

## Technical Data Sheet

**BV421 Mouse Anti-Human CD20****Product Information**

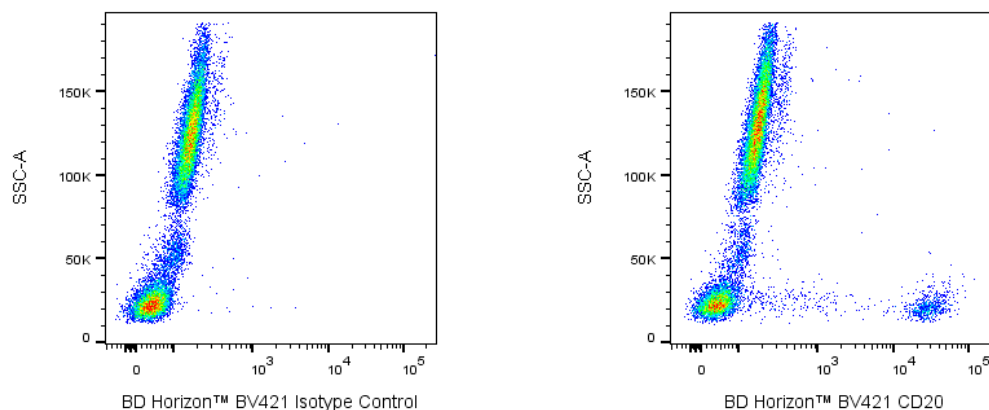
<b>Material Number:</b>	<b>562873</b>
<b>Alternate Name:</b>	MS4A1; B1; Bp35; LEU-16; S7
<b>Size:</b>	100 Tests
<b>Vol. per Test:</b>	5 µl
<b>Clone:</b>	2H7
<b>Immunogen:</b>	Human 6.16c1.3 B cell line
<b>Isotype:</b>	Mouse (C57BL/6) IgG2b, κ
<b>Reactivity:</b>	QC Testing: Human Tested in Development: Rhesus, Cynomolgus, Baboon
<b>RRID:</b>	AB_2737857
<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, 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 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.

This clone also cross-reacts with a subset of peripheral blood lymphocytes, but not monocytes nor granulocytes, of baboon and both rhesus and cynomolgus macaque monkeys. The distribution on lymphocytes is similar to that seen with normal human donor lymphocytes, namely bright staining on B lymphocytes and weak reactivity on a small subset of CD3-positive T lymphocytes.

The antibody was conjugated to BD Horizon™ BV421 which is part of the BD Horizon Brilliant™ Violet family of dyes. With an Ex Max near 407 nm and Em Max near 421 nm, BD Horizon™ BV421 can be excited by the violet laser (405 nm) and detected with a 450/50 nm filter. BD Horizon™ BV421 conjugates are very bright, often exhibiting a 10 fold improvement in brightness compared to Pacific Blue™ conjugates. Due to nearly identical excitation and emission properties but different spillover characteristics, BD Horizon™ BV421, Pacific Blue™, and BD Horizon™ V450 cannot be used simultaneously.



**Multiparameter flow cytometric analysis of CD20 expression on human peripheral blood leucocyte populations.** Whole blood was stained with either BD Horizon™ BV421 Mouse IgG2b, κ Isotype Control (Cat. No. 562748; Left Plot) or with BD Horizon™ BV421 Mouse Anti-Human CD20 antibody (Cat. No. 562873; Right Plot). Erythrocytes were lysed with BD Pharm Lyse™ Lysing Buffer (Cat. No. 555899). Bivariate pseudocolor density plots showing the correlated expression of CD20 (or Ig Isotype Control staining) versus side light-scatter (SSC-A) derived from gated events with the forward light-scatter characteristics of intact leucocytes. Flow cytometry and data analysis was performed using a BD LSRFortessa™ X-20 Cell Analyzer System and FlowJo™ software.

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562873 Rev. 3



## 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 to the dye under optimum conditions and unconjugated antibody and free dye were removed.

## Application Notes

### Application

Flow cytometry	Routinely Tested
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### Recommended Assay Procedure:

BD® CompBeads can be used as surrogates to assess fluorescence spillover (Compensation). When fluorochrome conjugated antibodies are bound to BD® CompBeads, they have spectral properties very similar to cells. However, for some fluorochromes there can be small differences in spectral emissions compared to cells, resulting in spillover values that differ when compared to biological controls. It is strongly recommended that when using a reagent for the first time, users compare the spillover on cells and BD® CompBeads to ensure that BD® CompBeads are appropriate for your specific cellular application.

For optimal and reproducible results, BD Horizon Brilliant™ Stain Buffer should be used anytime BD Horizon Brilliant™ dyes are used in a multicolor flow cytometry panel. Fluorescent dye interactions may cause staining artifacts which may affect data interpretation. The BD Horizon Brilliant™ Stain Buffer was designed to minimize these interactions. When BD Horizon Brilliant™ Stain Buffer is used in the multicolor panel, it should also be used in the corresponding compensation controls for all dyes to achieve the most accurate compensation. For the most accurate compensation, compensation controls created with either cells or beads should be exposed to BD Horizon Brilliant™ Stain Buffer for the same length of time as the corresponding multicolor panel. 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)
554657	Stain Buffer (BSA)	500 mL	(none)
563794	Brilliant Stain Buffer	100 Tests	(none)
566349	Brilliant Stain Buffer	1000 Tests	(none)
566385	Brilliant Stain Buffer Plus	1000 Tests	(none)
555899	Lysing Buffer	100 mL	(none)
349202	Lysing Solution 10X Concentrate	100 mL	(none)
562748	BV421 Mouse IgG2b, κ Isotype Control	50 µg	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-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. 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.
5. 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).
6. BD Horizon Brilliant Violet 421 is covered by one or more of the following US patents: 8,158,444; 8,362,193; 8,575,303; 8,354,239.
7. 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.
8. Species cross-reactivity detected in product development may not have been confirmed on every format and/or application.
9. Please refer to [www.bdbiosciences.com/us/s/resources](http://www.bdbiosciences.com/us/s/resources) for technical protocols.
10. Please refer to <http://regdocs.bd.com> to access safety data sheets (SDS).
11. Pacific Blue™ is a trademark of Life Technologies Corporation.

### References

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