Blood Cell Protein Extraction Kit

Cat: EX1190
Size: 50T/100T
Storage: 2-8℃, valid for 1 year.

Kit Components:

Kit Components	50T	100T	Storage
Reagent A: Protein Extract Solution A	50mL	100mL	2-8℃
Reagent B: Protease Inhibitor Mixture	100μL	200μL	-20℃

Note:

1. Protease inhibitors can also be stored at 2-8°C before use without open lid. Store at -20°C after opening the lid for use.
2. 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.
3. Please use the reagent as soon as possible after unpacking!

Introduction:

Blood cell protein extraction kit is suitable for extracting total protein from all kinds of animal whole blood cells. The extraction process is simple and convenient, and can be completed within 1h.

This kit contains a unique formula 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.

This kit does not contain EDTA and is compatible with downstream applications such as metal chelation and chromatography.

Self-prepared Reagents and Instruments:

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

Product Features:

1. Easy to use, shorten the time of protein extraction to 30min to 1h.
2. Containing protein stabilizer, the extracted protein is stable.
3. The background interference is low when the protein concentration is detected by UV.

Protocols:

First, use precautions

1. Before the formal experiment, please select several samples to do pre-experiment, in order 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. All reagents in the process of 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.
7. In the downstream experiment, if the enzyme activity of specific protease or phosphatase is detected, the extract can be without protease or phosphatase inhibitors. Pay attention to the low temperature operation during the extraction process to shorten the centrifugation time.
8. Do not mix with other brands of reagents, otherwise it will affect the use effect.
9. Contamination of the sample or reagent with bacteria or fungi or cross-contamination of reagents may result in false results.

Second, blood cell protein extraction

1. Extraction liquid preparation:

Add 1μL protease inhibitor mixture into every 500μL cold blood cell protein extraction solution, mix well and put on ice for use.

2. Protein extraction

(1) Take 500μL fresh anticoagulant blood and centrifuge 3000×g for 10min.
(2) Remove the upper plasma and collect the lower blood cell precipitate.
(3) Wash the blood cells with PBS and precipitate them twice.
(4) Add 500μL-1mL cold protein extract, blow and mix well, and oscillate at 4℃ for 20-30min.
(5) Centrifuge at 4℃, 12000-14000×g, for 10min.
(6) Inhale the supernatant into another pre-cooled clean centrifuge tube to obtain the total blood cell protein.

Analysis of Common Problems:

1. Low protein concentration?

Some tissue samples may not be fully lysed when treated, resulting in low protein concentration. 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.

2. What method is used to quantify 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.

3. Is the extracted protein active?

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

Note:

1. This kit is intended for scientific research only and is not intended for diagnosis or treatment.
2. 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.
3. All samples and exposed glassware should be disposed of in accordance with the prescribed procedure after the experiment is completed.
4. Avoid skin or mucous membranes coming into contact with the reagent.