

# Monoclonal Anti-human SOX17-Alexa Fluor® 488

Catalog Number: IC19241G

Lot Number: ACEO01

100 Tests

## Reagents Provided

### Alexa Fluor® 488-conjugated mouse monoclonal anti-human SOX17:

Supplied as 50 µg of antibody in 0.5 mL saline containing up to 0.5% BSA and 0.1% sodium azide.

Clone #: 614013

Isotype: mouse IgG<sub>1</sub>

## 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 bearing SOX17 within a population and qualitatively determine the density of SOX17 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 mouse immunized with purified, *E. coli*-derived, recombinant human SOX17 (Asp177 - Val414; Accession # Q9H612). The IgG fraction of the tissue culture supernatant was purified by Protein A or G affinity chromatography. The purified antibody was then conjugated to Alexa Fluor® 488 fluorochrome. Intracellular expression of SOX17 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.

## Background Information

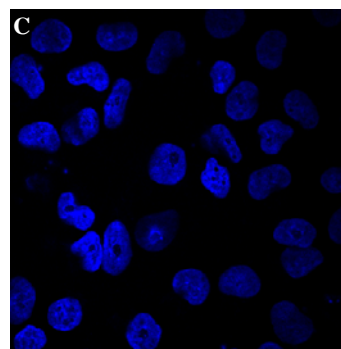
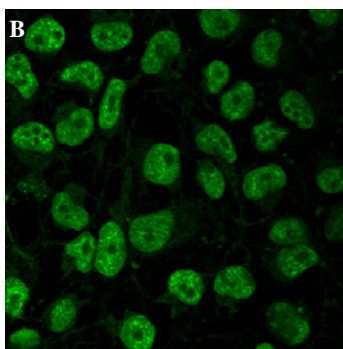
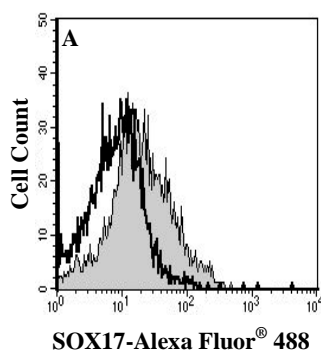
SOX17 is a member of the SOX family of transcription factors that bind DNA via a high mobility group (HMG) domain. SOX17 is suggested to play an important role in endoderm development.

## 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<sup>6</sup> 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 mouse IgG<sub>1</sub> 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.



(A) Definitive endoderm-differentiated BG01V human embryonic stem cells were stained with Alexa Fluor® 488-conjugated anti-human SOX17 (Catalog # IC19241G, filled histogram) or Alexa Fluor® 488-conjugated isotype control (Catalog # IC002G, open histogram). The same cells were also visualized by immunocytochemistry following staining with Alexa Fluor® 488-conjugated anti-human SOX17 (Catalog # IC19241; B) and counterstaining with DAPI (C).

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