

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

Catalog No: E-BC-E001

Specification: 100 Assays

Elabscience[®]Mitochondrial Extraction Assay 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

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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 mitochondrial from animal tissue.

Detection principle

The principle of mitochondrial separation and purification: after rupturing cells mechanically, the debris and huge organelles are removed by low-speed differential centrifugation, then purified mitochondria will be obtained by high-speed differential centrifugation.

Kit components & storage

Item	Component	Specification	Storage
Reagent 1	Lysis Buffer	60 mL × 1 vial	-20 °C, 12 months
Reagent 2	Mito-Wash Buffer	30 mL × 1 vial	-20 °C, 12 months, shading light
Reagent 3	Store Buffer	15 mL × 1 vial	-20 °C, 12 months

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

Place all reagents in ice water for pre-cooling for at least 15 min after they have returned to the solution state before use.

Operating steps

- ① Weight 0.05 g~0.1 g of fresh tissue and wash with PBS (0.01 M, pH 7.4) at 2-8°C to remove blood. Blot the water with absorbent paper. Cut the tissue into pieces with scissors and place them into a pre-cooled 5 mL glass homogenizer. Add 0.5 mL of pre-cooled lysis buffer. Grind the tissue up and down in the ice bath for about 30-40 times.
- ② Transfere tissue homogenate to a 2 mL pre-cooled EP tube, centrifuge at 500 g at 4 °C for 5 min.
- ③ Transferre the supernatant to a new 2 mL pre-cooled EP tube, centrifuge at 500 g at 4 °C for 5 min.
- ④ Transferre the supernatant to a new 2 mL pre-cooled EP tube, centrifuge at 15000 g at 4 °C for 15 min.
- ⑤ Discard the supernatant, resuspend the precipitate with 0.2 mL of pre-cooled mito-wash buffer, then centrifuge with 11000 g at 4 °C for 10 min.
- ⑥ Discard the supernatant, resuspend the precipitate with 50-100 µL of pre-cooled store buffer. The prepared mitochondrial extract can be used immediately or be stored at -70 °C , avoiding 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 carefully 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 should 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 results 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 calculate the possible usage of sample and reserve sufficient samples before use.