Technical Data Sheet

BB515 Mouse Anti-Human CD27

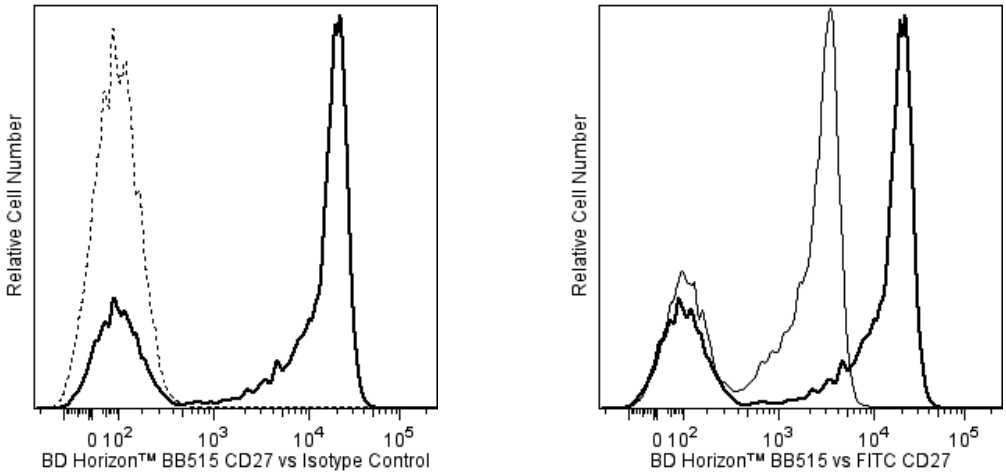
Product Information

Material Number:	**564642**
Alternate Name:	TNFRSF7; TNF receptor superfamily, member 7; T14; Tp55; S152
Size:	100 Tests
Vol. per Test:	5 µl
Clone:	M-T271
Immunogen:	Human T-CLL cells
Isotype:	Mouse (BALB/c) IgG1, κ
Reactivity:	QC Testing: Human Tested in Development: Rhesus, Cynomolgus, Baboon
Workshop:	IV T187; V 5T CD27.03
Storage Buffer:	Aqueous buffered solution containing ≤0.09% sodium azide.

Description

The M-T271 monoclonal antibody specifically binds to CD27. CD27 presents as a type I transmembrane, disulphide-linked 110 kDa homodimer comprised of two polypeptide chains. The CD27 molecule is a lymphocyte-specific member of the TNF/NGF-R family, and is expressed on a subset of human thymocytes and on the majority of mature T lymphocytes, activated B cells and NK cells. CD27 is highly induced on T cells after TCR stimulation. CD27 binds to CD70 (also known as, CD27 ligand or CD27L) and may be involved in cellular interaction of T and B lymphocytes.

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.



Flow cytometric analysis of CD27 expression on human peripheral blood lymphocytes - 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; dashed line histogram) or BD Horizon BB515 Mouse Anti-Human CD27 antibody (Cat. No. 564642/564643; bold solid line histogram). Alternatively, cells were stained with FITC Anti-Human CD27 antibody (Cat. No. 555440/557329/560986; thin solid line histogram). Erythrocytes were lysed with BD FACS Lysing Solution (Cat. No. 349202).*

Overlaid histograms are shown to facilitate staining comparisons between: BB515 Anti-CD27 antibody versus its Ig Isotype Control (Left Panel), and BB515 Anti-CD27 antibody versus FITC Anti-CD27 antibody (Right Panel). The fluorescence histograms showing CD27 expression (or Ig Isotype control staining) were 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.

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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

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
555440	FITC Mouse Anti-Human CD27	100 Tests	M-T271
557329	FITC Mouse Anti-Human CD27	50 Tests	M-T271
560986	FITC Mouse Anti-Human CD27	25 Tests	M-T271
349202	BD FACS™ Lysing Solution	100 mL	(none)
555899	Lysing Buffer	100 mL	(none)
563794	Brilliant Stain Buffer	100 Tests	(none)
554656	Stain Buffer (FBS)	500 mL	(none)
554657	Stain Buffer (BSA)	500 mL	(none)
564416	BB515 Mouse IgG1, κ Isotype Control	100 µg	X40
564643	BB515 Mouse Anti-Human CD27	25 Tests	M-T271
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 × 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. 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. For fluorochrome spectra and suitable instrument settings, please refer to our Multicolor Flow Cytometry web page at www.bdbiosciences.com/colors.
5. 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.
6. Species cross-reactivity detected in product development may not have been confirmed on every format and/or application.
7. Please refer to http://regdocs.bd.com to access safety data sheets (SDS).
8. Alexa Fluor® is a registered trademark of Life Technologies Corporation.
9. Please refer to www.bdbiosciences.com/us/s/resources for technical protocols.

References

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Morimoto C. Cluster report: CD27. In: Schlossman SF. Stuart F. Schlossman .. et al., ed. *Leucocyte typing V : white cell differentiation antigens : proceedings of the fifth international workshop and conference held in Boston, USA, 3-7 November, 1993.* Oxford: Oxford University Press; 1995:356-357. (Clone-specific: Flow cytometry)

Bigler RD, Donat TL, Boselli CM. Definition of three epitopes of the CD27 molecule [P 120->55] present on activated normal lymphocytes. In: Knapp W. W. Knapp .. et al., ed. *Leucocyte typing IV : white cell differentiation antigens.* Oxford New York: Oxford University Press; 1989:351-352. (Clone-specific: Blocking, Flow cytometry)

Kato K, Cantwell MJ, Sharma S, Kipps TJ. Gene transfer of CD40-ligand induces autologous immune recognition of chronic lymphocytic leukemia B cells. *J Clin Invest.* 1998; 101(5):1133-1141. (Clone-specific: ELISA, Flow cytometry)

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