Platelet Protein Extraction Kit

Cat: EX1210
Size: 50T/100T
Storage: 2-8℃, valid for 1 year.

Kit Components:

Kit Components	50T	100T	Storage
Reagent A: Protein Extract	25mL	50mL	2-8℃
Reagent B: Platelet Washing Solution	25mL	50mL	2-8℃
Reagent C: 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°C. Take it out of the refrigerator and return to room temperature or 37°C 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:

Platelet protein extraction kit is suitable for extracting total protein from various animal platelets. The extraction process is simple and convenient and can be completed within 1h.

This kit contains a unique formula that effectively dissolves the membrane components. 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.

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 1h
2. Containing protein stabilizer, the extracted protein is stable.

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. The following methods of platelet collection are for reference only. Automatic platelet separator can be used to separate platelets when conditions permit, and gel filtration can be used to collect platelets to reduce the degree of activation. Methods commonly used in the laboratory or selected according to literature can also be used.
6. The recovery rate of platelet protein is about 70-80%.
7. Do not mix with other brands of reagents, otherwise it will affect the effect of use.
8. Contamination of the sample or reagent with bacteria or fungi or cross-contamination of reagents may result in false results.

Second, platelet protein extraction

1. Take 1-5mL of ACD anticoagulant blood and centrifuge at room temperature 200×g for 10min.
2. Collect the upper plasma and discard the lower blood cell precipitate layer.
3. Centrifuge the upper plasma layer at room temperature 200×g for 5min.
4. Centrifuge the upper plasma layer at room temperature 3000×g for 10min.
5. Discard the supernatant, add 500μL platelet washing solution B to the precipitate and wash once, centrifuge 3000×g for 10min.
6. Discard the supernatant, add 200-500μL cold protein extract solution A and 2μL protease inhibitor mixture into the precipitation, blow and mix well, and shake at 4℃ for 20min.
7. Centrifuge at 4℃, 12000-16000×g, for 15min.
8. Inhale the supernatant into another pre-cooled clean centrifuge tube to obtain the platelet protein.
9. The protein extract was quantified and divided into -80℃ refrigerator for reserve or directly used in downstream experiment.

Analysis of Common Problems:

1. Low protein concentration?

Processing part of the tissue 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.

2. 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.

3. 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.

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 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.