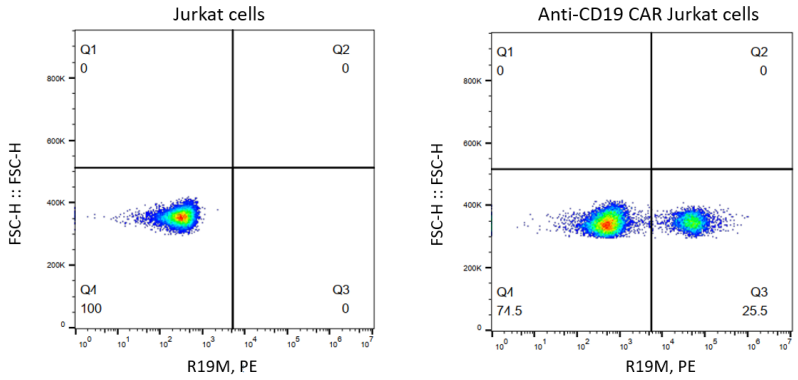
Technical Data Sheet

Rabbit Anti-Mouse FMC63 scFv Monoclonal Antibody, PE

Product Information	
Product No.	200105
Size	25 Tests
Recommended Vol. per Test	1 μL
Antibody Types	Monoclonal
Antibody Format	Whole IgG
Clone	R19M
Immunogen	scFv region of a CD19-specific mouse mAb clone FMC63
Conjugate	PE
Excitation/Emission Max	488/578nm，561/578nm
Host Species	Rabbit
Reactivity	Mouse
Storage Buffer	Aqueous buffered solution containing protein stabilizer and ≤0.05% ProClin 300
Storage conditions	2-8°C, store in dark

Description

The rabbit monoclonal antibody R19M 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 cell line Jurkat cells. *Jurkat cells were transduced with lentivirus encoding anti-CD19 CAR and cultured. 2×10^5 cells were stained for the expression of Rabbit Anti-Mouse FMC63 scFv Monoclonal Antibody, PE (Product No. 200105, right panel). Non-transduced Jurkat cells were used as a control for gating of CAR expression (left panel).*

Preparation & Storage

- Store undiluted at 2-8°C.
- Avoid prolonged exposure to light.
- The monoclonal antibody was purified by Protein A.
- The antibody was conjugated with PE under optimum conditions, and unincorporated dye was removed.

Application Notes

Application

Flow cytometry	Routinely Tested

Recommended Antibodies to Include in the Detection Process

Product name	Product No.
Anti-human CD45 Antibody	602148
Anti-human CD14 Antibody	602240
Anti-human CD8 Antibody	602044
Anti-human CD3 Antibody	603943/604043
Anti-human CD4 Antibody	604344
Rabbit Monoclonal IgG Isotype Control, PE	700306

FACS Protocol

- BioSwan reagents can be used with or without an isotype control to assess the amount of nonspecific antibody binding. If you want to set a control, the recommended reagent to use is the following: Rabbit IgG Isotype Control, PE (Product No. 700306).

(Optional) For Whole Blood Sample

1. Pipette 1 µL Rabbit Anti-Mouse FMC63 scFv Monoclonal Antibody, PE into the bottom of the tube.
2. Add dead cell staining solution and additional fluorochrome-conjugated antibodies into the bottom of the tube.
3. Pipette 100 µL of well-mixed, anticoagulated whole blood into the bottom of the tube. Mix gently and thoroughly.

   Note Avoid smearing sample down the side of the tube. If the sample remains on the side of the tube, it will not be stained with the reagents.
4. Incubate for 25 minutes in the dark at room temperature (18-25°C).
5. Pipette Red Blood Cell Lysis Solution to the tube. Mix gently and thoroughly. Incubate for 15 minutes in the dark at room temperature (18-25°C).
6. Add 500 µL FACS buffer to the tube. Mix well and centrifuge at 300g for 5 minutes at room temperature (18-25°C). Aspirate supernatant completely.
7. Repeat step 6 twice.
8. Add a suitable amount of FACS buffer to resuspend cell and analysis by flow cytometry.

(Optional) For Cell Sample

1. Harvest the cells and wash the cells twice by FACS buffer.
2. Count the cells number and the viability.
3. Resuspend the cell suspension to a concentration up to 1×10^6 nucleated cells per 100 µL of buffer.
4. Add 1 µL Rabbit Anti-Mouse FMC63 scFv Monoclonal Antibody, PE, dead cell staining solution and additional fluorochrome. Mix gently and thoroughly.
5. Incubate for 25 minutes in the dark at room temperature (18-25°C).
6. Add 500 µL FACS buffer to the tube. Mix well and centrifuge at 300g for 5 minutes at room temperature (18-25°C). Aspirate supernatant completely.
7. Repeat step 6 twice.
8. Add a suitable amount of FACS buffer to resuspend cell and analysis by flow cytometry.

Product Notices

1. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
2. Caution: Antibody solutions containing ProClin 300 should be handled with care. Do not take internally and avoid all contact with the skin, mucosa and eyes.

Intellectual Product Notices

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Application References

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3. Sangya Agarwal et al., "Production of Human CRISPR-Engineered CAR-T Cells," ***Journal of Visualized Experiments***, no. 169 (March 15, 2021): 62299, https://doi.org/10.3791/62299.
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