

### **Product Data Sheet**

Lymphocyte Separation Medium, Density 1.077 g/ml Product No. GB-LSM-100A, GB-LSM-500A

Packing. 100 ml, 500 ml

Storage. 2-8°C

#### **General Information:-**

The Lymphocyte Separation Medium (LSM) is a sterile, ready-to-use reagent for the in vitro isolation of mononuclear cells (lymphocytes and monocytes) from human whole blood, buffy coats, bone marrow and several other starting materials. The separation solution contains Ficoll™ density gradient media. Ficoll™ is a hydrophilic polymer with a molecular weight of 400 kDa. It is used to form multiple layers of different cell types during density gradient centrifugation. Erythrocytes and granulocytes sediment to the bottom of the tube and constitute the major content of the pellet. Mononuclear cells remain at the interphase between plasma and LSM.

# **Product Specifications:-**

Appearance	Clear liquid
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#### **Instructions for Use:-**

## **Separation of Mononuclear cells from Whole Blood:**

- Fill 50 ml size centrifuge tubes with 15 ml Lymphocyte Separation Medium (D = 1.077 g/ml, at +20°C).
- ♣ Dilute anticoagulated blood with equal parts of PBS and mix gently by inverting the tube.
- Carefully layer the diluted blood sample (20-25 ml) onto the 15 ml Lymphocyte Separation Medium. Important: Do not mix the LSM and the diluted blood sample.
- ♣ The separation process is performed by centrifugation at 800 g for 20 min at 18°C to 20°C. Important: brake should be turned off to maintain the separated layers.
- ♣ The lymphocytes and other mononuclear cells (70 % to 100 % enrichment) concentrate in the interphase (white cloudy layer) between the plasma and the separation solution.



- Transfer the white Interphase completely with a sterile Pasteur pipette in a new sterile 50 ml size tube.
- Fill up with PBS or culture medium and wash twice at 300 g for 5-10 min.
- ♣ Resuspend the cell pellet in the appropriate medium for further application. Cell counting can be done by using the standard methods.

### Reactivity and Stability:-

The reactivity and stability of Ficoll™ are based on its hydroxyl groups and on the glycoside bonds within the sucrose residues. It is stable in alkaline and neutral solutions. At pH values lower than 3, it is rapidly hydrolysed, especially at elevated temperatures. In neutral solutions, however, Ficoll™ can be sterilized by autoclaving at 110°C for 30 minutes, without affecting the reactivity. Avoid heavily oxidizing or reducing substances.

This product is for research use only.