Technical Data Sheet

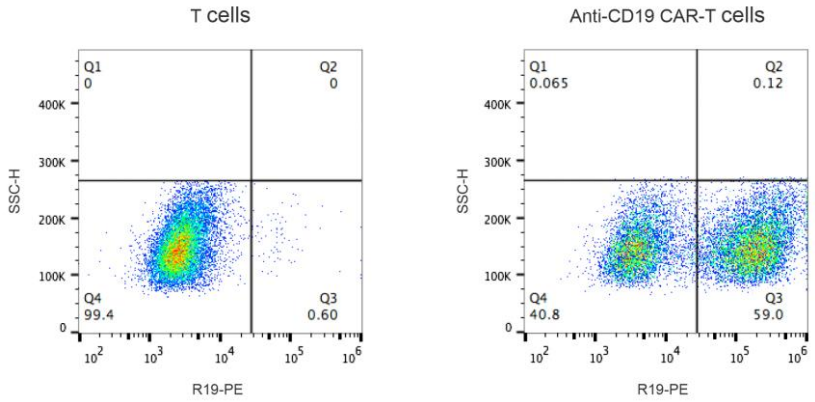
Rabbit Anti-Mouse FMC63 scFv Polyclonal Antibody, Biotin

Product Information

Material Number:	500014
RRID	AB_2857948
Size:	100 Tests
Vol. per Test:	1 μL
Antibody Types:	Polyclonal
Immunogen:	scFv region of a CD19-specific mouse mAb clone FMC63
Host Species:	Rabbit
Reactivity:	Mouse
Storage Buffer:	Aqueous buffered solution containing protein stabilizer and ⩽0.03% sodium azide

Description

The rabbit polyclonal antibody R19P specifically binds to the scFv region of a CD19-specific mouse monoclonal antibody (mAb, clone FMC63). CD19 antigen is a B-cell specific cell surface antigen, which is expressed in all B-cell lineage malignancies and normal B-cells. The scFv region of FMC63 has been used to develop CD19-specific chimeric antigen receptor (CAR) T cells utilized in clinical trials.



***Flow cytometric analysis of anti-CD19 CAR expression on human T cells**. Human T cells were lentivirally transduced with anti-CD19 CAR and cultured for 7 days. 2×10^5 cells were stained for the expression of anti-CD19 CAR with Rabbit Anti-Mouse FMC63 scFv Polyclonal Antibody, Biotin (Cat. No. 500014, right panel). Secondary staining was carried out with Streptavidin PE (Cat. No. SAPE-100). Non-transduced T cells were used as a control for gating of CAR expression (left panel). Acquisition of >10, 000 events was performed.*

Preparation and Storage

Shipped at 4°C. Store at 4°C short term (1-2 weeks). Store at -20°C in small aliquots for long term storage. Avoid freeze/thaw cycle.
The polyclonal antibody was purified by Protein A.
The antibody was conjugated with biotin under optimum conditions, and unreacted biotin was removed.

Application Notes

Application

Flow cytometry	Routinely Tested

Product Notices

1. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
2. An isotype control should be used at the same concentration as the antibody of interest.
3. Caution: Sodium azide yields highly toxic hydrazoic acid under acidic conditions. Dilute azide compounds in running water before discarding to avoid accumulation of potentially explosive deposits in plumbing.

FACS Protocol

1. Harvest the cells and wash the cells once by FACS buffer (PBS containing 2% of BSA).

2. Count the cells number and the viability, aliquot up to 2×10^5 live cells into each tube. (Note: the cell viability must be ≥ 95%.)
3. Resuspend cells in 100 μL of diluted Blocking Reagent (Cat. No. BLR-100, 1:100 diluted in FACS buffer) for 45 min at 4°C.
4. Wash the cells once by FACS buffer.
5. Resuspend cells in 100 μL of diluted Rabbit Anti-Mouse FMC63 scFv Polyclonal Antibody, Biotin (Cat. No. 500014, 1:100 diluted in FACS buffer) for 30 min at 4°C.
6. Wash the cells once by FACS buffer.
7. Resuspend cells in 100 μL of diluted Streptavidin PE (Cat. No. SAPE-100, 1:200 diluted in FACS buffer) for 30 min at 4°C.
8. Wash the cells 3 times by FACS buffer and resuspend the cells in 200 μL PBS per sample.
9. Transfer the cells into flow tube and analyze on Flow Cytometer. Acquisition of >10, 000 events is performed.