

Reagents Provided

Peridinin-Chlorophyll-Protein-Complex (PerCP)-conjugated mouse monoclonal anti-human B7-H2: Supplied as 25 µg of antibody in 1 mL saline containing up to 0.5% BSA and 0.1% sodium azide.

Clone #: 136726

Isotype: mouse IgG_{2b}

Reagents Not Provided

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

Storage

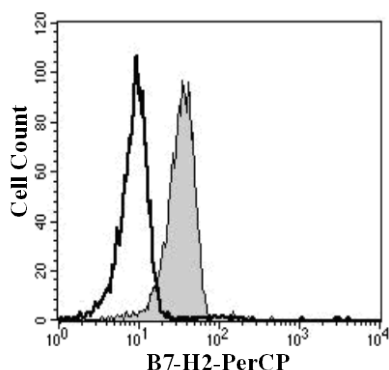
Reagents are stable for **twelve months** from the date of receipt when stored in the dark at 2° - 8° C.

Intended Use

Designed to quantitatively determine the percentage of cells bearing B7-H2 within a population and qualitatively determine the density of B7-H2 on cell surfaces by flow cytometry.

Product Description

This antibody was produced from a hybridoma resulting from the fusion of a mouse myeloma with B cells obtained from a mouse immunized with purified, NS0-derived, recombinant human B7-H2 (rhB7-H2; aa 19 - 258; Accession # O75144) extracellular domain. The IgG fraction of the tissue culture supernatant was purified by Protein G affinity chromatography. The purified antibody was then conjugated to PerCP fluorochrome. Cell surface expression of B7-H2 is determined by flow cytometry. PerCP has a maximum absorption of 482 nm and 564 nm and a maximum emission of 675 nm.



U937 cells were stained with PerCP-conjugated anti-human B7-H2 (Catalog # FAB165C, filled histogram) or PerCP-conjugated isotype control (Catalog # IC0041C, open histogram).

Background Information

Human B7-H2, also called B7RP-1 and ICOSL, is a 60 kDa member of the B7 family of immune costimulatory proteins. It is expressed on resting B cells, dendritic cells, monocytes, alveolar epithelial cells, and placental trophoblasts. B7-H2 interactions with ICOS promote lymphocyte differentiation and activation. Within the ECD, human B7-H2 shares 49% and 54% amino acid sequence identity with mouse and rat B7-H2, respectively.

Flow Cytometry Validation

This antibody has been tested for flow cytometry using U937 cells.

- 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 1 - 2.5 x 10⁵ 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 mouse IgG_{2b} 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.