

Technical Data Sheet

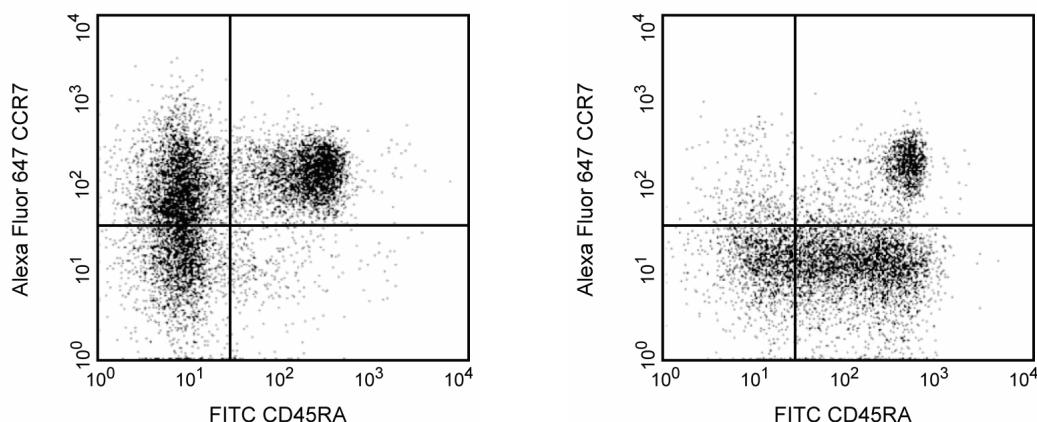
Alexa Fluor® 647 Rat Anti-Human CD197 (CCR7)

Product Information

Material Number:	557734
Alternate Name:	CCR7, BLR-2, EBI-1, CMKBR7
Size:	100 tests
Vol. per Test:	5 µl
Clone:	3D12
Immunogen:	Human CCR7 protein
Isotype:	Rat IgG2a, κ
Reactivity:	QC Testing: Human
Storage Buffer:	Aqueous buffered solution containing BSA and ≤0.09% sodium azide.

Description

The monoclonal antibody 3D12 reacts with the human CC chemokine receptor, CCR7. CCR7 (previously known as BLR-2, EBI-1 and CMKBR7), a seven-transmembrane, G-protein-coupled receptor, is the specific receptor for CC chemokines, MIP-3β/Exodus 3/ELC/ CCL19 and 6Ckine/Exodus 2/SLC/TCA4/CCL21. It has been shown that CCR7 mRNA is expressed mainly in lymphoid tissues including spleen, lymph nodes and tonsil. CCR7 mRNA was also detected in peripheral T and B lymphocytes, in bone marrow and cord blood CD34-positive cells and mature dendritic cells. The human CCR7 gene, unlike other CC chemokine receptor genes, has been mapped to chromosome 17q12. The immunogen used to generate 3D12 hybridoma was the N-terminus as well as parts of the second extracellular loop of human CCR7 protein. The monoclonal antibody 3D12 recognizes an epitope mapping to the N-terminus of human CCR7.



Detection of CCR7 expression on CD4 and CD8-positive human peripheral blood lymphocytes by Alexa Fluor® 647 conjugated anti-human CCR7 antibody 3D12. Human peripheral leukocytes were stained with 5 µl/test of Alexa Fluor® 647 conjugated 3D12 and anti-human CD45RA-FITC (Cat. No. 555488). The data shown are derived from the CD4-positive (based on staining with anti-human CD4-PE, Cat. No. 555347, left panel) and CD8-positive (based on staining with anti-human CD8-PE, Cat. No. 555367, right panel) lymphocyte gated populations and displayed as bivariate dot plots.

Preparation and Storage

The monoclonal antibody was purified from tissue culture supernatant or ascites by affinity chromatography.

The antibody was conjugated to Alexa Fluor® 647 under optimum conditions, and unreacted Alexa Fluor® 647 was removed.

Store undiluted at 4°C and protected from prolonged exposure to light. Do not freeze.

Application Notes

Application

Flow cytometry	Routinely Tested
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Recommended Assay Procedure:

Chemokine receptors are known to internalize during manipulation resulting in low frequency expression. Immunophenotyping studies of chemokine receptors need to be performed on freshly collected whole blood (<24 Hrs). Incubation with the antibody should be done in the dark. Cellular manipulation, such as Ficoll separation, freezing, or exposure to cold temperatures prior to staining have been shown to cause a decrease in staining intensity and inconsistent results.

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A multiple-step staining procedure is strongly recommended to amplify immunofluorescent signals in the flow cytometric analysis of human CCR7 expression. Purified anti-Human CCR7, clone 3D12, Cat. No. 552175 is recommended for detecting low frequency expression.

Suggested Companion Products

Catalog Number	Name	Size	Clone
557857	Alexa Fluor® 647 Rat IgG2a, κ Isotype Control	100 tests	R35-95
555488	FITC Mouse Anti-Human CD45RA	100 tests	HI100
555367	PE Mouse Anti-Human CD8	100 tests	RPA-T8
555347	PE Mouse Anti-Human CD4	100 tests	RPA-T4
552175	Purified Rat Anti-Human CD197 (CCR7)	0.1 mg	3D12

Product Notices

1. This reagent has been pre-diluted for use at the recommended Volume per Test. We typically use 1×10^6 cells in a 100-μl experimental sample (a test).
2. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.
3. For fluorochrome spectra and suitable instrument settings, please refer to our Fluorochrome Web Page at www.bdbiosciences.com/colors.
4. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
5. Alexa Fluor® 647 fluorochrome emission is collected at the same instrument settings as for allophycocyanin (APC).
6. The Alexa Fluor®, Pacific Blue™, and Cascade Blue® dye antibody conjugates in this product are sold under license from Molecular Probes, Inc. for research use only, excluding use in combination with microarrays, or as analyte specific reagents. The Alexa Fluor® dyes (except for Alexa Fluor® 430), Pacific Blue™ dye, and Cascade Blue® dye are covered by pending and issued patents.
7. Alexa Fluor is a registered trademark of Molecular Probes, Inc., Eugene, OR.
8. Source of all serum proteins is from USDA inspected abattoirs located in the United States.
9. 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.

References

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