Item number: EX2261
Specification: 50T/100T
Store: 2-8℃ (Store at 2-8℃ before using protease inhibitor without lid, store at -20℃ after using lid)

Kit composition

Components	50T	100T	Save
Reagent A: Mitochondrial extract Solution A	55mL	110mL	2-8℃
Reagent B: Mitochondrial extract B	27mL	55mL	2-8℃
Reagent C: protein extract solution C	11mL	22mL	2-8℃
Reagent D: protease inhibitor mixture D	100μL	100μL×2	- 20℃
Instructions	1 copy	1 copy	

Product introduction

Mitochondria are organelles in eukaryotic cells covered by two layers of membranes. They are important organelle structures in cells to produce energy and are the main places for aerobic respiration. The energy substances in the cell -- fat, sugar and some amino acids are finally oxidized here, and ATP is generated by coupling phosphorylation to supply the physiological activities of the cell. The study of mitochondrial structure and function is usually carried out in isolated mitochondria.

The plant mitochondrial protein extraction kit provides a complete set of reagents, which can be extracted in a simple and rapid way within 1 hour.

The kit contains a unique formula that effectively dissolves mitochondrial membrane components. The kit contains a protease inhibitor mixture that prevents 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.

Experimental Steps

1. Extraction solution preparation: Add 2μL of reagent D to every 200μL of cold reagent C, mix well and put on ice for use.
2. Take 200-300mg of fresh plant sample leaves, wash and dry with PBS, then remove leaf stems and coarse veins. Use surgical scissors to cut up as much as possible.
3. Fully homogenize with A homogenizer after adding 1mL of reagent A or fully homogenize with a homogenizer/homogenizer after adding the right amount of reagent A.
4. Centrifuge the homogenate at 4℃, 100×g, for 1min.
5. Transfer the supernatant to the new tube with the 1mL gun tip. Centrifuge 500×g for 5min, discard the precipitation, and take the supernatant.

6. Transfer the supernatant to a new tube, centrifuge 800×g for 5min, discard the precipitation, and collect the supernatant.
7. Transfer the supernatant to the new tube, centrifuge 2000×g for 5min, discard the precipitation, and collect the supernatant.
8. The supernatant was transferred to the new tube and centrifuged at 12000×g for 20min. Discard the supernatant and collect the precipitation.
9. 500μL reagent B was added to the precipitate for re-suspension.
10. Centrifuge the suspension at 12000×g for 20min, discard the supernatant, and collect the precipitation.
11. Add 100-200μL of reagent C to the precipitation and mix thoroughly. Oscillate at 4℃ for 20-40min.
12. Centrifuge at 4℃, 14000×g for 15min.
13. Inhale the supernatant into another pre-cooled clean centrifuge tube to obtain mitochondrial protein.
14. The protein extract was quantified and divided into -80℃ refrigerator for reserve or directly used in downstream experiment.

Matters needing attention

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 touched glassware should be disposed of according to the prescribed procedure after completion.
4. Avoid skin or mucous membranes coming into contact with the reagent.

Related Products

R0020 Normal RIPA Lysate (tissue/cell)
PR1910 Rainbow 180 Broad Spectrum Protein Marker (11-180KD)
PC0020 BCA protein concentration determination kit
P1020 1 x PBS Buffer (pH7.2-7.4)
P10405 5 x Protein Loading buffer (including DTT)