

## NanoSep™ CD4 Beads

Size / Cat.No.: 2mL / TL-623

### Product Name

Generic Name NanoSep™ CD4 Beads

### Product Information

Endotoxin	< 2 EU/mL
Reactive species	human
Storage Temperature	2-8°C
Validity period	3 months

### Product Description

NanoSep™ CD4 beads are suitable for separation of human CD4<sup>+</sup> T cells and the size of the magnetic beads is 50nm. CD4<sup>+</sup> T cells can be separated by anti-human CD4 monoclonal antibody conjugated on the magnetic beads, which can specifically recognize CD4<sup>+</sup> T cells. NanoSep™ CD4 beads can be used for enrichment or depletion of CD4<sup>+</sup> T cells of PBMC efficiently.

### Protocol

- 1.1 Resuspend human PBMC cells in PBS buffer containing 1% HSA and count the cells. A total of 10<sup>7</sup> cells were taken into a 1.5mL Ep tube and centrifuge at 1500rpm for 5min.
- 1.2 Discard the supernatant and resuspend cells in 80 μL of MACS Running buffer (130-091-221). Add 20 μL of NanoSep™ CD4 beads, mix well and incubate for 15 minutes at 2–8°C.
- 1.3 Place LS column (130-042-401) in the magnetic field of a MACS Separator (130-042-303). Rinse twice with 1mL MACS Running buffer.
- 1.4 Add 1 mL of MACS Running buffer after the samples had completed incubation. Centrifuge at 1500rpm for 5 minutes. Discard supernatant completely.
- 1.5 Resuspend cells in 1mL MACS Running buffer and load cell suspension onto the column. Collect unlabeled cells which pass through and wash column with 1mL of MACS Running buffer. Perform washing steps twice. This is the unlabeled cell fraction.
- 1.6 Remove column from the separator and place it on a suitable collection tube (eg:15mL centrifuge tube). Pipette 3mL of MACS Running buffer onto the column. Immediately flush out fraction with the magnetically labeled cells by firmly applying the plunger supplied with the column.
- 1.7 Centrifuge the magnetically labeled cells suspension at 1500rpm for 5min.

1.8 Discard the supernatant and resuspend cells in 1mL of DPBS solution. The cells could be counted and detected by flow cytometry.

**Notice:**

1. The magnetic beads need to be thoroughly mixed when incubated with cells to improve separation efficiency.

**References**

1. David M Barrett, Nathan Singh, Xiaojun Liu, Shuguang Jiang, Carl H June, Stephan A Grupp, Yangbing Zhao (2014). Relation of clinical culture method to T-cell memory status and efficacy in xeno graft models of adoptive immunotherapy. *Cytotherapy*. 16(5):619-30.