

# LarSep Columns

## 1. Product Information

Product Name	Model	Size
LarSep Columns	HCSC-25	25 pcs/box
LarSep Columns	HCSC-10	10 pcs/box

## 2. Description

The LarSep columns are for research use only.

**Principle:** the column is placed in a strong and high-gradient magnetic field, the magnetic labeled cells stay in the column due to the magnetic force when passing through the column, while the unlabeled cells flow out. Thus achieving the purpose of separating or depleting cells. The separated cells can be directly used for downstream experiments such as cell culture and flow cytometry etc.

## 3. Specifications

- Capacity: recommended number of total cells:  $10^7 \sim 2 \times 10^9$ , recommended number of labeled cells:  $10^5 \sim 10^8$ . When separating cells larger than lymphocytes, capacity of the column may decrease.
- Make sure buffer added in the previous step drains away before adding buffer of the next step (i.e. no continuous droplets are dripping from the lower port of the column); the column should not run dry.
- Columns are sterilized and for single use.

## 4. Transportation and storage

Shipping at 10-35°C;

Store at 10-35°C, keep dry and protected from light. Valid for 3 years.

## 5. Requirements for reagents and instruments

Buffer: phosphate buffered saline(PBS) pH 7.2, containing 0.5% bovine serum albumin(BSA) and 2 mM EDTA

Streptavidin MicroBeads

30  $\mu$ m cell filter

**Note:** keep buffer at 2~8 °C. To prevent air bubbles from blocking the column, wash the column with buffer containing no air bubbles before separation.

## 6. Method for use

### 6.1 Preparation of columns

- (1) Put the column in a suitable magnetic field (LarSep Columns can be adapted to RWD LSC Separator, Cat No. LSC-S1 and Miltenyi MidiMACS™ Separator, Cat No.130-042-302, QuadroMACS™ Separator, Cat No.130-091-051).
- (2) Put a collection tube below the column.
- (3) Wash the column with 2mL buffer, collect the effluent, and make sure that there is no continuous droplet dripping from the lower port of the column.
- (4) Discard the effluent and replace the collection tube.

**Note:** the column should be used immediately after washing.

### 6.2 Magnetic separation

**Note:** before adding buffer in the following steps, make sure that all the buffer added in the previous step drains away (i.e. no continuous droplets are dripping from the lower port of the column).

- (1) Resuspend cells with 500 $\mu$ L buffer if there are no more than  $10^8$  cells. Increase buffer if there are more than  $10^8$  cells. (Filter sample before separation if cell concentration is too high or there are too much cell clumps and aggregates)

- (2) Add the cell suspension into the column and collect the effluent (containing unlabeled cells).
- (3) Wash the column with 2-3mL buffer and collect the effluent, and mix it with the effluent collected in step (2). Repeat washing for 2-3 times.
- (4) When buffer added in the previous step drains away, remove the column from the magnetic field and replace the collection tube with a new one.
- (5) Add 1~2 mL buffer into the column and then flush out the buffer with the plunger supplied with the column to obtain the magnetic labeled cells.
- (6) (Optional) To improve purity of the magnetic labeled cells, repeat steps (2) to (5) to enrich the magnetic labeled cells.

## 7. Precautions

- (1) The columns are valid for 3 years, and RWD does not guarantee its validity when it is expired.
- (2) All operations should be performed under sterilized conditions.
- (3) Do not combine the columns with other magnetic substances other than Streptavidin MicroBeads, or the sample may be damaged.
- (4) The columns are not suitable for samples larger than 30  $\mu\text{m}$ . Please filter and resuspend the cell suspension before magnetic separation.
- (5) Cell suspensions or buffers with cell clumps and aggregates may block the column and affect the separation. Filtering and dilution are recommended before separation.

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