Monoclonal Anti-human/mouse SSEA-3-Alexa Fluor® 488

Catalog Number: FAB1434G
Lot Number: ADFJ01
100 Tests

Reagents Provided

Alexa Fluor® 488-conjugated rat monoclonal anti-human/mouse SSEA-3: Supplied as 10 µg of antibody in 0.5 mL saline containing up to 0.5% BSA and 0.09% sodium azide.

Clone #: MC-631

Isotype: rat IgM

Reagents Not Provided

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

Storage

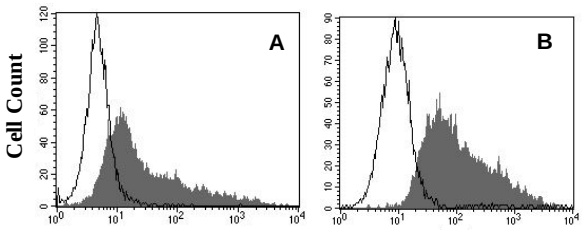
Reagents are stable for **twelve months** from the date of receipt when stored in the dark at 2-8 °C.

Intended Use

Designed to quantitatively determine the percentage of cells bearing SSEA-3 within a population and qualitatively determine the density of SSEA-3 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 four to eight cell stage mouse embryos. The IgM fraction of the tissue culture supernatant was purified by IgM-specific affinity chromatography. The purified antibody was then conjugated to Alexa Fluor® 488 fluorochrome. Cell surface expression of SSEA-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.



SSEA-3-Alexa Fluor® 488

A) BG01V human embryonic stem cells, or B) NTera-2 human pluripotent embryonic carcinoma cells, were stained with Alexa Fluor® 488-conjugated anti-human/mouse SSEA-3 (Catalog # FAB1434G; filled histograms), or Alexa Fluor® 488-conjugated isotype control (open histograms).

Background Information

SSEA-3, also known as glycolipid GB5, is expressed on the surface of human teratocarcinoma stem cells (EC), human embryonic germ cells (EG), and human embryonic stem cells (ES).[1] Expression of SSEA-3 is down-regulated following differentiation of human EC cells. In contrast, the differentiation of murine EC and ES cells may be accompanied by an increase in SSEA-3 expression.[2, 3]

References

1. Zhou, D. *et al*. (2000) J. Biol. Chem. **275**:22631.
2. Thomson, J.A. & J.S. Odorico (2000) Trends Biotechnol. **18**:53.
3. Draper, J.S. *et al*. (2002) J. Anat. **200**:249.

Flow Cytometry Validation

This antibody has been tested for flow cytometry using BG01V or NTera-2 cells.

1. Cells may be Fc-blocked with 1 µg of human IgG/10^5 cells for 15 minutes at room temperature. Do not wash excess blocking IgG from this reaction.
2. After blocking, 5 µL of conjugated antibody was added to up to 1 x 10^6 cells and incubated for 30 minutes at room temperature.
3. 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).
4. 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 IgM 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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