

Reagents Provided

Peridinin-Chlorophyll-Protein-Complex (PerCP)-conjugated goat monoclonal anti-human BTLA: Supplied as 50 µg of antibody in 1 mL saline containing up to 0.5% BSA and 0.1% sodium azide.

Isotype: goat IgG

Reagents Not Provided

- Flow Cytometry Staining Buffer (Catalog # FC001) or other BSA-supplemented saline buffer.

Storage

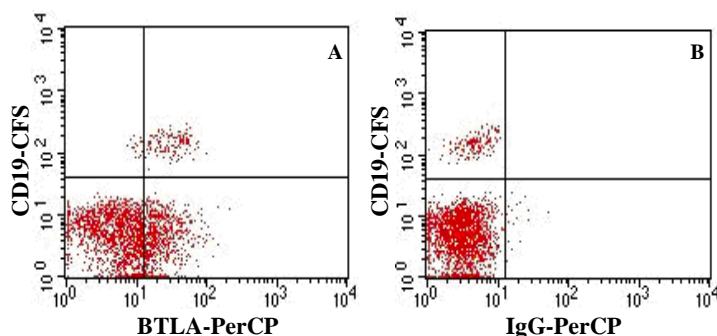
Reagents are stable for **twelve months** from the date of receipt when stored in the dark at 2° - 8° C.

Intended Use

Designed to quantitatively determine the percentage of cells bearing BTLA within a population and qualitatively determine the density of BTLA on cell surfaces by flow cytometry.

Product Description

This antibody was produced in goats immunized with purified, NS0-derived, recombinant human B and T Lymphocyte Attenuator (rhBTLA; aa 25 - 150; Accession # Q7Z6A9 with V105M, S138G and M148V substitutions). Human BTLA specific IgG was purified by human BTLA affinity chromatography. The purified antibody was then conjugated to PerCP fluorochrome. Cell surface expression of BTLA is determined by flow cytometry. PerCP has a maximum absorption of 482 nm and 564 nm and a maximum emission of 675 nm.



Whole blood lymphocytes were stained with either A) PerCP-conjugated anti-human BTLA (Catalog # FAB3354C) or B) PerCP-conjugated isotype control (Catalog # IC108C) and CFS-conjugated anti-human CD19 (Catalog # FAB4867F).

Background Information

BTLA, also known as CD272, is a 70 kDa type I membrane glycoprotein that belongs to the CD28 family of receptors. CD28 receptors are Ig superfamily proteins with an extracellular IgV-like domain that function as co-stimulatory or co-inhibitory T cell signaling molecules. BTLA is expressed on B cells, T cells, and dendritic cells. Binding of BTLA to its ligand, herpesvirus entry mediator (HVEM, TNFRSF14) triggers a potent inhibitory signal that blocks T cell activation. The amino acid sequence of the extracellular domain of BTLA shares only 34% identity with that of mouse BTLA. A human BTLA variant lacking the transmembrane domain has been reported.

Flow Cytometry Validation

This antibody has been tested for flow cytometry using whole blood lymphocytes.

- Cells may be Fc-blocked with 1 µg of human IgG/10⁵ cells for 15 minutes at room temperature. Do not wash excess blocking IgG from this reaction.
- After blocking, 10 µL of conjugated antibody was added to up to 1 x 10⁶ cells and incubated for 30 minutes at room temperature.
- Unbound antibody was removed by washing the cells twice in Flow Cytometry Staining Buffer (Catalog # FC001). Note that whole blood requires a RBC lysis step at this point using Flow Cytometry Human Lyse Buffer (Catalog # FC002).
- The cells were resuspended in Flow Cytometry Staining Buffer for final flow cytometric analysis. As a control for this analysis, cells in a separate tube should be treated with PerCP-labeled goat IgG antibody. This procedure may need to be modified, depending upon the cell type and final utilization. Individual users may need to titrate to determine the optimal reagent amount for their specific use.

Warning: Contains sodium azide as a preservative - sodium azide may react with lead and copper plumbing to form explosive metal azides. Flush with large volumes of water during disposal.