

Lymphocyte Separation Medium

Product Name

English Name: Lymphocyte Separation Medium

Product Performance

Filling Volume/CatalogueNumber: 100mL/AS-37

200mL/AS-37L-200

Storage Conditions: Store at 4 - 30°C in the dark.

Expiry Date: 24months

Appearance: Liquid

Intended Use

This product is a sterile ready - to - use working solution, which is used for the isolation and purification of mononuclear cells from anticoagulated human peripheral blood, cord blood, or tissue samples. After opening, it should be stored at 4°C in the dark and used within one month. For research use only.

Instructions for Use

1. Dilute the blood sample 2 - 4 times with a sterile diluent such as PBS buffer or normal saline. Appropriately increasing the dilution factor of the blood sample can help improve the purity of mononuclear cells.
2. Add 15 - 20 mL of the separation medium into a 50 - mL conical - bottom centrifuge tube, and then slowly add 20 - 30 mL of the diluted whole blood or tissue cell suspension onto the surface of the separation medium.
3. Centrifuge at 400g for 15 - 30 minutes at room temperature, ensuring that the rotor speed decreases smoothly.
4. Take out the centrifuge tube and slowly aspirate the top layer, avoiding contact with the mononuclear cell layer.
5. Slowly transfer the mononuclear cell layer to another 50 - mL conical - bottom centrifuge tube.
6. Add sterile washing solution (normal saline, PBS buffer, etc.) and mix well. Then centrifuge at 300g for 10 minutes at room temperature and carefully discard the supernatant.
7. To remove residual platelets, resuspend the cells in 30 - 50 mL of sterile washing solution, centrifuge at 200g for 10 - 15 minutes at room temperature, and carefully discard the supernatant.
8. The above step is optional and will remove most of the platelets.
9. Resuspend the cells with buffer or culture medium for subsequent operations such as detection and culture.

Precautions

1. The SLymphocyte Separation Medium should be used at room temperature.
2. When separating mononuclear cells, the centrifuge speed should increase rapidly and decrease slowly to ensure the smooth descent of the rotor.

References

1. Se Jin Im, Masao Hashimoto, Michael Y Gerner, Junghwa Lee, Haydn T Kissick, Matheus C Burger, Qiang Shan, J Scott Hale, Judong Lee, Tahseen H Nasti, Arlene H Sharpe, Gordon J

Freeman, Ronald N Germain, Helder I Nakaya, Hai-Hui Xue, Rafi Ahmed, Defining CD8⁺ T cells that provide the proliferative burst after PD-1 therapy, *Nature*, 15(9) (2016):417-421.

2. G Atsaldi, G C Astaldi, U Topuz, L Guarina, Lymphocyte immunological patterns in leukaemia: a review, *Haematologia (Budap)*, 9(1-2)1975:21-33.