Polyclonal
Anti-human Galectin-3-Alexa Fluor® 488

Catalog Number: IC1154G
Lot Number: ABWO01
100 Tests

Reagents Provided

Alexa Fluor® 488-conjugated goat polyclonal anti-human Galectin-3: Supplied as 10 µg of antibody in 0.5 mL saline containing up to 0.5% BSA and 0.1% sodium azide.

Isotype: goat IgG

Reagents Not Provided

Flow Cytometry Fixation Buffer (Catalog # FC004) or other 4% paraformaldehyde fixation buffer.

Flow Cytometry Permeabilization/Wash Buffer I (1X) (Catalog # FC005) or other saponin-containing saline buffer.

Storage

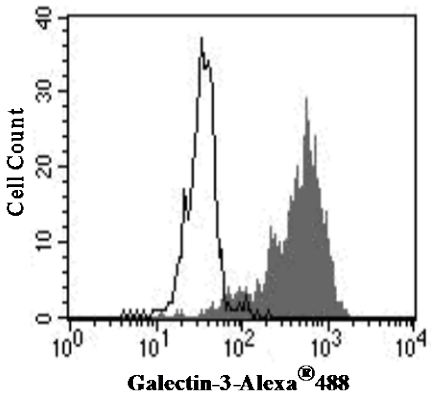
Reagents are stable for **twelve months** from the date of receipt when stored in the dark at 2-8 °C.

Intended Use

Designed to quantitatively determine the percentage of cells containing Galectin-3 within a population and qualitatively determine the density of intracellular Galectin-3 by flow cytometry.

Product Description

Produced in goats immunized with purified *E. coli*-derived recombinant human Galectin-3 (rhGalectin-3). Human Galectin-3 specific IgG was purified by human Galectin-3 affinity chromatography. The purified antibody was then conjugated to Alexa Fluor® 488 fluorochrome. Intracellular expression of Galectin-3 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.



PBMC monocytes were stained with Alexa Fluor® 488-conjugated anti-human Galectin-3 (Catalog # IC1154G; filled histogram) or Alexa Fluor® 488-conjugated isotype control (Catalog # IC108G; open histogram).

Background Information

Galectin-3, also known as Mac-2, L29, CBP35 and εBP, is a member of a large family of carbohydrate-binding proteins with specificity for N-acetyl-lactosamine-containing glycoproteins. At least 14 mammalian galectins, which share structural similarities in their carbohydrate recognition domains (CRD), have been identified to date. Galectin-3 is expressed in tumor cells, macrophages, activated T cells, osteoclasts, epithelial cells, and fibroblasts.

Flow Cytometry Validation

For intracellular staining, cells must first be fixed and permeabilized. We recommend the use of 4% PFA as a fixative and a 0.1% saponin balanced salt solution for permeabilization and washing (see Reagents Not Provided).

1. Cells were harvested and washed twice in saline buffer.
2. Cell surface staining may be done at this point following the manufacturer's staining procedure.
3. Up to 1 x 10^6 cells were resuspended in 0.5 mL of cold Flow Cytometry Fixation Buffer (Catalog # FC004) and incubated at room temperature for 10 minutes.
4. Following fixation, the cells were washed twice in saline buffer then once in Flow Cytometry Permeabilization/Wash Buffer I (Catalog # FC005).
5. After permeabilization, 5 µL of conjugated antibody was added and the cells were incubated for 30 minutes at room temperature **in the dark**.
6. The cells were washed twice with Flow Cytometry Permeabilization/Wash Buffer I.
7. The cells were resuspended in saline 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 goat IgG antibody. This procedure may need to be modified, depending on 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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