

## Technical Data Sheet

## BB515 Mouse Anti-Human CD35

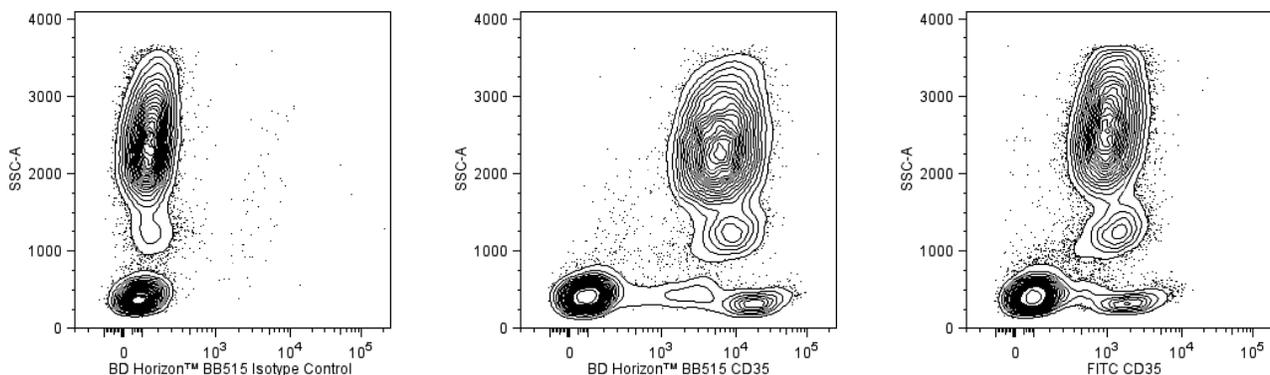
## Product Information

<b>Material Number:</b>	<b>566021</b>
<b>Alternate Name:</b>	CR1; Complement receptor type 1; C3b/C4b receptor; C3BR; C4BR; KN
<b>Size:</b>	25 Tests
<b>Vol. per Test:</b>	5 µl
<b>Clone:</b>	E11
<b>Immunogen:</b>	Human Cells of the Monocyte Lineage
<b>Isotype:</b>	Mouse IgG1, κ
<b>Reactivity:</b>	QC Testing: Human Reported Reactivity: Rhesus, Cynomolgus, Baboon
<b>Workshop:</b>	III 204
<b>Storage Buffer:</b>	Aqueous buffered solution containing ≤0.09% sodium azide.

## Description

The E11 monoclonal antibody specifically binds to CD35. CD35 is also known as Complement receptor type 1 (CR1), C3b/C4b receptor, C3BR, C4BR, Immune adherence receptor, or KN. CD35 is a type I transmembrane glycoprotein that exists in four allelic forms of 160, 190, 220 and 250 kDa. CD35 serves as a receptor for complement fragments C3b, iC3b, C3dg, C4b, iC3, and iC4. It enhances phagocytosis by neutrophils and monocytes and regulates complement activation. It is expressed on erythrocytes, granulocytes, monocytes, B cells, and some dendritic cells, T cells, and NK cells. It binds complement components C3b and C4b, mediating. This antibody cannot inhibit the phagocytic capacity of granulocytes. The CD35 antibody is useful in studies of cells that express complement receptors.

The antibody was conjugated to BD Horizon BB515 which is part of the BD Horizon Brilliant™ Blue family of dyes. With an Ex Max near 490 nm and an Em Max near 515 nm, BD Horizon BB515 can be excited by the blue laser (488 nm) laser and detected with a 530/30 nm filter. This dye has been exclusively developed by BD Biosciences and is up to seven times brighter than FITC with less spillover into the PE channel. Due to similar excitation and emission properties, BB515, FITC, and Alexa Fluor® 488 cannot be used simultaneously. It is not recommended to use BB515 in cocktails that include Streptavidin conjugates as it may cause high background.



**Multiparameter flow cytometric analysis of CD35 expression on human peripheral blood leucocytes - Staining comparisons between BD Horizon™ BB515 and FITC-conjugated antibodies.** Human whole blood was stained with either BD Horizon BB515 Mouse IgG1, κ Isotype Control (Cat. No. 564416; Left Plot), BD Horizon BB515 Mouse Anti-Human CD35 antibody (Cat. No. 565330/566021; Middle Plot), or FITC Mouse Anti-Human CD35 antibody (Cat. No. 555452; Right Plot). The erythrocytes were lysed with BD FACS™ Lysing Solution (Cat. No.349202). Two-parameter flow cytometric contour plots showing the correlated expression of CD35 (or Ig Isotype control staining) versus side light-scatter (SSC) signals were derived from gated events with the forward and side light-scatter characteristics of intact leucocyte populations. 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™ BB515 under optimum conditions and unconjugated antibody was removed.

## Application Notes

## Application

Flow cytometry	Routinely Tested
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United States 877.232.8995 Canada 866.979.9408 Europe 32.2.400.98.95 Japan 0120.8555.90 Asia Pacific 65.6861.0633 Latin America/Caribbean 55.11.5185.9995

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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 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 CompBead to ensure that BD Comp beads are appropriate for your specific cellular application.

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).

For optimal results, it is recommended to perform 2 washes after staining with antibodies. Cells may be prepared, stained with antibodies and washed twice with wash buffer per established protocols for immunofluorescence staining, prior to acquisition on a flow cytometer. Performing fewer than the recommended wash steps may lead to increased spread of the negative population.

### Suggested Companion Products

Catalog Number	Name	Size	Clone
564416	BB515 Mouse IgG1, $\kappa$ Isotype Control	100 $\mu$ g	X40
349202	BD FACSTM Lysing Solution	100 mL	(none)
555899	Lysing Buffer	100 mL	(none)
565330	BB515 Mouse Anti-Human CD35	50 Tests	E11
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)

### 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. The manufacture, use, sale, offer for sale, or import of this product is subject to one or more patents or pending applications. This product, and only in the amount purchased by buyer, may be used solely for buyer's own internal research, in a manner consistent with the accompanying product literature. No other right to use, sell or otherwise transfer (a) this product, or (b) its components is hereby granted expressly, by implication or by estoppel. Diagnostic uses require a separate license.
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 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.
7. Please refer to <http://regdocs.bd.com> to access safety data sheets (SDS).
8. Please refer to [www.bdbiosciences.com/us/s/resources](http://www.bdbiosciences.com/us/s/resources) for technical protocols.

### References

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- Lin G-X, Yang X, Hollemweguer E, et al. Cross-reactivity of CD antibodies in eight animal species. In: Mason D. David Mason .. et al., ed. *Leukocyte typing VII : white cell differentiation antigens : proceedings of the Seventh International Workshop and Conference held in Harrogate, United Kingdom*. Oxford: Oxford University Press; 2002:519-523. (Clone-specific: Flow cytometry)
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- Hogg N, Ross GD, Jones DB, Slusarenko M, Walport MJ, Lachmann PJ. Identification of an anti-monocyte monoclonal antibody that is specific for membrane complement receptor type one (CR1). *Eur J Immunol*. 1984; 14(3):236-243. (Immunogen: Blocking, Fluorescence microscopy, Functional assay, Immunofluorescence, Immunohistochemistry, Immunoprecipitation, Inhibition, Radioimmunoassay)
- Nickells M, Hauhart R, Krych M, et al. Mapping epitopes for 20 monoclonal antibodies to CR1. *Clin Exp Immunol*. 1998; 112(1):27-33. (Clone-specific: Dot Blot, ELISA)
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