Technical Data Sheet

BV605 Rat Anti-Mouse CD62L

Product Information

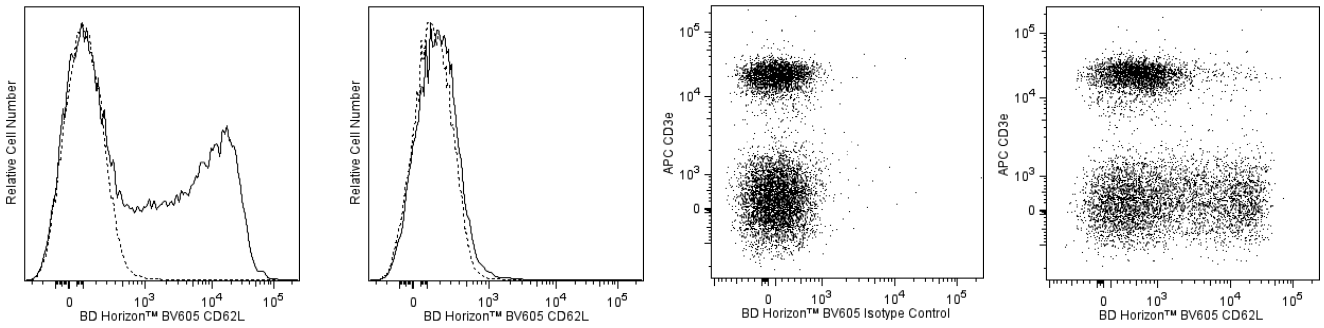
Material Number:	**563252**
Alternate Name:	Sell; L-selectin; LECAM-1; LAM-1; Lnhr; Ly-22; Ly-m22; Lyam-1
Size:	50 µg
Concentration:	0.2 mg/ml
Clone:	MEL-14
Immunogen:	C3H/eb mouse B lymphoma 38C-13
Isotype:	Rat (F344) IgG2a, κ
Reactivity:	QC Testing: Mouse
Storage Buffer:	Aqueous buffered solution containing BSA and ≤0.09% sodium azide.

Description

The MEL-14 monoclonal antibody specifically binds to CD62L (L-selectin), a 95 kDa (on neutrophils) or 74 kDa (on lymphocytes) receptor with lectin-like and Epidermal Growth Factor-like domains. In the mouse, L-selectin is detected on most thymocytes, with the highest levels of expression on an immunocompetent subset and a population of dividing progenitor cells, and on peripheral leukocytes, including subsets of B and T lymphocytes, neutrophils, monocytes, and eosinophils. This member of the selectin adhesion molecule family appears to be required for lymphocyte homing to peripheral lymph nodes and to contribute to neutrophil emigration at inflammatory sites. L-selectin is rapidly shed from lymphocytes and neutrophils upon cellular activation; metalloproteinases may mediate the release of CD62L ectodomains from the cell surface. The level of CD62L expression, along with other markers, distinguishes naive, effector, and memory T cells. L-selectin binds to sialytaed oligosaccharide determinants on high endothelial venules (HEV) in peripheral lymph nodes. In vitro studies have demonstrated that CD34, GlyCAM-1, and MAdCAM-1, all recognized by mAb MECA-79 (anti-mouse PNAd Carbohydrate Epitope, Cat. No. 553863), may be ligands for CD62L. MEL-14 mAb blocks in vitro binding of lymphocytes to peripheral lymph node HEV and inhibits in vivo lymphocyte extravasation into peripheral lymph nodes and late stages of leukocyte rolling.

This antibody is conjugated to BD Horizon BV605 which is part of the BD Horizon Brilliant™ Violet family of dyes. With an Ex Max of 407-nm and Em Max of 602-nm, BD Horizon BV605 can be excited by a violet laser and detected with a standard 610/20-nm filter set. BD Horizon BV605 is a tandem fluorochrome of BD Horizon BV421 and an acceptor dye with an Em max at 605-nm. Due to the excitation of the acceptor dye by the green (532 nm) and yellow-green (561 nm) lasers, there will be significant spillover into the PE and BD Horizon PE-CF594 detectors off the green or yellow-green lasers. BD Horizon BV605 conjugates are very bright, often exhibiting brightness equivalent to PE conjugates and can be used as a third color off of the violet laser.

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).



Left Panels - Flow cytometric analysis of CD62L on mouse bone marrow cells. *Bone marrow cells from a BALB/c mouse were left untreated (Left Panel) or were cultured (1 hour) with Phorbol 12-Myristate 13-Acetate (PMA; Middle Left Panel). The cells were then stained with either BD Horizon™ BV605 Rat Anti-Mouse CD62L antibody (Cat. No. 563252, solid line histogram) or with BD Horizon™ BV605 Rat IgG2a, κ Isotype Control (Cat. No. 563144, dashed line histogram). Fluorescence histograms were derived from gated events with the forward and side light-scatter characteristics of viable bone marrow cells. Flow cytometric analysis was performed using a BD™ LSR II Flow Cytometer System.*

Right Panels - Multicolor flow cytometric analysis of CD62L expression on mouse splenocytes. *Splenic leucocytes from a BALB/c mouse were stained with APC Hamster Anti-Mouse CD3e (Cat. No. 553066/561826) and with either BD Horizon™ BV605 Rat IgG2a, κ Isotype Control (Middle Right Panel) or BD Horizon™ BV605 Rat Anti-Mouse CD62L antibody (Right Panel). Two-color flow cytometric dot plots showing the expression of CD62L (or Ig Isotype control staining) versus CD3e were derived from gated events with the forward and side light-scatter characteristics of viable leucocytes. 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 with BD Horizon™ BV605 under optimum conditions, and unconjugated antibody and free BD Horizon™ BV605 were removed.

Application Notes

Application

Flow cytometry	Routinely Tested

Suggested Companion Products

Catalog Number	Name	Size	Clone
554656	Stain Buffer (FBS)	500 mL	(none)
555899	Lysing Buffer	100 mL	(none)
563144	BV605 Rat IgG2a, κ Isotype Control	50 µg	R35-95
553066	APC Hamster Anti-Mouse CD3e	0.1 mg	145-2C11
561826	APC Hamster Anti-Mouse CD3e	25 µg	145-2C11
563794	Brilliant Stain Buffer	5 mL	(none)

Product Notices

1. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
2. Source of all serum proteins is from USDA inspected abattoirs located in the United States.
3. An isotype control should be used at the same concentration as the antibody of interest.
4. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.
5. Please observe the following precautions: Absorption of visible light can significantly alter the energy transfer occurring in any tandem fluorochrome conjugate; therefore, we recommend that special precautions be taken (such as wrapping vials, tubes, or racks in aluminum foil) to prevent exposure of conjugated reagents, including cells stained with those reagents, to room illumination.
6. 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.
7. For fluorochrome spectra and suitable instrument settings, please refer to our Multicolor Flow Cytometry web page at www.bdbiosciences.com/colors.
8. Although every effort is made to minimize the lot-to-lot variation in the efficiency of the fluorochrome energy transfer, differences in the residual emission from BD Horizon™ BV421 may be observed. Therefore, we recommend that individual compensation controls be performed for every BD Horizon™ BV605 conjugate.
9. CF™ is a trademark of Biotium, Inc.

References

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