

## Various Animal Peripheral Blood Mononuclear Cell Isolation Solution<sup>V02</sup>

**Size:** 3X200mL/kit

**Storage:** This product is sensitive to light, should avoid light storage at room temperature, shelf life of 2 years. After sterile opening, save at room temperature.

### Product Introduction

This product is a sterile, low endotoxin level density gradient separation liquid used for separating peripheral blood mononuclear cells (PBMCs) from animals. The separation principle is based on the difference in density between blood cells, which allows cells of a certain density to distribute according to the corresponding density gradient through centrifugation, thus separating PBMCs from peripheral blood. It is suitable for separating PBMCs from anticoagulated blood of animals. The PBMCs separated under sterile conditions can be used for culture.

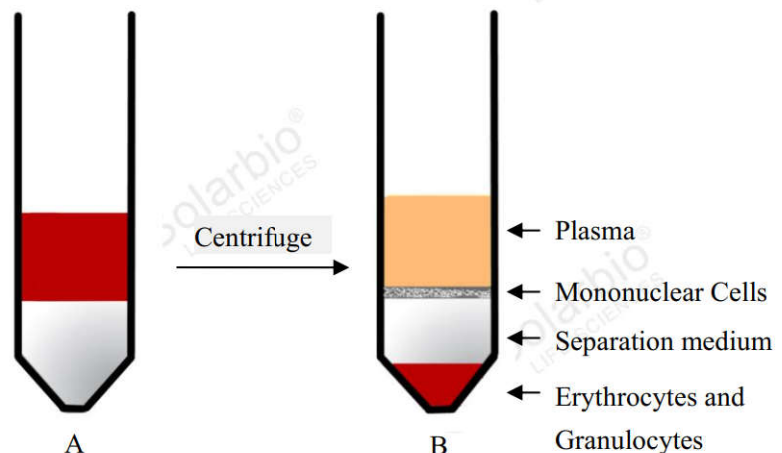
This product is for research use only.

### Kit compositions

Various Animal Peripheral Blood Mononuclear Cell Isolation	200mL
Whole blood and tissue dilution solution	200mL
Cell washing solution	200mL

### Protocols(only for reference)

- 1、 Collect blood in a tube containing anticoagulant (EDTA or heparin) or use defibrinated blood. Dilute the blood with an equal volume of Dilution buffer or phosphate buffered saline solution.
- 2、 Add an appropriate amount of separation solution to the centrifuge tube (when the volume of diluted blood is less than 3mL, add 3mL of separation solution; 3mL or more, add an equal volume of separation solution. But the total volume of the two can not exceed two-thirds of the centrifuge tube, otherwise it will affect the separation effect), the diluted blood is tiled to the separation liquid above the liquid level, pay attention to keep the interface of the two liquid levels clear. (You can use a Pasteurized straw to draw the blood, and then carefully spread the blood on the separation solution, because the density difference between the two will form an obvious layered interface. If more samples are added for a long time, it is normal for red blood cells to agglomerate and sink before centrifugation.)
- 3、 At room temperature, horizontal rotor 500~1000g, centrifugation for 20~30minutes (the larger the volume of blood, the larger the centrifugal force required, the longer the centrifugation time, the best separation conditions need to explore, the maximum centrifugal speed does not exceed 1200g).
- 4、 After centrifugation, there will be obvious stratification: the top layer is the diluted plasma layer, the middle is the clear separation liquid layer, the white membrane layer between the plasma and the separation liquid is the lymphocyte layer, and the bottom of the centrifuge tube is the red blood cells and granulocytes.
- 5、 Carefully suck the tunica albuginea cells into a 15mL clean centrifuge tube, and wash the tunica albuginea cells with 10mL PBS or cell wash solution. Centrifuge at 250g for 10minutes.
- 6、 The supernatant was discarded, and the cells were resuspended by adding 5mL of cell wash solution, 250g, and centrifuged for 10minutes.
- 7、 The supernatant was discarded and the cells were resuspended for later use.







## Note

- A. Mix it upside down before opening. This separation solution is a sterile product. In order to prolong the storage time of the separation solution, please unseal it under sterile conditions to avoid microbial contamination.
- B. The separation solution should always be kept at room temperature (18°C~25°C) when used. If the indoor temperature is low, the separation solution can be preheated. Centrifugation at 4°C or lower temperature may cause the white film layer to be unclear.
- C. Blood samples should preferably be fresh anticoagulated (within 2h of blood collection). In order to maintain the activity of neutrophils, freezing and cold storage should be avoided.
- D. Dilute blood or wash cells, do not use buffer and culture medium containing Ca, Mg ions, its formation will lead to blood cell agglutination, greatly reduce the cell yield and purity.
- E. Due to the electrostatic interaction of some plastic products (such as polystyrene), it may cause the cell to hang on the wall, affecting the separation effect.
- F. The viscosity or temperature difference of blood samples may affect the separation effect, so the number of centrifugation and centrifugation time can be adjusted to find the best separation condition.
- G. If the separated cells are to be further cultured, pay attention to maintain aseptic operation throughout the process to avoid microbial contamination.
- H. Excessive suction of cell layers and separation fluid layers can result in the removal of granulocytes at the interface between the separation fluid and the suction cup, thereby increasing the number of contaminated granulocytes. Excessive suction of plasma layers may result in contamination of granulocytes with plasma proteins and platelets.
- I. The cell dispersion coefficient and cell charge of different animal blood in different specific gravity separation solution are different, which should be mentioned when the user formulates the separation solution. The specific gravity of the required separation solution, the species of animal and the name of the cells to be separated should be provided.

## Related products

- YA0902 Disposable Pasteurized Straw
- S9020 Superior Fetal Bovine Serum
- 31800 RPMI Medium 1640
- A Variety of Other Animal and Other Cell Separations and Kits

## Reference

- [1] Boyum A. Separation of leucocytes from blood and bone marrow. Scand J Clin Lab Invest Suppl. 1968; 97: 7.
- [2] Ting A, Morris PJ. A technique for lymphocyte preparation from stored heparinized blood. Vox Sang. 1971 Jun; 20(6): 561-3.
- [3] Boyum A. Separation of Blood Leucocytes, Granulocytes and Lymphocytes Tissue Antigens. 1974; 4(4): 269-74.
- [4] Weisbart RH, Webb WF, Bluestone R, Goldberg LS. A simplified method for lymphocyte separation. Vox Sang. 1972; 23(5): 478-80.

Note: For more literature on the use of this product, please refer to Solarbio's official website.

