

Technical Data Sheet

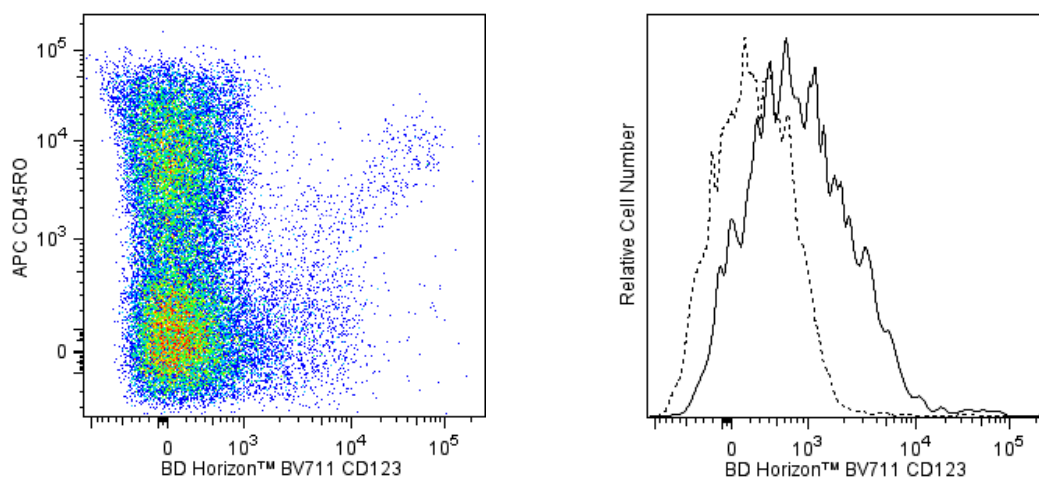
BV711 Mouse Anti-Human CD123**Product Information**

| | |
|-------------------------|---|
| Material Number: | 563161 |
| Alternate Name: | IL3RA; IL-3RA; IL-3R α ; IL-3R-alpha; Interleukin-3 receptor subunit alpha |
| Size: | 50 Tests |
| Vol. per Test: | 5 μ l |
| Clone: | 9F5 |
| Immunogen: | Human IL-3R α Transfected Cell Line |
| Isotype: | Mouse (BALB/c) IgG1, κ |
| Reactivity: | QC Testing: Human |
| Workshop: | VI C-67 |
| Storage Buffer: | Aqueous buffered solution containing BSA and $\leq 0.09\%$ sodium azide. |

Description

The 9F5 monoclonal antibody specifically binds to CD123. CD123 is the 70 kDa IL-3 receptor α chain (IL-3R α) that associates with the 120-140 kDa β subunit (CD131/Common β -chain/ β c) to form the functional IL-3 receptor complex. The β c chain is also shared with distinct α chain subunits to form the functional heterodimeric receptors for interleukins IL-5 and GM-CSF. IL-3R α is expressed on a subset of peripheral blood dendritic cells, myeloid precursors, basophils, mast cells, macrophages, and megakaryocytes. Reports indicate that IL-3R α is also expressed on lymphocytes. The IL-3R plays an important role in hematopoietic progenitor cell growth and differentiation. This antibody does not block binding of IL-3 to the IL-3 receptor.

The antibody was conjugated to BD Horizon BV711 which is part of the BD Horizon Brilliant™ Violet family of dyes. This dye is a tandem fluorochrome of BD Horizon BV421 with an Ex Max of 405-nm and an acceptor dye with an Em Max at 711-nm. BD Horizon BV711 can be excited by the violet laser and detected in a filter used to detect Cy™5.5 / Alexa Fluor® 700-like dyes (eg, 712/20-nm filter). Due to the excitation and emission characteristics of the acceptor dye, there may be moderate spillover into the Alexa Fluor® 700 and PerCP-Cy5.5 detectors. However, the spillover can be corrected through compensation as with any other dye combination.



Multicolor flow cytometric analysis of CD123 expression on human peripheral blood cells. Human whole blood was stained with APC Mouse Anti-Human CD45RO (Cat. No. 559865/560899) and FITC Mouse Anti-Human CD14 (Cat. No. 555397/557153/561712) antibodies and either BD Horizon™ BV711 Mouse IgG1, κ Isotype Control (Cat. No. 563044; dashed line histogram) or BD Horizon™ BV711 Mouse Anti-Human CD123 antibody (Cat. No. 563161; solid line histogram and dot plot). The erythrocytes were lysed with BD Pharm Lyse™ Lysing Buffer (Cat. No. 555899). The two-color flow cytometric dot plot (Left Panel) shows the correlated expression of CD123 versus CD45RO for gated events with the forward and side light-scatter characteristics of viable lymphocytes. The overlapping CD123 and Ig Isotype Control fluorescence histograms (Right Panel) was derived from CD14-positive gated events (monocytes). Flow cytometric analysis was performed using a BD™ LSR II Flow Cytometer System.

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563161 Rev. 2



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™ BV711 under optimum conditions, and unconjugated antibody and free BD Horizon™ BV711 were removed.

Application Notes

Application

Flow cytometry

Routinely Tested

Recommended Assay Procedure:

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. 563794/566349) or the BD Horizon Brilliant Stain Buffer Plus (Cat. No. 566385).

Suggested Companion Products

| Catalog Number | Name | Size | Clone |
|----------------|-------------------------------------|------------|--------|
| 554656 | Stain Buffer (FBS) | 500 mL | (none) |
| 563044 | BV711 Mouse IgG1, k Isotype Control | 50 µg | X40 |
| 555899 | Lysing Buffer | 100 mL | (none) |
| 559865 | APC Mouse Anti-Human CD45RO | 100 Tests | UCHL1 |
| 560899 | APC Mouse Anti-Human CD45RO | 25 Tests | UCHL1 |
| 555397 | FITC Mouse Anti-Human CD14 | 100 Tests | M5E2 |
| 557153 | FITC Mouse Anti-Human CD14 | 50 Tests | M5E2 |
| 561712 | FITC Mouse Anti-Human CD14 | 25 Tests | M5E2 |
| 349202 | BD FACSTM Lysing Solution | 100 mL | (none) |
| 554657 | Stain Buffer (BSA) | 500 mL | (none) |
| 566385 | Brilliant Stain Buffer Plus | 1000 Tests | (none) |
| 566349 | Brilliant Stain Buffer | 1000 Tests | (none) |
| 563794 | Brilliant Stain Buffer | 100 Tests | (none) |

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. 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. Alexa Fluor® is a registered trademark of Molecular Probes, Inc., Eugene, OR.
5. 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.
6. For fluorochrome spectra and suitable instrument settings, please refer to our Multicolor Flow Cytometry web page at www.bdbiosciences.com/colors.
7. BD Horizon Brilliant Violet 711 is covered by one or more of the following US patents: 8,110,673; 8,158,444; 8,227,187; 8,455,613; 8,575,303; 8,354,239.
8. BD Horizon Brilliant Stain Buffer is covered by one or more of the following US patents: 8,110,673; 8,158,444; 8,575,303; 8,354,239.
9. Cy is a trademark of GE Healthcare.
10. Please refer to www.bdbiosciences.com/pharming/protocols for technical protocols.

References

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Korpelainen EI, Gamble JR, Smith WB, et al. The receptor for interleukin 3 is selectively induced in human endothelial cells by tumor necrosis factor alpha and potentiates interleukin 8 secretion and neutrophil transmigration. *Proc Natl Acad Sci USA*. 1993; 90(23):11137-41. (Clone-specific: Flow cytometry, Immunofluorescence)

Macardie PJ, Chen Z, Shih CY, et al. Characterization of human leucocytes bearing the IL-3 receptor. *Cell Immunol*. 1996; 168(1):59-68. (Clone-specific: Flow cytometry)

Smith WB, Guida L, Sun Q, et al. Neutrophils activated by granulocyte-macrophage colony-stimulating factor express receptors for interleukin-3 which mediate class II expression. *Blood*. 1995; 86(10):3938-3944. (Clone-specific: Flow cytometry)

Sun Q, Woodcock JM, Rapoport A, et al. Monoclonal antibody 7G3 recognizes the N-terminal domain of the human interleukin-3 (IL-3) receptor alpha-chain and functions as a specific IL-3 receptor antagonist. *Blood*. 1996; 87(1):83-92. (Clone-specific: Immunoprecipitation, Western blot)