

## Technical Data Sheet

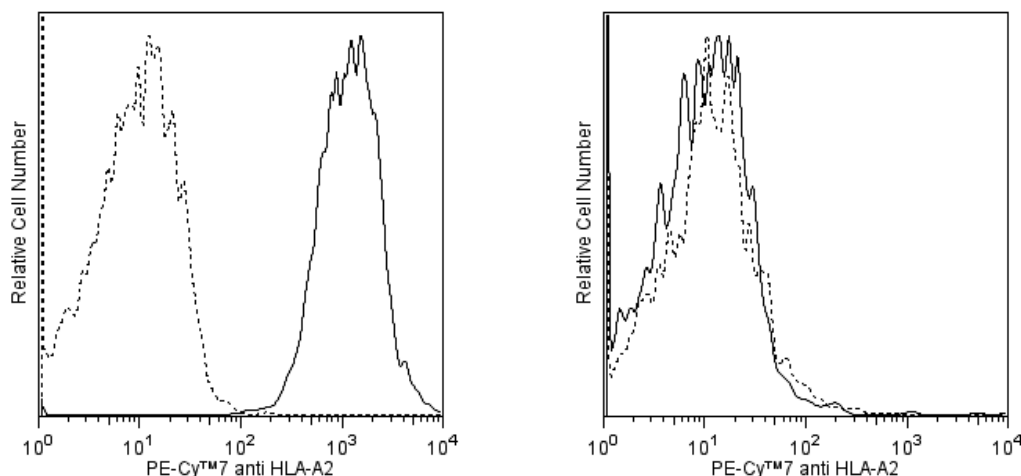
## PE-Cy™7 Anti-Human HLA-A2

## Product Information

Material Number:	561347
Alternate Name:	HLA class I histocompatibility antigen A2 alpha chain
Size:	50 tests
Vol. per Test:	5 µl
Clone:	BB7.2
Isotype:	Mouse IgG2b, κ
Reactivity:	QC Testing: Human
Storage Buffer:	Aqueous buffered solution containing BSA and ≤0.09% sodium azide.

## Description

The monoclonal antibody BB7.2 specifically binds to the α subunit of the human leukocyte antigen-A2 (HLA-A2), a class I molecule of the major histocompatibility complex (MHC). The MHC gene locus encodes a group of highly polymorphic, cell-surface proteins that play a broad role in the immune response to protein antigens. MHC molecules function by binding and presenting small antigenic protein fragments to antigen-specific receptors expressed by T cells (TCR). Human (*human leukocyte antigen*/HLA) MHC molecules comprise two major classes, MHC class I and class II. Functionally, class I MHC molecules can bind peptides derived from intracellular antigens (eg, viral and some bacterial antigens) that are specifically recognized by CD8+ T cells, and class II MHC molecules recognize antigens derived from pathogens multiplying in intracellular vesicles, and those derived from ingested extracellular bacteria. When presented on the cell surface by the MHC class II molecules, these antigens are recognized by CD4+ T cells. TCR recognize both processed peptides bound to MHC, as well as regions of the MHC molecule itself. CD4 and CD8 accessory molecules strengthen formation of the TCR-MHC complex through their interaction with non-polymorphic regions of the MHC molecule.



**Flow cytometric analysis of human HLA-A2 on lymphocytes from HLA-A2-positive and -negative donors.** Human whole blood from either an HLA-A2-positive (Left Panel) or an HLA-A2-negative (Right Panel) donor was stained with the PE-Cy™7 Mouse Anti-Human HLA-A2 antibody (Cat. No. 561347; solid line histogram) or with a PE-Cy™7 Mouse IgG2b, κ isotype control (Cat. No. 560542; dashed line histogram). The erythrocytes were lysed with BD PharmLyse™ Lysing Buffer (Cat. No. 555899). The fluorescence histograms were derived from events with the forward and side light-scatter characteristics of viable lymphocytes. Flow cytometry was performed using a BD™ LSR II Flow Cytometer System.

## Preparation and Storage

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

The antibody was conjugated with PE-Cy7 under optimum conditions, and unconjugated antibody and free PE-Cy7 were removed.

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

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## Application Notes

### Application

Flow cytometry

Routinely Tested

### Suggested Companion Products

Catalog Number	Name	Size	Clone
555899	Lysing Buffer	100 ml	(none)
554656	Stain Buffer (FBS)	500 ml	(none)
560542	PE-Cy <sup>TM</sup> 7 Mouse IgG2b, $\kappa$ Isotype Control	0.1 mg	27-35

### Product Notices

1. This reagent has been pre-diluted for use at the recommended Volume per Test. We typically use  $1 \times 10^6$  cells in a 100- $\mu$ l experimental sample (a test).
2. An isotype control should be used at the same concentration as the antibody of interest.
3. Source of all serum proteins is from USDA inspected abattoirs located in the United States.
4. Please refer to [www.bdbiosciences.com/pharmingen/protocols](http://www.bdbiosciences.com/pharmingen/protocols) for technical protocols.
5. Cy is a trademark of Amersham Biosciences Limited. This conjugated product is sold under license to the following patents: US Patent Nos. 5,486,616; 5,569,587; 5,569,766; 5,627,027.
6. 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.
7. 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.
8. For fluorochrome spectra and suitable instrument settings, please refer to our Fluorochrome Web Page at [www.bdbiosciences.com/colors](http://www.bdbiosciences.com/colors).
9. Warning: Some APC-Cy7 and PE-Cy7 conjugates show changes in their emission spectrum with prolonged exposure to formaldehyde. If you are unable to analyze fixed samples within four hours, we recommend that you use BD<sup>TM</sup> Stabilizing Fixative (Cat. No. 338036).
10. This product is subject to proprietary rights of Amersham Biosciences Corp. and Carnegie Mellon University and made and sold under license from Amersham Biosciences Corp. This product is licensed for sale only for research. It is not licensed for any other use. If you require a commercial license to use this product and do not have one return this material, unopened to BD Biosciences, 10975 Torreyana Rd, San Diego, CA 92121 and any money paid for the material will be refunded.
11. PE-Cy7 is a tandem fluorochrome composed of R-phycoerythrin (PE), which is excited by 488-nm light and serves as an energy donor, coupled to the cyanine dye Cy7, which acts as an energy acceptor and fluoresces maximally at 780 nm. PE-Cy7 tandem fluorochrome emission is collected in a detector for fluorescence wavelengths of 750 nm and higher. 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 PE may be observed. Therefore, we recommend that individual compensation controls be performed for every PE-Cy7 conjugate. PE-Cy7 is optimized for use with a single argon ion laser emitting 488-nm light, and there is no significant overlap between PE-Cy7 and FITC emission spectra. When using dual-laser cytometers, which may directly excite both PE and Cy7, we recommend the use of cross-beam compensation during data acquisition or software compensation during data analysis.

### References

Bjorkman PJ, Saper MA, Samraoui B, Bennett WS, Strominger JL, Wiley DC. Structure of the human class I histocompatibility antigen, HLA-A2. *Nature*. 1987; 329(6139):506-512. (Biology)

Bjorkman PJ, Saper MA, Samraoui B, Bennett WS, Strominger JL, Wiley DC. The foreign antigen binding site and T cell recognition regions of class I histocompatibility antigens. *Nature*. 1987; 329(6139):512-518. (Biology)

Romero P, Dunbar PR, Valmori D. Ex vivo staining of metastatic lymph nodes by class I major histocompatibility complex tetramers reveals high numbers of antigen-experienced tumor-specific cytolytic T lymphocytes. *J Exp Med*. 1998; 188(9):1641-1650. (Biology)