

Reagents Provided

Alexa Fluor® 488-conjugated rat monoclonal anti-human Claudin-1:
Supplied as 25 µg of antibody in 0.5 mL saline containing up to 0.5% BSA and 0.1% sodium azide.

Clone #: 421203

Isotype: rat IgG_{2A}

Reagents Not Provided

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

Storage

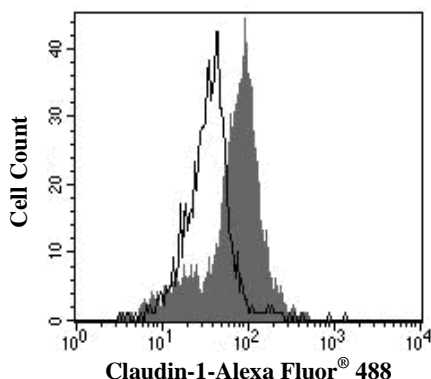
Reagents are stable for **twelve months** from the date of receipt when stored in the dark at 2-8 °C.

Intended Use

Designed to quantitatively determine the percentage of cells bearing Claudin-1 within a population and qualitatively determine the density of Claudin-1 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 rat immunized with CHO cells transfected with human Claudin-1 (Accession # O95832). The IgG fraction of the tissue culture supernatant was purified by Protein G affinity chromatography. The purified antibody was then conjugated to Alexa Fluor® 488 fluorochrome. Cell surface expression of Claudin-1 is determined by flow cytometry using 488 nm wavelength excitation and monitoring emitted fluorescence with a detector optimized to collect peak emissions at 515-545 nm.



PC-3 cells were stained with Alexa Fluor® 488-conjugated anti-human Claudin-1 (Catalog # FAB4618G; filled histogram) or Alexa Fluor® 488-conjugated isotype control (Catalog # IC006G; open histogram).

Background Information

Claudin-1 is a 23 kDa multipass membrane protein in the Claudin family of epithelial tight junction proteins. It is expressed by epithelial cells in a wide variety of tissues, as well as by Langerhans cells and dendritic cells. It is up- or down-regulated in many cancers and is required for entry of hepatitis C virus into hepatocytes. Human Claudin-1 shares 91% aa sequence identity with mouse and rat Claudin-1.

Flow Cytometry Validation

This antibody has been tested for flow cytometry using PC-3 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, 5 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 Alexa Fluor® 488-labeled rat IgG_{2A} 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.

Legal

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