

Monoclonal Anti-mouse Semaphorin 4F-APC

Catalog Number: FAB7200A

Lot Number: ADLC01

100 Tests

Reagents Provided

Allophycocyanin (APC)-conjugated rat monoclonal anti-mouse Semaphorin 4F: Supplied as 10 µg of antibody in 1 mL saline containing up to 0.5% BSA and 0.09% sodium azide.

Clone #: 780225

Isotype: rat IgG_{2A}

Reagents Not Provided

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

Storage

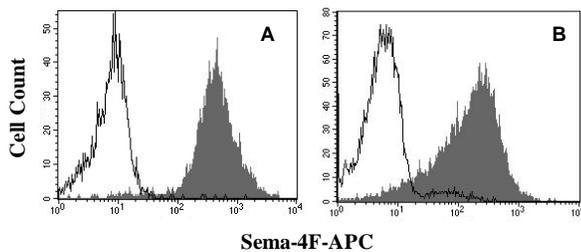
Reagents are stable for **twelve months** from the date of receipt when stored in the dark at 2-8 °C.

Intended Use

Designed to quantitatively determine the percentage of cells bearing Semaphorin 4F within a population and qualitatively determine the density of Semaphorin 4F 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 purified NS0-derived recombinant mouse Semaphorin 4F (aa 41-667). The IgG fraction of the tissue culture supernatant was purified by Protein A or G affinity chromatography. The purified antibody was then conjugated to APC fluorochrome. Cell surface expression of Semaphorin 4F is determined by flow cytometry using 620-650 nm wavelength excitation and monitoring emitted fluorescence with a detector optimized to collect peak emissions at 660-670 nm.



A) Mouse cortical stem cells (MCSC), or B) bEnd.3 cells, were stained with APC-conjugated anti-mouse Semaphorin 4F (Catalog # FAB7200A; filled histograms) or APC-conjugated isotype control (Catalog # IC006A; open histograms).

Background Information

Semaphorin 4F (Sema4F; also known as Sema W) is a 90-100 kDa, class IV member of the semaphorin family of proteins. It is expressed by select cell types, including Schwann cells and neurons, and likely acts as an inhibitor of cell proliferation by Schwann cells, and a regulator of postsynaptic glutamatergic synapses on neuron dendrites. Mature mouse Sema4F is a type I transmembrane protein that is 737 amino acids (aa) in length. It contains a 627 aa extracellular region (aa 41-667) that is characterized by the presence of one Sema domain (aa 71-493), a PSI region (aa 518-569), and a C2-type Ig-like domain (aa 586-641). Over aa 41-667, mouse Sema4F shares 92% and 98% aa identity with human and rat Sema4F, respectively.

Flow Cytometry Validation

This antibody has been tested for flow cytometry using mouse cortical stem cells (MCSC) and bEnd.3 cells.

- Cells may be Fc-blocked with 1 µg of mouse 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 Mouse Lyse Buffer (Catalog # FC003).
- 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 APC-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.