



PREP-FU Cell isolation Kit

REF P-006/2EU

General information

1 INTENDED USE

- 1.1 PREP-FU Cell isolation Kit is intended for isolation of lymphocytes from whole peripheral blood.
- 1.2 PREP-FU Cell isolation Kit can be used for research purposes.
- 1.3 PREP-FU Cell isolation Kit is designed for lymphocytes isolation from the 50 analyzed samples (including negative controls).

2 KIT FEATURES

- 2.1 Kit contents
 - Lymphocytes isolation solution - 50 tubes (500 µL in each).
- 2.2 Overall time needed to perform the analysis – 1 hour.

3 WARNING AND PRECAUTIONS

- 3.1 The laboratory makeup should comply with the requirements regulating work with microorganisms of I-IV classes of pathogenicity.
- 3.2 Handle and dispose all biological samples, reagents and materials used to carry out the assay as if they were able to transmit infective agents. Avoid direct contact with the biological samples reagents and materials used to carry out the assay. Any material coming in contact with the biological samples must be treated for at least 30 minutes with disinfecting solution or autoclaved for 1 hour at 121 °C before disposal.
- 3.3 Molecular biology procedures, such as nucleic acids extraction, reverse transcription, amplification and detection require qualified staff to avoid the risk of erroneous results, especially due to the degradation of nucleic acids contained in the samples or sample contamination by amplification products.
- 3.4 Plastic tubes do not contain phthalates. Do not breathe gas/fumes/vapour/spray produced by the components of the kit. Do not eat/drink components of the kit. Avoid contact with eyes. Do not use the kit after the expiry date provided. Only use the reagents provided in the kit and those recommended by manufacturer. Do not mix reagents from different batches. Do not use reagents from third party manufacturers' kits.
- 3.5 Significant health effects are NOT anticipated from routine use of this kit when adhering to the instructions listed in the current manual.

4 MATERIALS AND EQUIPMENT

The following equipment and materials are required when working with PREP-FU Cell isolation Kit:

- centrifuge for 1.5 mL tubes with max. RCF not lower than 13000g;
- microcentrifuge/vortex;
- refrigerator with a freezer chamber;
- pump with a trapping flask for supernatant removal;
- 1.5 mL plastic tubes;
- rack for 1.5 mL tubes;
- vacuum blood collection tubes (Vacuette for example), containing ethylenediaminetetraacetic acid disodium
- salt (EDTA) or sodium citrate anticoagulant;
- semi-automatic single channel adjustable volume pipettes which allow pipetting fluid volumes 200-1000 µL;
- disposable filter tips for 1000 µL automatic pipettes labeled RNAase-free, DNAase-free;
- disposable powder free textured gloves;
- container for the discharge of waste tips, tubes and other consumables;
- sterile saline (0,9% NaCl).

5 SAMPLE COLLECTION AND PREPARATION

5.1 Sample collection

Blood should be taken into 2.0 or 4.0 mL vacuum blood collection tubes (Vacuette for example), containing ethylenediaminetetraacetic acid disodium salt (EDTA) in final concentration of 2.0 mg/mL. Invert the tube 2-3 times to mix the blood with anticoagulant.



- 5.2 The use of heparin anticoagulant is not allowed.
Sample storage and transportation requirements

Storage of whole blood at room temperature (from 18 °C to 25 °C) for not longer than 24 hours is accepted.

6 LYMPHOCYTES ISOLATION

On that step use only "DNAse/RNAse free" labeled tips.

- 6.1 Mark two of 1.5 mL tubes (A and B) for each sample to be tested and negative control ("C-").
- 6.2 Add 500 µL of sterile buffered saline into the tubes marked A. Add 1000 µL of sterile buffered saline into the tubes marked B.
- 6.3 Add 500 µL of peripheral blood premixed by pipetting into the corresponding A tubes. Mix contents of tubes A by pipetting. Do not add blood to the "C-" tube.
- 6.4 Add 500 µL of sterile buffered saline into the "C-" A tube.
- 6.5 Carefully layer (add very slowly) 1.0 mL of tube A content onto the Lymphocytes isolation solution (Ficoll-Urographin). When layering do not mix the Ficoll and the diluted blood sample. Close tubes tightly.
- 6.6 Centrifuge the tubes at 4000 rpm for 20 min.
- 6.7 Transfer the interphase (approximately 500 µL) into the corresponding tubes B. Mix contents of tubes B by pipetting. Close tubes tightly.
- 6.8 Centrifuge the tubes at 4000 rpm for 10 min.
- 6.9 Remove supernatant completely avoiding contact of the pipette tip with the precipitate.
- 6.10 Add 500 µL of sterile buffered saline, close the tubes and vortex for 3-5 seconds.
- 6.11 Centrifuge the tubes at 4000 rpm for 10 min.
- 6.12 Remove supernatant completely avoiding contact of the pipette tip with the precipitate. Precipitate contains lymphocytes ready for nucleic acid extraction.



Isolated lymphocytes can be stored at temperature from minus 18 °C to minus 22 °C for no longer than 1 month.

7 STORAGE, SHIPPING AND HANDLING REQUIREMENTS

- 7.1 Shelf life – 6 months since the date of production in compliance with all transportation, storage and operation conditions.
- 7.2 The reagent kit must be stored at temperature from minus 18 °C to minus 22 °C over the storage period.
- 7.3 Repeated freezing-thawing cycles are not recommended. After thawing, store Lymphocytes isolation solution at the temperature between 2 °C and 8 °C for a month.
- 7.4 Transportation is possible for not more than 1 week at the temperature between 2 °C and 8 °C.
- 7.5 Strict compliance with the product instruction is necessary for kit performance assurance.

Contact our customer service department regarding quality issues with the kit:











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Key to symbols

	Temperature limitation		Consult instructions for use		Catalogue number
	Expiration date		Manufacturer		Batch code
	Date of manufacture		Number of tests		Do not expose to sunlight
	Caution				