Technical Data Sheet

BV510 Mouse Anti-Human CD25

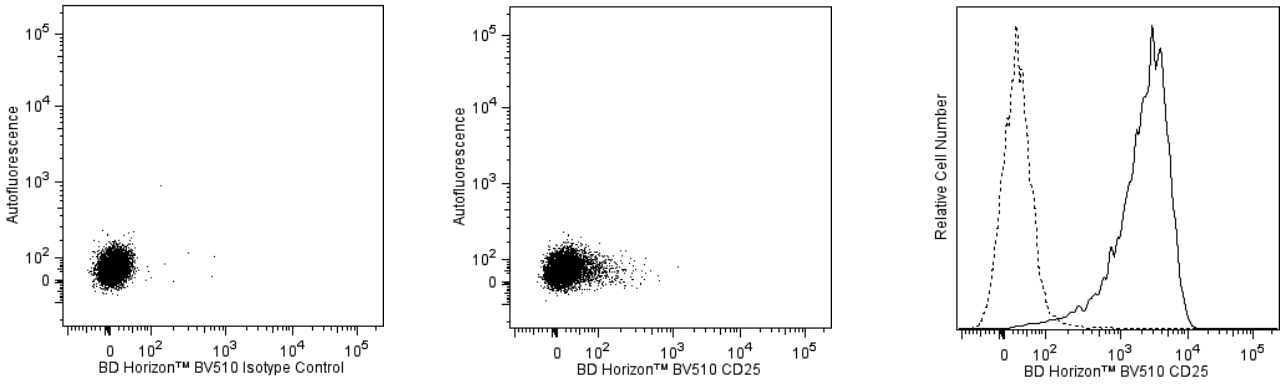
Product Information

Material Number:	**563352**
Alternate Name:	IL-2R; IL2RA; IL-2Rα; TCGFR; TAC antigen; p55
Size:	100 tests
Vol. per Test:	5 µl
Clone:	M-A251
Isotype:	Mouse IgG1, κ
Reactivity:	QC Testing: Human
Workshop:	IV A053
Storage Buffer:	Aqueous buffered solution containing BSA and ≤0.09% sodium azide.

Description

The M-A251 monoclonal antibody specifically binds to the 55 kDa type I transmembrane glycoprotein known as the low-affinity interleukin-2 receptor alpha chain subunit (IL-2Rα). CD25 is expressed on regulatory T cells and on activated lymphocytes (T and B) and monocytes. It associates with the IL-2Rβ/CD122 and the IL-2Rγ/CD132 receptor chains to form the high-affinity IL-2R complex. CD25 expression on T and B lymphocytes is upregulated by antigenic or mitogenic stimulation. Soluble CD25/IL-2Rα is produced as a consequence of lymphocyte stimulation and is found in biological fluids following inflammatory responses.

The antibody was conjugated to BD Horizon™ BV510 which is part of the BD Horizon™ Brilliant Violet™ family of dyes. With an Ex Max of 405-nm and Em Max at 510-nm, BD Horizon™ BV510 can be excited by the violet laser and detected in the BD Horizon™ V500 (525/50-nm) filter set. BD Horizon™ BV510 conjugates are useful for the detection of dim markers off the violet laser.



Flow cytometric analysis of CD25 expression on unstimulated and stimulated human peripheral blood lymphocytes.

Left and Middle Panels: Whole blood was stained with either BD Horizon™ BV510 Mouse IgG1, κ Isotype Control (Cat. No. 562946; Left Panel) or BD Horizon™ BV510 Mouse Anti-Human CD25 antibody (Cat. No. 563352/563351; Middle Panel). The erythrocytes were subsequently lysed with BD Pharm Lyse™ Lysing Buffer (Cat. No. 555899). The two-color flow cytometric dot plot shows the correlated expression of CD25 (or Ig Isotype control staining) versus cellular autofluorescence derived from gated events with the forward light-scatter characteristics of viable lymphocytes.

Right Panel: Phytohemagglutinin-stimulated (3 days) human peripheral blood mononuclear cells were stained with either BD Horizon™ BV510 Mouse Anti-Human CD25 antibody (solid line histogram) or with BD Horizon™ BV510 Mouse IgG1, κ Isotype Control (dashed line histogram). The fluorescence histograms were derived from events with the forward and side light-scatter characteristics of viable lymphoblasts.

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™ BV510 under optimum conditions, and unconjugated antibody and free BD Horizon™ BV510 were removed.

Application Notes

Application

Flow cytometry	Routinely Tested

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Recommended Assay Procedure:

Use of Anti-CD25 antibodies conjugated with brighter fluorochromes, eg, phycoerythrin, allophycocyanin, PE-Cy™7, or BD Horizon™ BV421, are recommended for staining cells that express relatively low levels of CD25 such as unstimulated regulatory T cells or memory T cells.

Suggested Companion Products

Catalog Number	Name	Size	Clone
554656	Stain Buffer (FBS)	500 ml	(none)
563351	BV510 Mouse Anti-Human CD25	25 tests	M-A251
562946	BV510 Mouse IgG1, k Isotype Control	50 µg	X40
555899	Lysing Buffer	100 ml	(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. 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. 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.
6. For fluorochrome spectra and suitable instrument settings, please refer to our Multicolor Flow Cytometry web page at www.bdbiosciences.com/colors.
7. Brilliant Violet™ 510 is a trademark of Sirigen.

References

Barclay NA, Brown MH, Birkeland ML, et al, ed. *The Leukocyte Antigen FactsBook.* San Diego, CA: Academic Press; 1997. (Biology)

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

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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