

## Technical Data Sheet

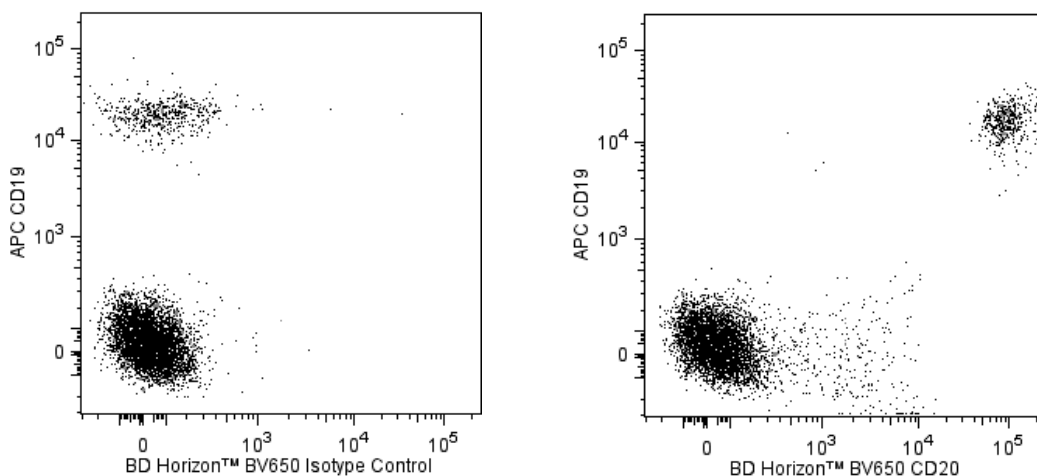
**BV650 Mouse Anti-Human CD20****Product Information**

<b>Material Number:</b>	<b>563780</b>
<b>Alternate Name:</b>	MS4A1; B1; Bp35; LEU-16; S7
<b>Size:</b>	100 tests
<b>Vol. per Test:</b>	5 µl
<b>Clone:</b>	2H7
<b>Immunogen:</b>	Human 6.16c1.3 B cell line
<b>Isotype:</b>	Mouse (C57BL/6) IgG2b, κ
<b>Reactivity:</b>	QC Testing: Human Tested in Development: Rhesus, Cynomolgus, Baboon
<b>Workshop:</b>	II B22; III B739, NL382; IV B201
<b>Storage Buffer:</b>	Aqueous buffered solution containing BSA and ≤0.09% sodium azide.

**Description**

The 2H7 monoclonal antibody specifically binds to CD20 that is encoded by the *MS4A1* (Membrane-spanning 4-domains, subfamily A, member 1) gene. CD20 is a 33-37 kDa unglycosylated four-transmembrane phosphoprotein. CD20 is expressed on pre-B-cells, resting and activated B cells and follicular dendritic cells but not on plasma cells. Low level CD20 expression is observed on a small subset of normal circulating T lymphocytes. The CD20 molecule is involved in the regulation of B-cell activation.

The antibody was conjugated to BD Horizon™ BV650 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 650-nm. BD Horizon™ BV650 can be excited by the violet laser and detected in a filter used to detect APC-like dyes (eg, 660/20-nm filter). Due to the excitation and emission characteristics of the acceptor dye, there will be spillover into the APC and Alexa Fluor® 700 detectors. However, the spillover can be corrected through compensation as with any other dye combination.



**Two-color flow cytometric analysis of CD20 expression on human peripheral blood lymphocytes.** Whole blood was stained with APC Mouse Anti-Human CD19 antibody (Cat. No. 555415/561742) and either BD Horizon™ BV650 Mouse IgG2b, κ Isotype Control (Cat. No. 563437; Left Panel) or BD Horizon™ BV650 Mouse Anti-Human CD20 antibody (Cat. No. 563779/563780; Right Panel). Erythrocytes were lysed with BD FACS™ Lysing Solution (Cat. No. 349202). Two color flow cytometric dot plots show the correlated expression of CD20 (or Ig Isotype Control staining) versus CD19 derived from 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 System.

**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™ BV650 under optimum conditions, and unconjugated antibody and free BD Horizon™ BV650 were removed.

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## Application Notes

### Application

Flow cytometry

Routinely Tested

### Suggested Companion Products

Catalog Number	Name	Size	Clone
554656	Stain Buffer (FBS)	500 ml	(none)
563437	BV650 Mouse IgG2b, $\kappa$ Isotype Control	50 $\mu$ g	27-35
563779	BV650 Mouse Anti-Human CD20	25 tests	2H7
349202	BD FACS™ Lysing Solution	100 ml	(none)
555899	Lysing Buffer	100 ml	(none)
555415	APC Mouse Anti-Human CD19	100 tests	HIB19
561742	APC Mouse Anti-Human CD19	25 tests	HIB19

### 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. 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. Species testing during development may have been performed with a different format of the same clone. Selected applications have been tested for cross-reactivity.
6. Alexa Fluor® is a registered trademark of Molecular Probes, Inc., Eugene, OR.
7. Brilliant Violet™ 650 is a trademark of Sirigen.
8. For fluorochrome spectra and suitable instrument settings, please refer to our Multicolor Flow Cytometry web page at [www.bdbiosciences.com/colors](http://www.bdbiosciences.com/colors).
9. Please refer to [www.bdbiosciences.com/pharming/en/protocols](http://www.bdbiosciences.com/pharming/en/protocols) for technical protocols.

### References

Clark EA, Yokochi T. Human B cell and B cell blast-associated surface molecules defined with monoclonal antibodies. In: Bernard A, Boumsell L, Dausset J, Milstein C, Schlossman SF, ed. *Leukocyte Typing*. Berlin: Springer-Verlag; 1984:339-346. (Clone-specific: Blocking, Flow cytometry, Immunoprecipitation)

Hultin LE, Hausner MA, Hultin PM, Giorgi JV. CD20 (pan-B cell) antigen is expressed at a low level on a subpopulation of human T lymphocytes. *Cytometry*. 1993; 14(2):193-204. (Biology)

Knapp W, Dörken B, Gilks WR, et al, ed. *Leukocyte Typing IV*. New York, NY: Oxford University Press; 1989:1-1182. (Biology)

Ledbetter JA, Clark EA. Surface phenotype and function of tonsillar germinal center and mantle zone B cell subsets. *Hum Immunol*. 1986; 15:30-43. (Immunogen: Blocking, Flow cytometry)

Loken MR, Shah VO, Dattilio KL, Civin CI. Flow cytometric analysis of human bone marrow. II. Normal B lymphocyte development. *Blood*. 1987; 70(5):1316-1324. (Biology)

Reinherz EL, Haynes BF, Nadler LM, Berstein ID, ed. *Leukocyte Typing II: Human B Lymphocytes*. New York: Springer-Verlag; 1986:1-560. (Clone-specific: Flow cytometry, Immunoprecipitation)

Schlossman SF, Boumsell L, Gilks W, et al, ed. *Leukocyte Typing V: White Cell Differentiation Antigens*. New York: Oxford University Press; 1995. (Biology)

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