Alexa Fluor™ 647-Labeled Human Siglec-3 / CD33 Protein, His TagStar Staining

Catalog # CD3-HA2H6

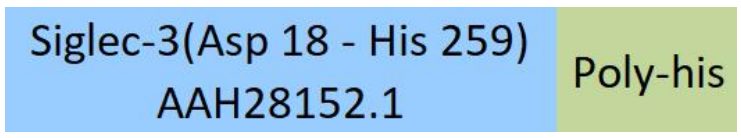
Synonym

CD33,SIGLEC3,gp67

Source

Alexa Fluor 647-Labeled Human Siglec-3, His Tag (CD3-HA2H6) is produced via conjugation of AF647 to Human Siglec-3, His Tag with a new generation site-specific technology under Star Staining labeling platform. Human Siglec-3, His Tag is expressed from human 293 cells (HEK293). It contains AA Asp 18 - His 259 (Accession # AAH28152.1).
Predicted N-terminus: Asp 18

Molecular Characterization



This protein carries a polyhistidine tag at the C-terminus.

Conjugate

AF647

Excitation Wavelength: 640 nm

Emission Wavelength: 672 nm

Purity

>95% as determined by SDS-PAGE.

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.

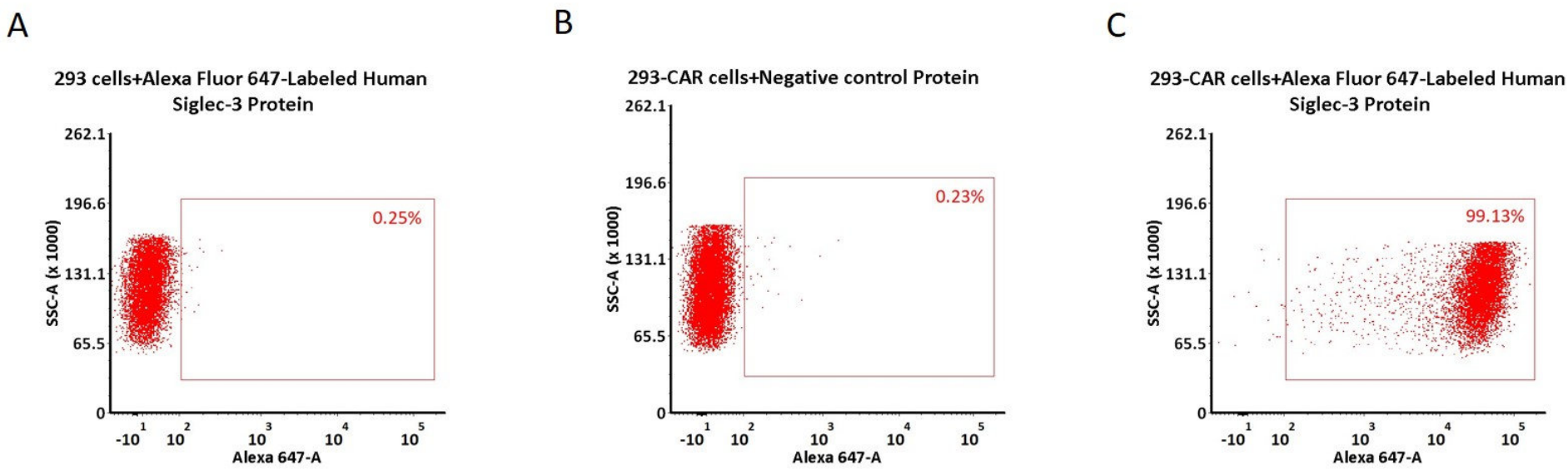
Please protect from light and avoid repeated freeze-thaw cycles.

This product is stable after storage at:

- -20°C to -70°C for 12 months in lyophilized state;
- -70°C for 3 months under sterile conditions after reconstitution.

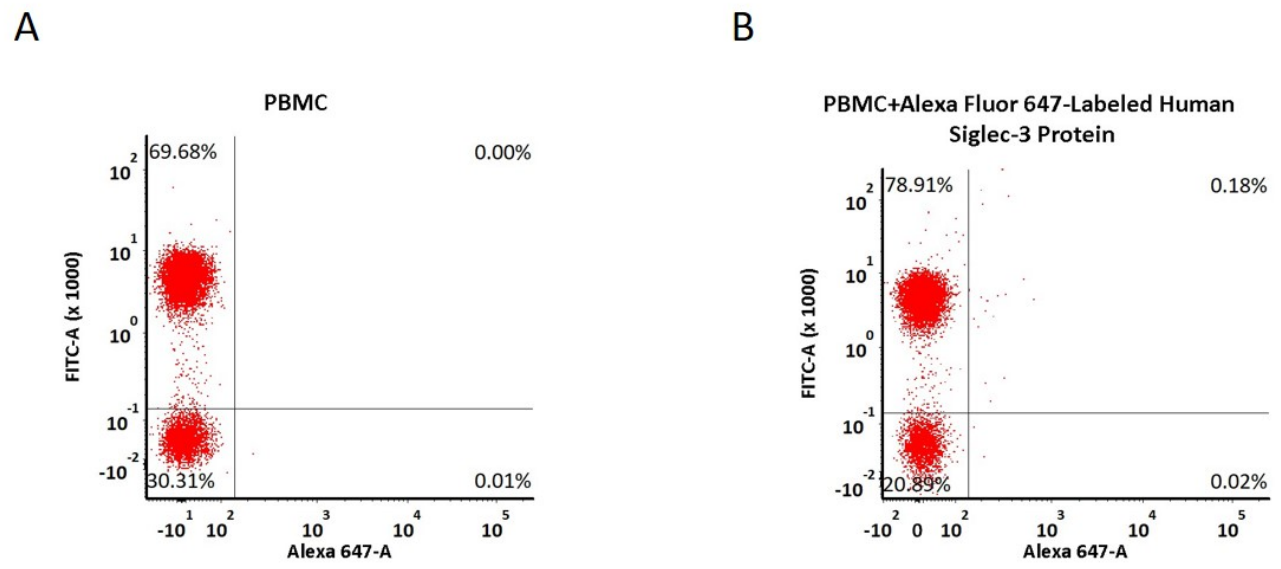
Evaluation of CAR expression

FACS Analysis of Anti-Siglec-3 CAR Expression



5e5 of 293 CAR cells transfected with anti-Siglec-3-scFv were stained with 100 μL of 3 μg/mL of Alexa Fluor 647-Labeled Human Siglec-3, His Tag (Cat. No. CD3-HA2H6) 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 Human Siglec-3, His Tag (Cat. No. CD3-HA2H6) 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).

Background

Myeloid cell surface antigen CD33 is also known as SIGLEC3, Siglecs (sialic acid binding Iglike lectins) and GP67, is a single-pass type I membrane protein which belongs to the immunoglobulin superfamily and SIGLEC (sialic acid binding Ig-like lectin) family. Human CD33 / Siglec-3 cDNA encodes a 364 amino acid (aa) polypeptide with a hydrophobic signal peptide, an N-terminal Ig-like V-type domain, one Ig-like C2-type domains, a transmembrane region and a cytoplasmic tail. CD33 / Siglec-3 usually considered myeloid-specific, but it can also be found on some lymphoid cells. In the immune response, CD33 / Siglec-3 may act as an inhibitory receptor upon ligand induced tyrosine phosphorylation by recruiting cytoplasmic phosphatase(s) via their SH2 domain(s) that block signal transduction through dephosphorylation of signaling molecules. CD33 / Siglec-3 induces apoptosis in acute myeloid leukemia.