

Reagents Provided

Allophycocyanin (APC)-conjugated mouse monoclonal anti-human FOLR3: Supplied as 10 µg of antibody in 1 mL saline containing up to 0.5% BSA and 0.1% sodium azide.

Clone #: 553720

Isotype: mouse IgG₁

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

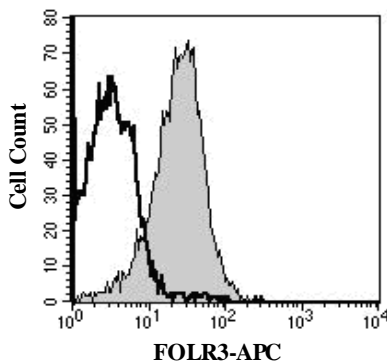
Reagents are stable for **twelve months** from the date of receipt when stored in the dark at 2° - 8° C.

Intended Use

Designed to quantitatively determine the percentage of cells containing FOLR3 within a population and qualitatively determine the density of intracellular FOLR3 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 FOLR3 (rhFOLR3; aa 1 - 245; Accession # P41439). The IgG fraction of the tissue culture supernatant was purified by Protein G affinity chromatography. The purified antibody was then conjugated to APC fluorochrome. Intracellular expression of FOLR3 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.



K562 (H₂O, non-responsive) cells were stained with APC-conjugated anti-human FOLR3 (Catalog # IC53191A, filled histogram) or isotype control (Catalog # IC002A, open histogram).

2009/12/25

Background Information

FOLR3 (folate receptor 3; also FR-γ and FOLBP-3) is a 32 kDa, secreted member of the folate receptor family. Although folate family receptors can internalize folate, they are generally not the principal conduits for folate uptake. FOLR3 will bind folate in milk and blood, possibly stabilizing this molecule. Its levels are low, however, and it is best known as a potential marker for malignancy. Human FOLR3 contains a 23 amino acid (aa) signal sequence and a 220 aa mature region. There are no definitive structural motifs. There is one isoform that shows a premature truncation after Leu104. This isoform does not bind folate. A second isoform shows a 53 aa substitution for aa 1 - 105. No definitive mouse counterpart to human FOLR3 has been reported.

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. 5×10^5 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, 10 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 APC-labeled mouse IgG₁ 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.