Histone Rapid Extraction kit (centrifugal column method)

Item No. : EX2410
Specification: 50T/100T
Validity: 2-8℃ storage, valid for one year.

Product content:

Components	Name	50T	100T	Storage conditions
Component A	Protein extract Liquid A	25mL	50mL	Store at 2-8℃
Component B	Protease inhibitor mixture B	100μL	200μL	Store at -20℃
Component C	Protein stabilizer C	100μL	200μL	Store at 2-8℃
Component D	Egg white centrifugal column	50 sets	100 sets	Store at RT

Note:

1. This kit is suitable for animal tissue.
2. The protease inhibitor can also be stored at 2-8℃ before use with the lid open. Store at -20℃ after opening the lid for use.
3. The protease inhibitor is solid at 2-8℃. Take it out of the refrigerator and return to room temperature or 37℃ water bath for a short time. When it becomes liquid, centrifuge it to the bottom of the tube and then open the lid.
4. Please use the reagent as soon as possible after unpacking.

Product Introduction:

The histone protein extraction kit provides a complete set of reagents suitable for extracting total protein from a variety of animal tissue samples. The extraction process is simple and convenient. The kit is equipped with centrifugal column for effective removal of non-protein impurities, not the use of centrifugal column to adsorb proteins.

This kit contains a unique formulation that effectively dissolves cell membrane components, including the plasma membrane, nuclear membrane, and various organelle membranes. The kit contains a protease inhibitor mixture that prevents the protease from degrading the protein and ensures the extraction of high purity proteins.

The proteins extracted from this kit can be used for downstream protein research experiments such as Western Blotting, protein electrophoresis, immunoprecipitation, ELISA, transcriptional activity analysis, Gel shift gel blocking assay, and enzyme activity determination. The proteins extracted by this kit are active proteins with natural protein conformation.

EDTA is not present in this kit and is compatible with downstream applications such as metal chelation and chromatography.

Bring your own reagents and instruments:

Centrifuge, oscillator, vortex mixer, pipette, refrigerator, ice box, PBS buffer, protein quantification kit, centrifuge tube, suction tip, disposable gloves

Product Features:

1. Easy to use.
2. Containing protein stabilizer, the extracted protein is stable.
3. The background interference is low when the protein concentration is detected by UV.
4. Protease inhibitors inhibited protein degradation, and the formulation of protease inhibitors was optimized. The protease inhibitor mixture consists of 6 separate protease inhibitors; Each

inhibitor specifically inhibits one or several protease activities. The composition of the mixture is optimized so that it can inhibit almost all important protease activities, including serine protease, cysteine protease, aspartate protease, alanyl-aminopeptidase, etc.

How to use:

First, use precautions:

1. The reagent in the rotating cap centrifuge tube should be centrifuged briefly before opening the cap, and the liquid on the inner wall of the cap should be thrown to the bottom of the tube to avoid the liquid spilling when opening the cap.
2. Protease inhibitor at 2-8℃ is a solid state, after taking out from the refrigerator, return to room temperature or 37℃ for a short time water bath, become a liquid state, centrifuge to the bottom of the tube and then open the lid.
3. All reagents used in the experiment must be pre-cooled; All utensils must be pre-cooled in a -20℃ refrigerator. The sample must be kept at a low temperature during the whole process.
4. If the solution of protease inhibitor is precipitated during storage, it will not affect the use, and it should be used normally after dissolution.
5. If the kit cannot be used up in a short time, the protease inhibitor mixture should not be added to the extraction solution all at once.
6. Other protease inhibitor products can be added as needed for your own experiment.

Two, operation steps

1. Extraction solution preparation:

Add 2μL protease inhibitor mixture and 2μL protein stabilizer to every 500μL protein extract, mix well and set aside on ice.

2. Take 50-100mg tissue and wash it off with PBS. Then cut into pieces with surgical scissors, add 500μL of protein extract and homogenize with a tissue homogenizer/homogenizer until no visible solids are visible to the naked eye.
3. The tissue homogenate is sucked into a pre-cooled clean centrifuge tube and oscillated at 4℃ for 10-20 minutes.
4. The liquid collection tube was put on, the extraction solution was sucked into the protein centrifuge tube and centrifuged for 5 minutes at 4℃, 10000g.
5. The total tissue protein was obtained by collecting the liquid in the tube.
6. The protein extract was quantified and divided into -80℃ refrigerator for reserve or directly used in downstream experiment.

Frozen samples:

1. For frozen tissue samples, follow the above procedure for tissue samples.
2. The frozen tissue sample is not washed with PBS, and can be treated by directly adding protein extract.

Analysis of common problems:

1. Slow cell lysis

In order to fully ensure the activity of the extracted protein, the extract adopts a unique formula of protective protein, which has mild cracking ability and wide downstream application. The cracking time can be extended appropriately.

2. Low protein concentration?

Handling part of the cell sample may not fully lyse, resulting in low protein concentrations. Just extend the processing time of reagent A appropriately. It is best to handle under the condition of continuous oscillation, and it can be mixed with a suction head at intervals of several minutes without an oscillator.

3. What is the method of quantifying the protein?

BCA method is recommended. The Bradford method is not suitable because reagent A contains components that interfere with the Bradford method, resulting in inaccurate quantification. If dialysis has been performed or the buffer system has been replaced with a desalting column, the Bradford method can be used for quantification.

4. Is the extracted protein active?

This kit does not contain ionic detergent components, does not destroy the protein structure, does not disrupt the original interaction between the proteins, and the proteins maintain their natural conformation and activity.

What to note:

1. Before the formal experiment, please select several samples for pre-experiment to optimize the experimental conditions and achieve the best experimental results.
2. Centrifuge the reagent in the screw cap microreagent tube briefly before opening the cap, and centrifuge the liquid on the cap and inside wall to the bottom of the tube to avoid reagent loss when opening the cap.
3. Do not mix with other brands of reagents, otherwise it will affect the use effect.
4. Contamination of the sample or reagent with bacteria or fungi or cross-contamination of reagents may result in false results.
5. It is best to use disposable suction heads, tubes, bottles, or glassware, and reusable glassware must be washed and thoroughly removed of residual cleaners before use.
6. All samples and exposed glassware should be disposed of in accordance with the prescribed procedure after the experiment is completed.