

Technical Data Sheet

BV605 Mouse Anti-Human CD73**Product Information**

Material Number:	563199
Alternate Name:	NT5E; 5' nucleotidase; 5'-NT; E5NT; Ecto-5'-nucleotidase; eN; eNT; NT; NT5
Size:	100 Tests
Vol. per Test:	5 µl
Clone:	AD2
Immunogen:	Pre-B leukemia cell line
Isotype:	Mouse IgG1, κ
Reactivity:	QC Testing: Human
Workshop:	V B-CD73.3
Storage Buffer:	Aqueous buffered solution containing BSA and ≤0.09% sodium azide.

Description

The AD2 monoclonal antibody specifically binds to ecto-5'-nucleotidase, a 70 kDa, glycosyl phosphatidylinositol (GPI)-anchored glycoprotein. CD73 is expressed on subsets of T and B lymphocytes, follicular dendritic cells, epithelial cells, endothelial cells and mesenchymal stem cells. Its expression on lymphocytes increases during T and B cell development. CD73 has enzymatic activity and catalyzes the dephosphorylation of adenosine monophosphate (AMP) converting it to adenosine. It has been suggested that CD73 can mediate costimulatory signals in T cell activation and adhesion of lymphocytes to endothelium.

This antibody is conjugated to BD Horizon BV605 which is part of the BD Horizon Brilliant™ 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).

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
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Suggested Companion Products

<u>Catalog Number</u>	<u>Name</u>	<u>Size</u>	<u>Clone</u>
554656	Stain Buffer (FBS)	500 mL	(none)
561527	Accutase™ Cell Detachment Solution	100 mL	(none)
562652	BV605 Mouse IgG1, κ Isotype Control	50 µg	X40
555899	Lysing Buffer	100 mL	(none)
563794	Brilliant Stain Buffer	5 mL	(none)

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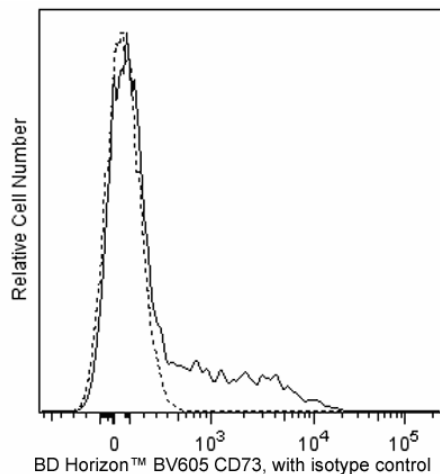
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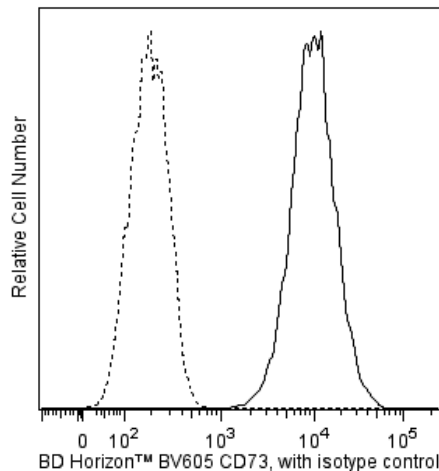
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Flow cytometric analysis of CD73 expression on human peripheral blood lymphocytes. Human whole blood was stained with BD Horizon™ BV605 Mouse Anti-Human CD73 antibody (Cat. No. 563199; solid line histogram) or with BD Horizon™ BV605 Mouse IgG1, κ Isotype Control (Cat. No. 562652; dashed line histogram). The erythrocytes were lysed with BD Pharm Lyse™ Lysing Buffer (Cat. No. 555899). The fluorescence histograms were derived from events with the forward and side light-scatter characteristics of viable lymphocytes. Flow cytometric analysis was performed using a BD™ LSR II Flow Cytometer System.



Flow cytometric analysis of CD73 expression on human mesenchymal stem cells. Human mesenchymal stem cells (Lonza) at passage 5 were harvested using Accutase™ Cell Detachment Solution (Cat. No. 561527) and stained with BD Horizon™ BV605 Mouse anti-Human CD73 antibody (Cat. No. 563199; solid line histogram) or a BD Horizon™ BV605 Mouse IgG1, κ Isotype Control (Cat. No. 562652; dashed line histogram). The fluorescence histograms were derived from gated events with the forward and side light-scatter characteristics of viable cells. Flow cytometry was performed using a BD LSRFortessa™ Flow Cytometry System.

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- μ l experimental sample (a test).
2. Source of all serum proteins is from USDA inspected abattoirs located in the United States.
3. An isotype control should be used at the same concentration as the antibody of interest.
4. Please refer to www.bdbiosciences.com/pharming/en/protocols for technical protocols.
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.
6. 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.
7. For fluorochrome spectra and suitable instrument settings, please refer to our Multicolor Flow Cytometry web page at www.bdbiosciences.com/colors.
8. 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™ BV421 may be observed. Therefore, we recommend that individual compensation controls be performed for every BD Horizon™ BV605 conjugate.
9. Accutase is a registered trademark of Innovative Cell Technologies, Inc.
10. CF™ is a trademark of Biotium, Inc.

References

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Nakamura T, Kubagawa H, Ohno T, Cooper MD. Characterization of an IgM Fc-binding receptor on human T cells. *J Immunol.* 1993; 151(12):6933-6941. (Clone-specific)

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Thomson LF, Ruedi JM, Glass A, et al. Production and characterization of monoclonal antibodies to the glycosyl phosphatidylinositol-anchored lymphocyte differentiation antigen ecto-5'-nucleotidase (CD73). *Tissue Antigens.* 1990; 35(1):9-19. (Biology)

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