Human Lymphocyte Separation Tube

Catalog Numbers: EXCM010 (15 mL tubes, 20 vials/pack)
EXCM011 (50 mL tubes, 25 vials/pack)

For Research Use Only. Not Intended for Diagnostic or Therapeutic Use.

EXREprotein™

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Product Description

This product is used for cell separation and purification by density gradient sedimentation according to the differences in cell densities with the help of the separation medium and centrifugation. After centrifugation, erythrocytes and granulocytes will have collected at bottom of the tube; PBMCs (peripheral blood mononuclear cells, including lymphocytes and monocytes) float on the surface of the separation medium, and a small part of cells may suspend in the separation medium. Lymphocytes are separated from the peripheral blood on the surface of the separation medium.

Shipping

Shipped at ambient temperature. Upon receipt, store immediately at 2 to 30°C and **protect from light**.

Storage

24 months from date of receipt at 2 to 30°C, **protected from light.**

Precautions

- Aseptic techniques should be followed when handling the product and cells.
- Protective clothing should be worn, and safe laboratory procedures should be followed.
- Blood should be collected using an anticoagulant at room temperature (20±5°C). Sample should not be stored longer than 2 hours at room temperature before use. Do not store sample refrigerated at 4°C.
- The optimum separation temperature is 20±5°C. Temperatures outside this range may affect separation.
- Blood does not require dilution prior to separation. Dilution will not affect the separation results when using the recommended volumes for the specific tube size.
- For a **15 mL separation tube, the recommended sample size is 3-6 mL**; and for a **50 mL separation tube, the recommended sample size is 15-25 mL**. Use of the separation tube is not recommended for volumes below the recommended range, and the dilution of the blood will not improve the separation effect on smaller volumes.
- If low PBMC layer or red blood cells are observed, to avoid cell loss, the washing frequency can be appropriately reduced to one wash. For other situations, it is recommended to wash twice.
- The main components are ficoll, diatrizoic acid and the polypropylene (PP) or polyethylene (PE) sieve plate.

Limitations

- This product is optimized to perform consistently. However, results may vary due to donor variability of primary cell populations and specific protocol design.
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Protocol

1. Take out the separation tube and observe whether there is free separation medium on the sieve plate material or air bubble below the sieve plate (Figure 1A). If yes, centrifuge at 800 x g for 1 minute at 20℃.
2. Pour blood, as shown in Figure 1B. The blood sample must be anticoagulant blood and does not require dilution.
3. Centrifuge at 800 x g for 15 minutes at 20℃. Set relatively low acceleration and deceleration (If there are ten gears and the tenth gear is the highest, acceleration and deceleration shall be adjusted to the third).
4. After suctioning out a portion of the plasma, directly pour the remaining small amount of plasma, PBMC layer, and a small amount of separation medium above the sieve plate into a clean centrifuge tube. As shown in Figure 1 (C), the PBMC layer is below the plasma and above the separation medium.
5. Wash with RPMI 1640 for 1-2 times (20℃, 250 x g, 10 minutes). Resuspend the lymphocytes with 0.9% (W/V) normal saline or a suitable culture medium for use.

Figure 1.

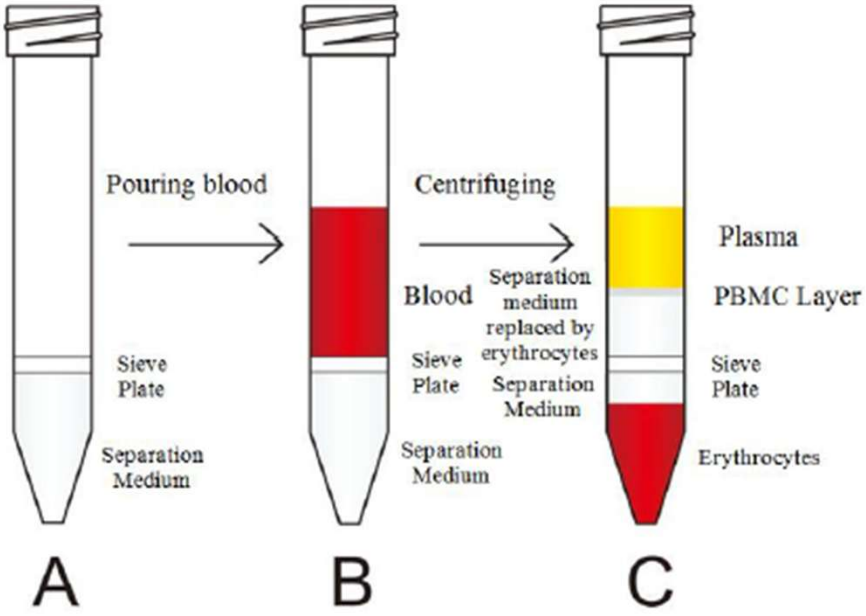

Figure 1: Process of Separating Blood with Lymphocyte Separation Tube for Human Peripheral Blood