Alexa Fluor™ 647-Labeled Monoclonal Anti-FMC63 Antibody, Mouse IgG1 (Y45)Star Staining

Catalog # FM3-AM534

Source

Alexa Fluor 647-Labeled Monoclonal Anti-FMC63 Antibody, Mouse IgG1 (Y45) is produced via conjugation of AF647 to Monoclonal Anti-FMC63 Antibody, Mouse IgG1 under optimal conditions with a new generation site-specific technology under Star Staining labeling platform.

Application

Flow Cytometry (Evaluation of Anti-CD19 (FMC63 scFv) CAR Expression).

Clone

Y45

Species

Mouse

Isotype

Mouse IgG1 | Mouse Kappa

Specificity

Specifically recognizes the antigen-recognition domain of FMC63 derived CARs.

Immunogen

Recombinant FMC63 scFv derived from HEK293 cells.

Conjugate

AF647

Excitation Wavelength: 640 nm

Emission Wavelength: 672 nm

Recommended Dilution

1:50

Formulation

Lyophilized from 0.22 μm filtered solution in PBS, pH7.4 with trehalose as protectant.

Contact us for customized product form or formulation.

Reconstitution

Please see Certificate of Analysis for specific instructions.

For best performance, we strongly recommend you to follow the reconstitution protocol provided in the CoA.

Storage

For long term storage, the product should be stored at lyophilized state at -20°C or lower.

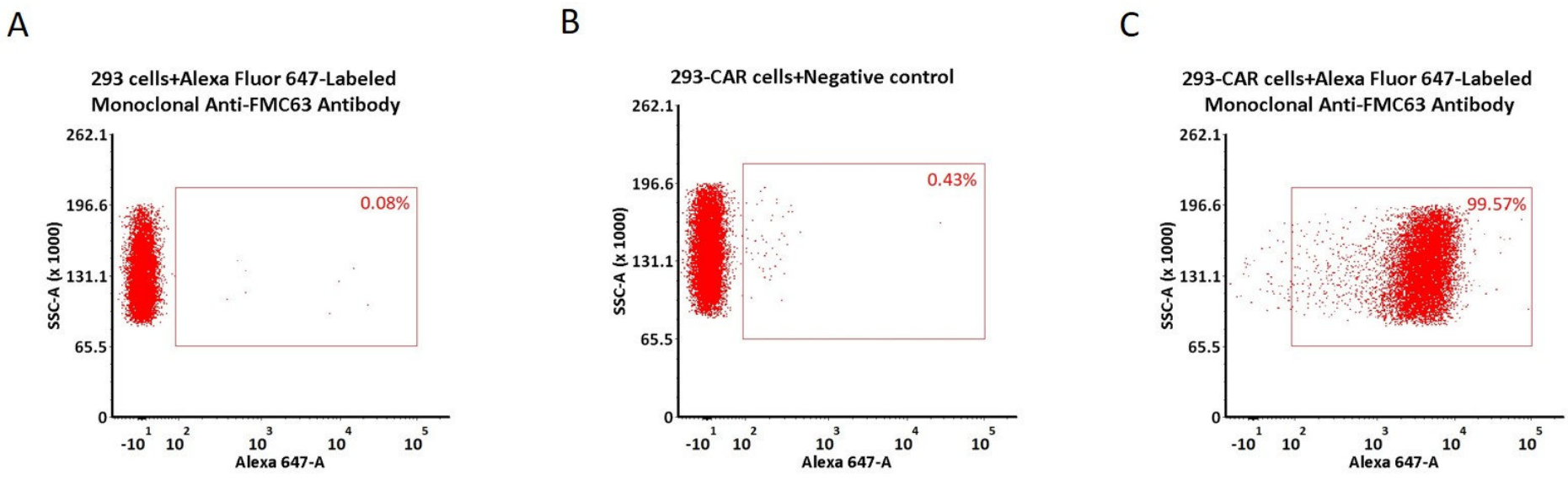
Please protect from light and avoid repeated freeze-thaw cycles.

This product is stable after storage at:

- -20°C to -70°C for 24 months in lyophilized state;
- -70°C for 12 months under sterile conditions after reconstitution.

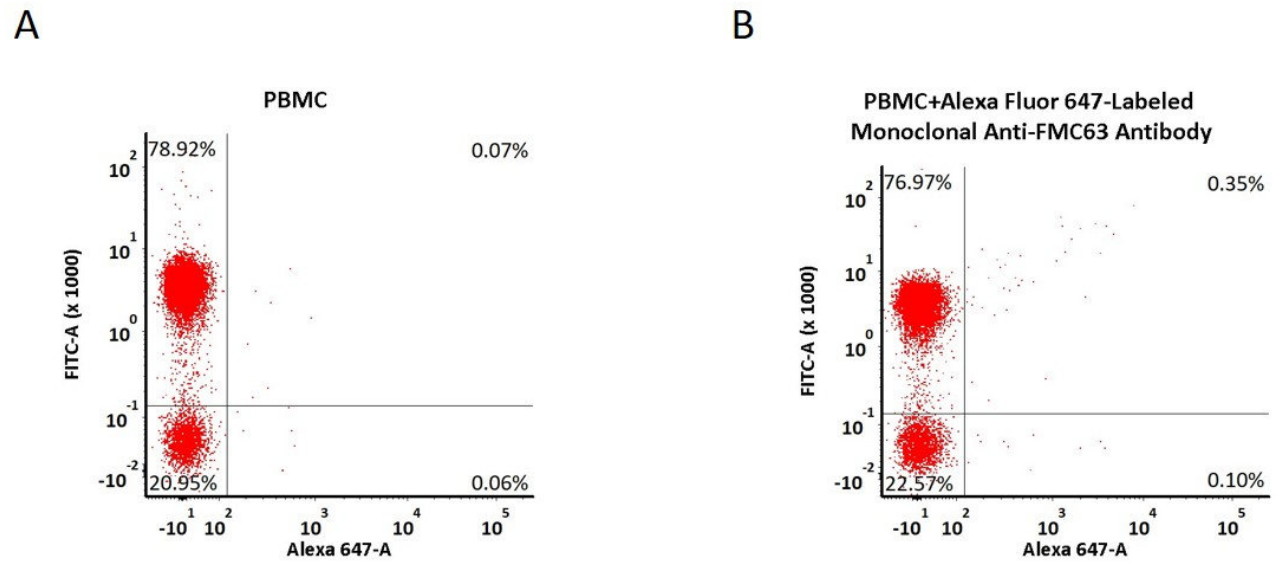
Evaluation of CAR expression

FACS Analysis of Anti-FMC63 CAR Expression



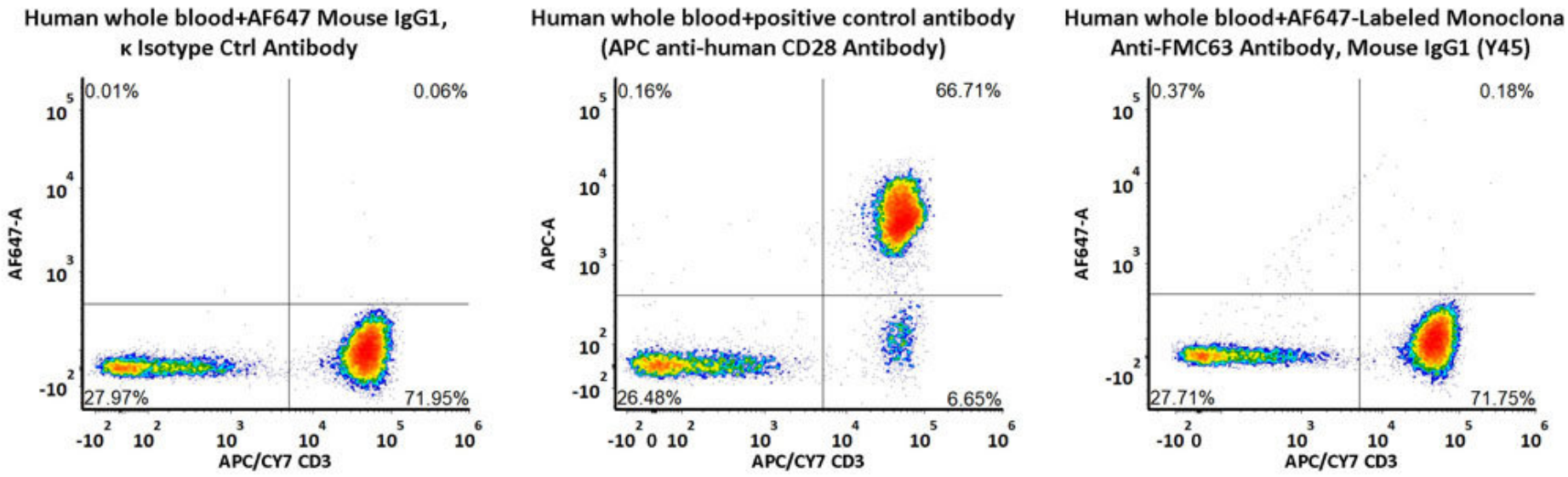
5e5 of anti-CD19 CAR-293 cells were stained with 100 µL of 1:50 dilution (2 µL stock solution in 100 µL FACS buffer) of Alexa Fluor 647-Labeled Monoclonal Anti-FMC63 Antibody, Mouse IgG1 (Y45) (Cat. No. FM3-AM534) and negative control protein respectively (Fig. C and B), and non-transfected 293 cells were used as a control (Fig. A). Alexa 647 signal was used to evaluate the binding activity (QC tested).

FACS Analysis of Non-specific binding to PBMCs



5e5 of PBMCs were stained with Alexa Fluor 647-Labeled Monoclonal Anti-FMC63 Antibody, Mouse IgG1 (Y45) (Cat. No. FM3-AM534) and anti-CD3 antibody, washed and then analyzed with FACS. FITC signal was used to evaluate the expression of CD3+ T cells in PBMCs, and Alexa 647 signal was used to evaluate the non-specific binding activity to PBMCs (QC tested).

FACS Analysis of Non-specific binding to Human whole blood



Non-specificity of Alexa Fluor™ 647-Labeled Monoclonal Anti-FMC63 Antibody, Mouse IgG1 (Y45) (Cat. No. FM3-AM534) binding to CD3+ cells present in human whole blood. 100 µl of human whole blood were simultaneously stained with APC/Cyanine7 anti-human CD3 Antibody and Alexa Fluor™ 647-Labeled Monoclonal Anti-FMC63 Antibody, Mouse IgG1 (Y45) (2 µL of the antibody stock solution in a final volume of 100 µL), compared with isotype control antibody and positive control antibody. Both APC/Cyanine7 and Alexa Fluor™ 647 positive signals was used to evaluate the non-specific binding activity to human CD3+ cells.

Background

FMC63 is an IgG2a mouse monoclonal antibody specific for CD19, which is a target for the immunotherapy of B lineage leukaemias and lymphomas. FMC63 scFv is the most commonly used ectodomain component of CD19-specific CARs. So far, most of reported CART19 trials contain the anti-CD19 scFv derived from FMC63, including the two FDA-approved CARs Kymriah and Yescarta.

Clinical and Translational Updates