissue up and down in the ice bath for about 30 times.

⑤ Transfere tissue homogenate to a 2 mL pre-cooled EP tube, place in the ice bath for 15 min.

⑥ Centrifuge at 12000 g at 4℃ for 15 min. The supernatant was the total protein extract. Place it on ice for detection.

⑦ The prepared total protein solution should be stored at -70℃ with avoiding of repeated freeze-thaw.

2. Total Protein Extraction of Cell

a、Cell collection

Suspension cell: Transferre cell suspension to pre-cooled EP tubes, centrifuge at 4℃ at 1000 g for 10 min to remove supernatant, wash with PBS（0.01 M，pH 7.4）at 2-8℃ once, centrifuge at 4℃ at 1000 g for 10 min to remove supernatant, leaving precipitation for use.

Adherent cell: Discard the culture solution and wash the cells twice with PBS (0.01 M, pH 7.4) at 2-8℃. Scrape down cells with cell scraping, or treat with EDTA solution, blown the cells off with a pipettor and transferre the cell suspension to a pre-cooled EP tube. Centrifuge at 4℃ at 1000 g for 10 min to remove supernatant, wash with PBS（0.01 M，pH 7.4）at 2-8℃ once, centrifuge at 4℃ at 1000 g for 10 min to remove supernatant, leaving precipitation for use.

b、Cell extraction

① Take 5×10^6 cells and add 0.5 mL of pre-cooled lysis working solution and 10 μL of precooled phenylmethylsulfonyl fluoride.

② Place on ice box for 15 min, vortex and mix every 5 min for 10 s each time.

③ Centrifuge at 12000 g at 4℃ for 15 min. The supernatant was the total protein extract, which was placed on ice for detection.

④ The prepared total protein supernatant should be stored at -70℃ to avoid repeated freeze-thaw.

Statement

1. This assay kit is for Research Use Only. We will not response for any arising problems or legal responsibilities causing by using the kit for clinical diagnosis or other purpose.
2. Please read the instructions carefμLly and adjust the instruments before the experiments. Please follow the instructions strictly during the experiments.
3. Protection methods must be taken by wearing lab coat and latex gloves.
4. If the concentration of substance is not within the detection range exactly, an extra dilution or concentration shoμLd be taken for the sample.
5. It is recommended to take a pre-test if your sample is not listed in the instruction book.
6. The experimental resμLts are closely related to the situation of reagents, operations, environment and so on. Elabscience will guarantee the quality of the kits only, and NOT be responsible for the sample consumption caused by using the assay kits. It is better to calcμLate the possible usage of sample and reserve sufficient samples before use.