

Reagents Provided

Allophycocyanin (APC)-conjugated goat polyclonal anti-human KLF4: Supplied as 10 µg of antibody in 1 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

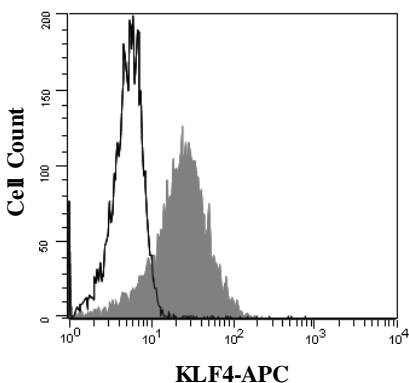
Reagents are stable for **twelve months** from the date of receipt when stored in the dark at 2° - 8° C.

Intended Use

Designed to quantitatively determine the percentage of cells containing KLF4 within a population and qualitatively determine the density of intracellular KLF4 by flow cytometry.

Product Description

This antibody was produced in goats immunized with purified, E. coli-derived, recombinant human Kruppel-like Factor 4 (rhKLF4; aa 2 - 470; Accession # O43474). Human KLF4 specific IgG was purified by human KLF4 affinity chromatography. The purified antibody was then conjugated to an APC fluorochrome. Intracellular expression of KLF4 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.



BGO1V cells were stained with APC-conjugated anti-human KLF4 (Catalog # IC3640A, filled histogram) or isotype control (Catalog # IC108A, open histogram).

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FOR RESEARCH USE ONLY. NOT FOR USE IN HUMANS.

Background Information

Human KLF4, also known as epithelial zinc finger protein (EZFP), is a 53 kDa (470 aa) member of the kruppel C2H2-type zinc finger protein family. It contains three C2H2-type zinc fingers at the carboxyl terminus that preferentially bind to cis-DNA elements that are G-C rich. KLF4 regulates the expression of target genes that are involved in different cellular functions. KLF4 is highly expressed in the epithelial cells of the skin and the gastrointestinal tract. Human and mouse KLF4 share 90% amino acid sequence identity.

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 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.

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