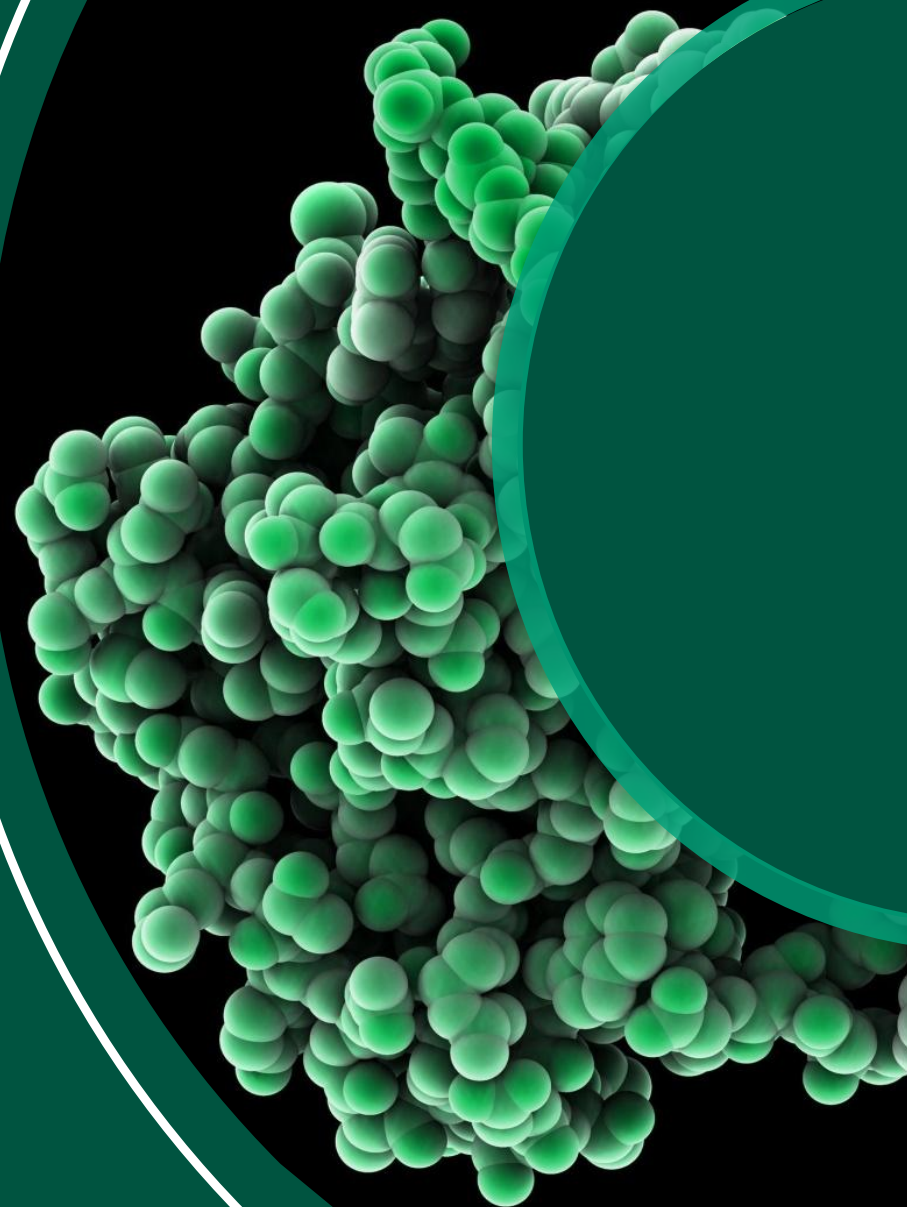


Site-Specific Labeling Technology Platform and Products



Why site-specific matters

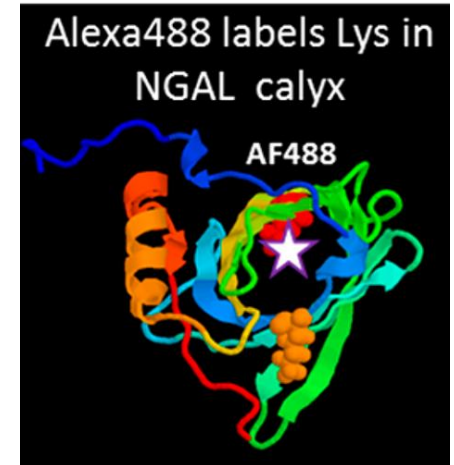
	Chemically Labeling	Site-Specific Labeling
Labeling site	Randomly attached (eg. Lys/Cys)	N- or C-terminus or both
Numbers of labels	Poorly controlled (3 - 10+)	1 or 2, precisely controlled
Impact on activity	May modify functional sites	Minimal/no impact
Homogeneity	High heterogeneity	High homogeneity
Reproducibility	Higher batch-to-batch variability	Consistent across batches
Typical usage	Applications where activity is less critical but stronger signal is desired (eg. WB, ELISA)	Applications requiring preserved protein activity and structural integrity (eg. Structural analysis, drug conjugation, single-molecule imaging...)

Why site-specific matters

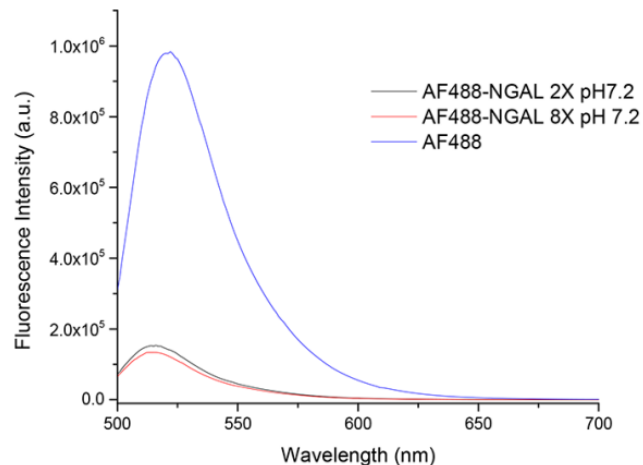
A case that random labeling interferes with activities

Table 2. Lists of Identified NGAL Peptides with AF488 Attachment from Sample AF488-NGAL (NHS) 2X, pH 7.2

peptide location	sequence	theoretical M.W., Da	experimental M.W., Da	relative signal intensity (%)
126–131	KVSQNR	730.4	1246.4	57
100–125	SYPLTSYLVRVVSTNYNQHAMVFFK	3020.5	3536.7	27
17–44	VPLQQNFQDNQFQGKQWYWGGLAGNALLR	3202.7	3718.7	6
74–82	KKKCDYWIR	1238.7	1754.7	3
132–141	EYFKITLYGR	1288.6	1804.7	2
74–76	KKK	402.3	918.3	2
83–110	TFVPGSQPGEFTLGNIKSYPLTSYLVR	3027.6	3543.6	1
100–141	SYPLTSYLVRVVSTNYNQHAMVFFKKVSQNREYFKITLYGR	5003.6	5519.7	1
100–143	SYPLTSYLVRVVSTNYNQHAMVFFKKVSQNREYFKITLYGRTK	5232.7	5748.8	<1



DOI: 10.1021/acs.analchem.8b01748 Anal. Chem. 2018, 90, 9695–9699



1. Random labeling can cause biased, site-preferred labelling
2. Random labeling can affect the activity if binds to active site
3. Random labeling would not always give higher signal intensity

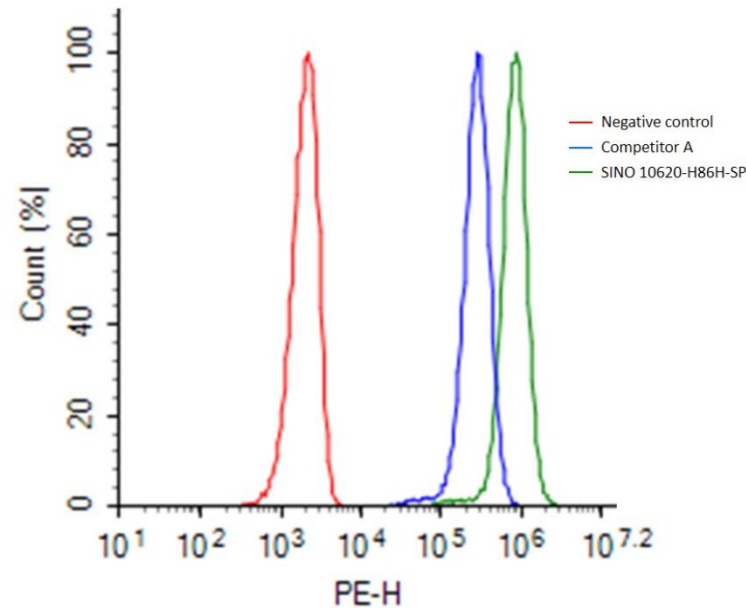
How we do site-specific labeling?



- ✓ **Labeling at a specific amino acid**
- ✓ **Precise labeling positions and quantities**
- ✓ **Maintain natural conformation and activity**
- ✓ **High consistency between and within batches**

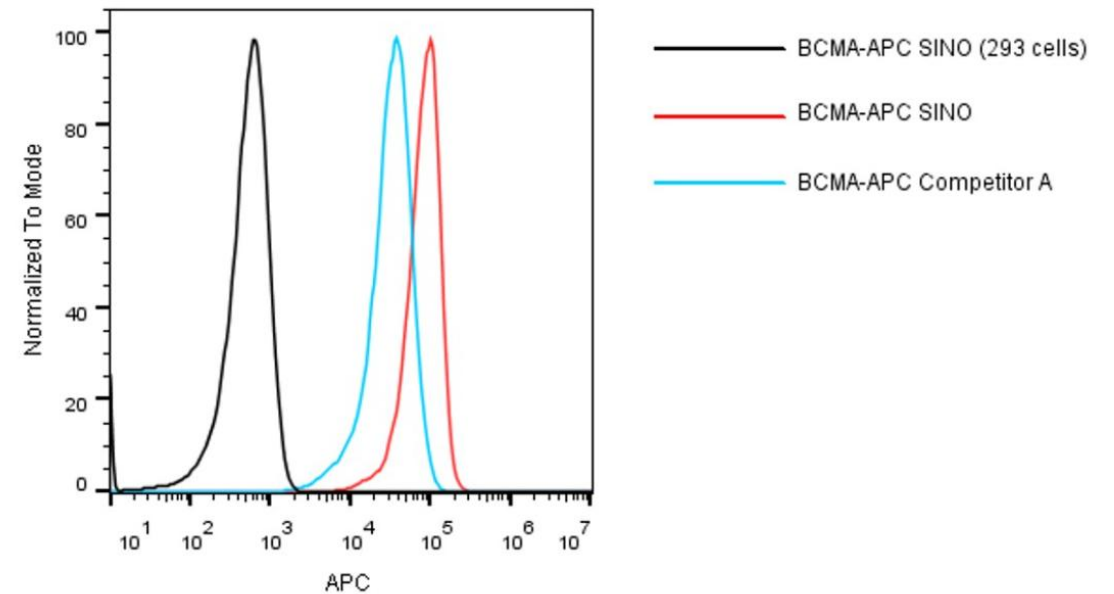
Higher efficiency

Recombinant Human BCMA Protein
(Site-Specific PE Conjugation, ECD, His Tag)
10620-H86H-SP



Flow cytometric analysis of anti-BCMA CAR expression. HEK293 cells were lentivirally transduced with anti-BCMA CAR. Non-transduced HEK293 cells were used as a control (shown on the left). 2 μ L (1 test) of site-specifically PE-conjugated human BCMA protein from two vendors (competitor A and SINO) was used. The results demonstrated that SINO's 10620-H86H-SP has a much higher fluorescence intensity than that of the competitor.

Recombinant Human BCMA Protein
(Site-Specific APC-Conjugated, ECD, His Tag)
10620-H86H-SA

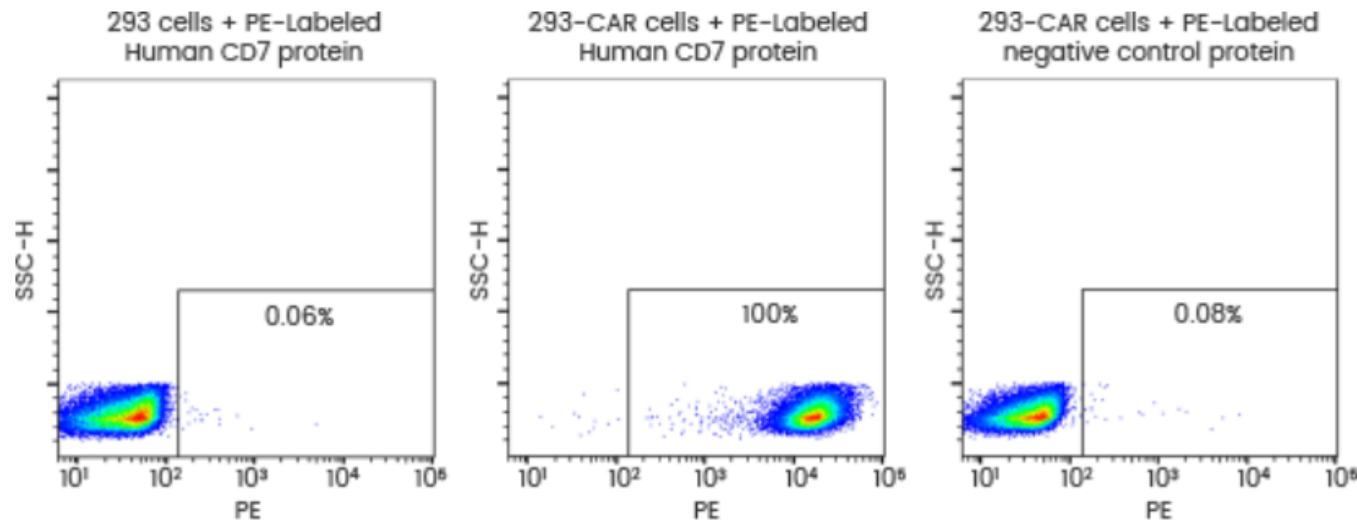


Flow cytometric analysis using APC conjugated BCMA protein. 5e5 of anti-BCMA CAR-293 cells were stained with APC-conjugated human BCMA protein from two vendors (competitor A and SINO). Non-transduced 293 cells were used as a control (Black Histogram). The results demonstrated that SINO's 10620-H86H-SA has a higher fluorescence intensity than that of the competitor.

Stringent Quality Control

CAR Cell Batch Release & Testing

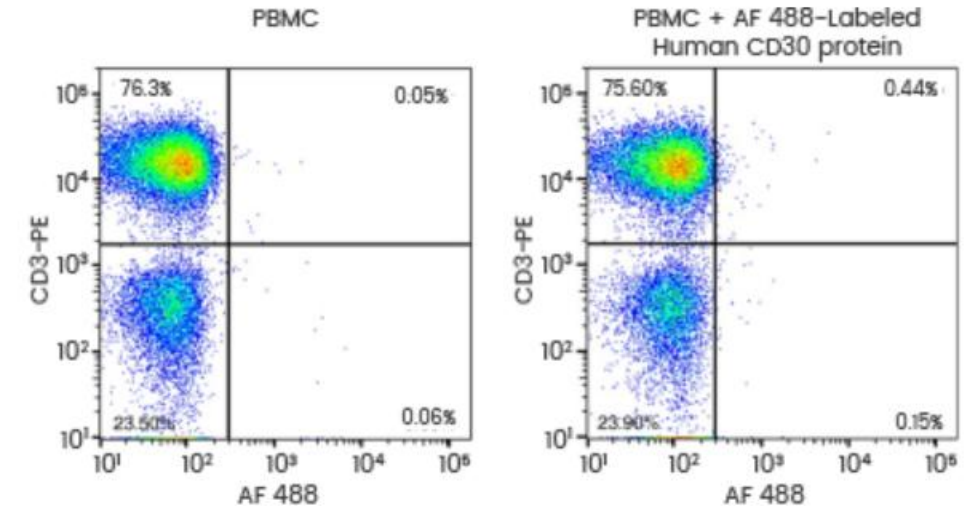
Human CD7 Protein
(Site-Specific PE Conjugation, His Tag)
Cat#: 11028-H86H-SP



Flow cytometric analysis of anti-CD7 CAR expression. 5×10^5 of anti-CD7 CAR-293 cells were stained with 100 μ L of 1:100 (1 μ L stock solution in 100 μ L FACS buffer) of PE-conjugated human CD7 Protein and negative control protein. Non-transduced 293 cells were used as a control (left). PE signal was used to evaluate the binding activity (QC tested).

PBMC Batch Release and Testing

Human CD30/TNFRSF8 Protein
(Site-Specific AF 488 Conjugation, His Tag)
Cat#: 10777-H86H-SD

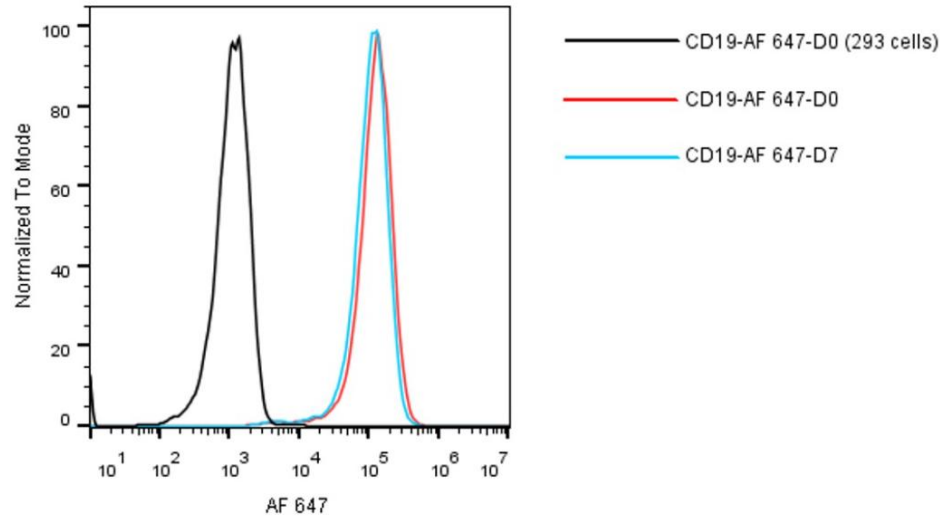


Binding activity of AF 488-conjugated CD30 protein to PBMC cells. PBMC cells were stained with PE conjugated anti-CD3 antibody and AF 488-conjugated human CD30 Protein and detected by flow cytometry. PE signal was used to evaluate the content of CD3+ T cells in PBMCs. AF 488 signal was used to evaluate the non-specific binding activity to PBMCs (QC tested).

Stringent Quality Control

Accelerated Stability

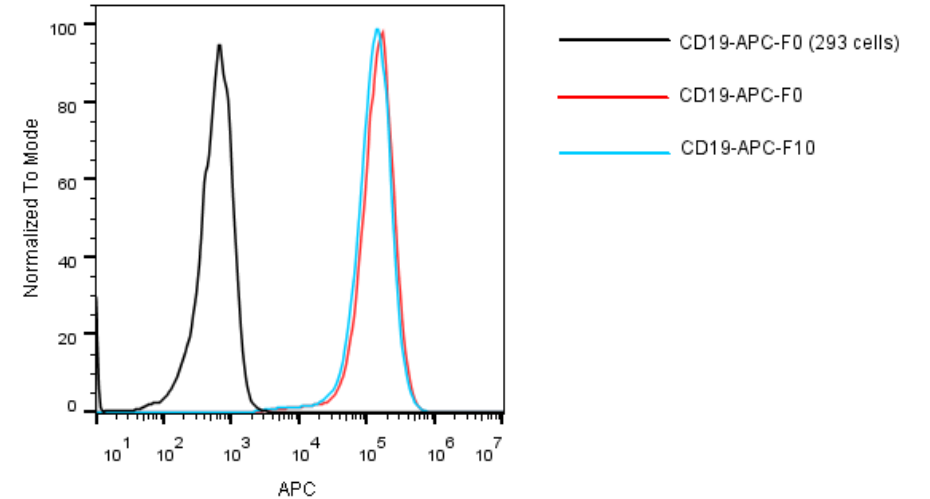
Recombinant Human CD19 Protein (Site-Specific AF 647-Conjugated, ECD, His Tag)
11880-H08W1-SG



Flow cytometric analysis using AF 647 conjugated CD19 protein. 5×10^5 of anti-CD19 CAR-293 cells were stained with AF 647-conjugated human CD19 protein (Site-Specific Conjugation, ECD, His Tag, Cat. No. 11880-H08W1-SG). Non-transduced 293 cells were used as a control (Black Histogram). The protein exhibited stable performance after 7 days of storage at 37°C , as measured on Day 0 and Day 7

Freeze-Thaw Stability

Recombinant Human CD19 Protein (Site-Specific APC-Conjugated, ECD, His Tag)
11880-H08W1-SA

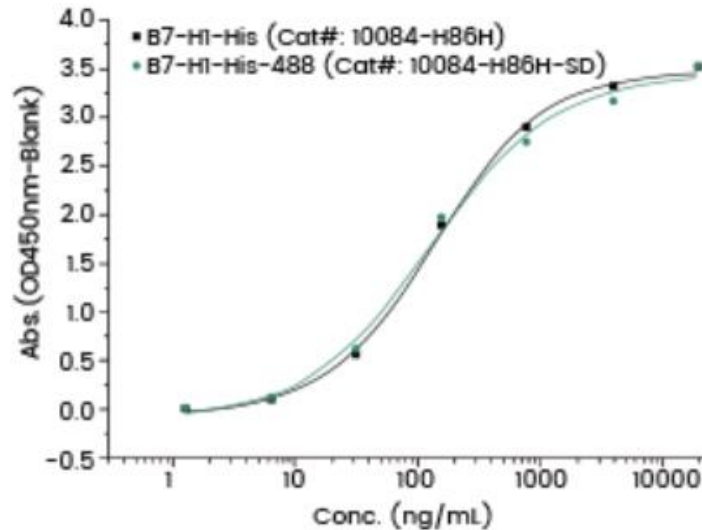


Flow cytometric analysis using APC conjugated CD19 protein. 5×10^5 of anti-CD19 CAR-293 cells were stained with APC-conjugated human CD19 protein (Site-Specific APC Conjugation, ECD, His Tag, Cat. No. 11880-H08W1-SA). Non-transduced 293 cells were used as a control (Black Histogram). The protein exhibited stable performance after multiple freeze-thaw cycles (10 cycles)

Stringent Quality Control

Consistent activity

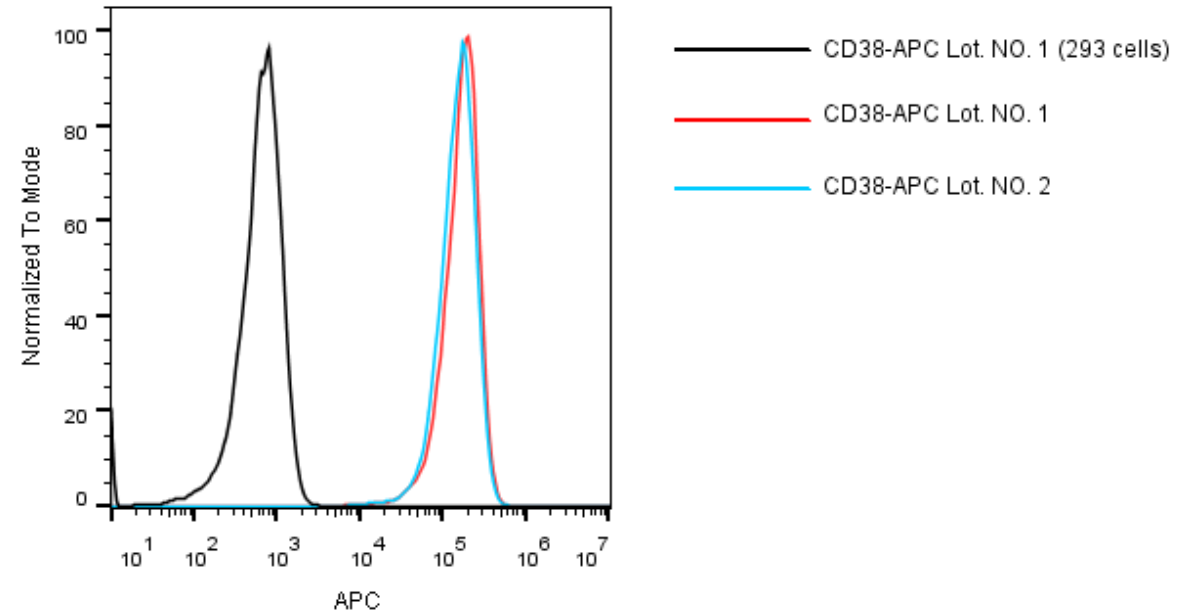
Human B7-H3 Protein
(Site-Specific AF 488 Conjugation)
Cat#: 11188-H86H-SD



Comparison of ELISA activity between pre-conjugation and different batches of site-specific AF 488 conjugated B7-H3 (routinely tested).

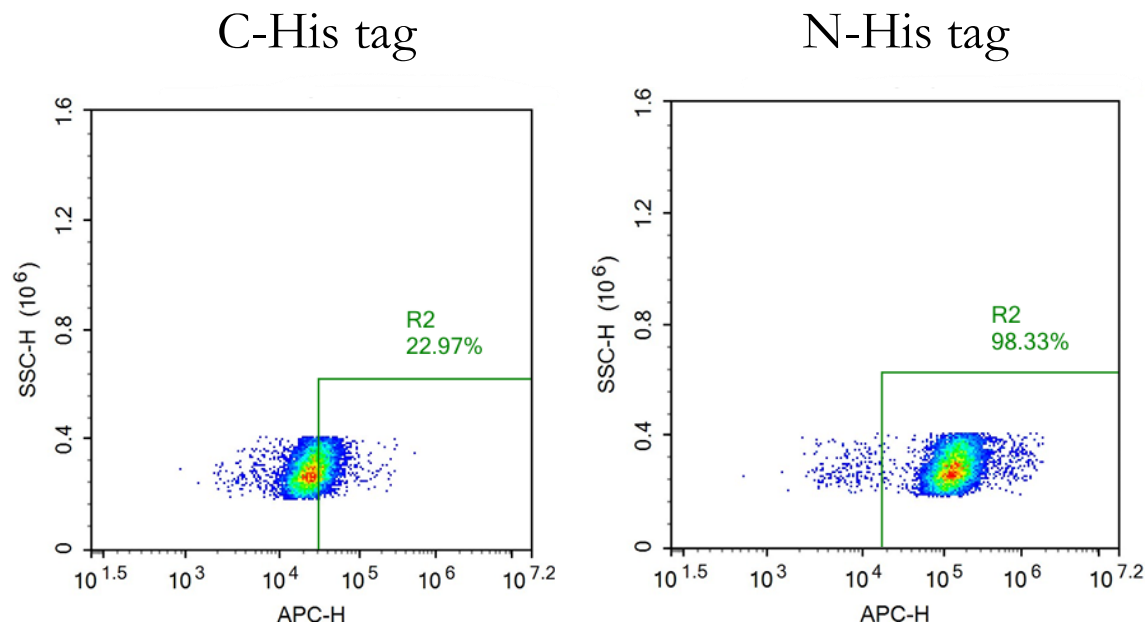
Lot-lot consistency

Recombinant Human CD38 Protein
(Site-Specific APC-Conjugated, ECD, His Tag)
10818-H85H-SA

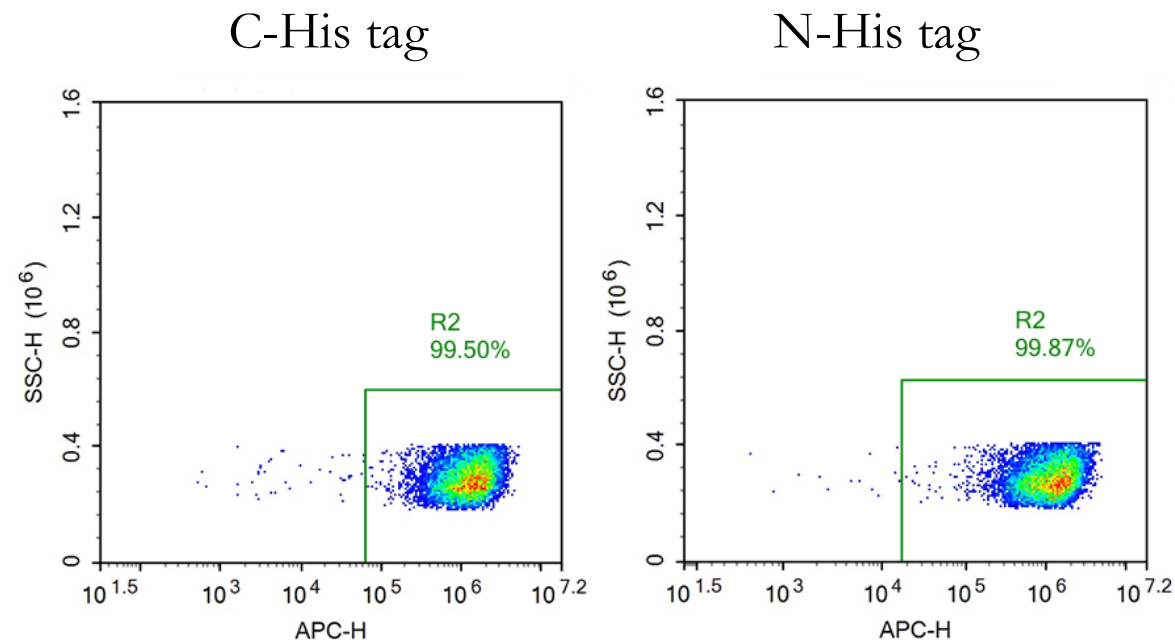


Flow cytometric analysis using different lots of APC conjugated CD38 protein. 5e5 of anti-CD38 CAR-293 cells were stained with 2 lots of APC-conjugated human CD38 protein (Site-Specific APC-Conjugated, ECD, His Tag, Cat. No. 10818-H85H-SA) at the same concentration. Non-transduced 293 cells were used as a control (Black Histogram). The results show high lot-to-lot consistency.

Special Product for Epitope-Tailored CAR Detection



For Anti C-terminal CD38 CAR (C-ter CD38 CAR sensitive)



For both Anti N-terminal and C-terminal CD38 CAR