# **Technical Data Sheet**

# BV605 Rat Anti-Human CD197 (CCR7)

#### **Product Information**

**Material Number:** 563711

Alternate Name: CCR7, BLR-2, EBI-1, CMKBR7

50 Tests Size Vol. per Test: 5 μl 3D12 Clone:

Human CCR7 protein Immunogen:

Isotype: Rat IgG2a, ĸ Reactivity: QC Testing: Human

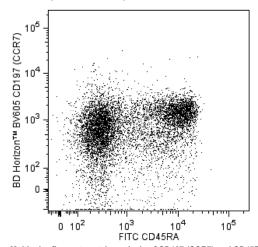
Storage Buffer: Aqueous buffered solution containing BSA and ≤0.09% sodium azide.

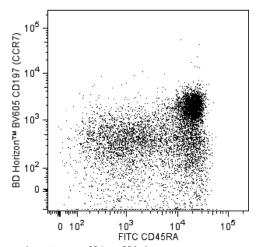
# Description

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.

This antibody is conjugated to BD Horizon BV605 which is part of the BD Horizon Brilliant<sup>™</sup> Violet family of dyes. With an Ex Max of 407-nm and Em Max of 602-nm, BD Horizon BV605 can be excited by a violet laser and detected with a standard 610/20-nm filter set. BD Horizon BV605 is a tandem fluorochrome of BD Horizon BV421 and an acceptor dye with an Em max at 605-nm. Due to the excitation of the acceptor dye by the green (532 nm) and yellow-green (561 nm) lasers, there will be significant spillover into the PE and BD Horizon PE-CF594 detectors off the green or yellow-green lasers. BD Horizon BV605 conjugates are very bright, often exhibiting brightness equivalent to PE conjugates and can be used as a third color off of the violet laser.

For optimal and reproducible results, BD Horizon Brilliant Stain Buffer should be used anytime two or more BD Horizon Brilliant dyes are used in the same experiment. Fluorescent dye interactions may cause staining artifacts which may affect data interpretation. The BD Horizon Brilliant Stain Buffer was designed to minimize these interactions. More information can be found in the Technical Data Sheet of the BD Horizon Brilliant Stain Buffer (Cat. No. 563794).





Multicolor flow cytometric analysis of CD197 (CCR7) and CD45RA coexpression patterns on CD4+ or CD8+ human peripheral blood lymphocytes. Human whole blood was stained with BD Horizon™ BV605 Rat Anti-Human CD197 (CCR7) (Cat. No. 563711), FITC Mouse Anti-Human CD45RA (Cat. No. 555488/561882), PerCP-Cy™5.5 Mouse Anti-Human CD4 (Cat. No. 560650), and Alexa Fluor® 647 Mouse Anti-Human CD8 (Cat. No. 557708) antibodies. Erythrocytes were lysed with BD FACS™ Lysing Solution (Cat. No. 349202). The two-color flow cytometric dot plots show the correlated expression patterns of CD45RA versus CD197 (CCR7) for CD4+ (Left Panel) or CD8+ (Right Panel) gated events with the forward and side light-scatter characteristics of intact lymphocytes. Flow cytometric analysis was performed using a BD™ LSR II Flow Cytometer

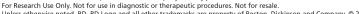
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#### **Preparation and Storage**

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

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

The antibody was conjugated with BD Horizon™ BV605 under optimum conditions, and unconjugated antibody and free BD Horizon™ BV605 were removed.

#### **Application Notes**

# Application

Flow cytometry	Routinely Tested

# **Suggested Companion Products**

Catalog Number	Name Name	Size	Clone	
554656	Stain Buffer (FBS)	500 mL	(none)	
563144	BV605 Rat IgG2a, κ Isotype Control	50 μg	R35-95	
560650	PerCP-Cy <sup>TM</sup> 5.5 Mouse Anti-Human CD4	50 Tests	RPA-T4	
557708	Alexa Fluor® 647 Mouse Anti-Human CD8	100 Tests	RPA-T8	
555488	FITC Mouse Anti-Human CD45RA	100 Tests	HI100	
561882	FITC Mouse Anti-Human CD45RA	25 Tests	HI100	
349202	BD FACS™ Lysing Solution	100 mL	(none)	
555899	Lysing Buffer	100 mL	(none)	
563794	Brilliant Stain Buffer	5 mL	(none)	

#### **Product Notices**

- 1. This reagent has been pre-diluted for use at the recommended Volume per Test. We typically use 1 × 10<sup>6</sup> cells in a 100-μl experimental sample (a test).
- 2. An isotype control should be used at the same concentration as the antibody of interest.
- 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.
- 4. Source of all serum proteins is from USDA inspected abattoirs located in the United States.
- 5. Please observe the following precautions: Absorption of visible light can significantly alter the energy transfer occurring in any tandem fluorochrome conjugate; therefore, we recommend that special precautions be taken (such as wrapping vials, tubes, or racks in aluminum foil) to prevent exposure of conjugated reagents, including cells stained with those reagents, to room illumination.
- Although every effort is made to minimize the lot-to-lot variation in the efficiency of the fluorochrome energy transfer, differences in the residual emission from BD Horizon<sup>TM</sup> BV421 may be observed. Therefore, we recommend that individual compensation controls be performed for every BD Horizon<sup>TM</sup> BV605 conjugate.
- 7. Alexa Fluor® is a registered trademark of Molecular Probes, Inc., Eugene, OR.
- 8. For fluorochrome spectra and suitable instrument settings, please refer to our Multicolor Flow Cytometry web page at www.bdbiosciences.com/colors.
- 9. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.
- 10. CFTM is a trademark of Biotium, Inc.

# References

Birkenbach M, Josefsen K, Yalamanchili R, Lenoir G, Kieff E. Epstein-Barr virus-induced genes: first lymphocyte-specific G protein-coupled peptide receptors. *Nature*. 1993; 67(4):2209-2220. (Biology)

Burgstahler R, Kempkes B, Steube K, Lipp M. Expression of the chemokine receptor BLR2/EBI1 is specifically transactivated by Epstein-Barr virus nuclear antigen 2. Biochem Biophys Res Commun. 1995; 215(2):737-743. (Biology)

Kim CH, Pelus LM, White JR, Broxmeyer HE. Macrophage-inflammatory protein-3 beta/EBI1-ligand chemokine/CK beta-11, a CC chemokine, is a chemoattractant with a specificity for macrophage progenitors among myeloid progenitor cells. *J Immunol*. 1998; 161(5):2580-2585. (Biology)

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Schweickart VL, Raport CJ, Godiska R, et al. Cloning of human and mouse EBI1, a lymphoid-specific G-protein-coupled receptor encoded on human chromosome 17q12-q21.2. *Genomics*. 1994; 23(3):643-650. (Biology)

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Yoshida R, Nagira M, Kitaura M, Imagawa N, Imai T, Yoshie O. Secondary lymphoid-tissue chemokine is a functional ligand for the CC chemokine receptor CCR7. J Biol Chem. 1998; 273(12):7118-7122. (Biology)

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