

# Monoclonal Anti-human OX40 Ligand/TNFSF4-APC

Catalog Number: FAB10541A Lot Number: ABLJ02

100 Tests

## **Reagents Provided**

Allophycocyanin (APC)-conjugated mouse monoclonal anti-human OX40 Ligand/TNFSF4: Supplied as 10  $\mu$ g of antibody in 1 mL saline containing up to 0.5% BSA and 0.1% sodium azide.

Clone #: 159403 Isotype: mouse IgG<sub>1</sub>

## **Reagents Not Provided**

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

## Storage

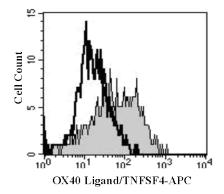
Reagents are stable for **twelve months** from the date of receipt when stored in the dark at 2° - 8° C.

#### **Intended Use**

Designed to quantitatively determine the percentage of cells bearing OX40 Ligand/TNFSF4 within a population and qualitatively determine the density of OX40 Ligand/TNFSF4 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, NS0-derived, recombinant human OX40 Ligand (rhOX40 Ligand) extracellular domain. The IgG fraction of the tissue culture supernatant was purified by Protein G affinity chromatography. The purified antibody was then conjugated to APC fluorochrome. Cell surface expression of OX40 Ligand/TNFSF4 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.



PBMCs stimulated with recombinant human GM-CSF (Catalog # 215-GM) and LPS were stained with APC-conjugated anti-human OX40 Ligand/TNFSF4 (Catalog # FAB10541A, filled histogram) or isotype control (Catalog # IC002A, open histogram).

## **Background Information**

OX40 Ligand, also known as gp34, is a type II transmembrane glycoprotein belonging to the TNF superfamily and is now designated TNFSF4. Engagement of OX40 Ligand by OX40 (CD134) on activated CD4<sup>+</sup> T cells promotes the proliferation and differentiation of activated B cells.

## Flow Cytometry Validation

This antibody has been tested for flow cytometry using stimulated peripheral blood mononuclear cells (PBMCs).

- 1. Cells may be Fc-blocked with 1  $\mu$ g of human IgG/10<sup>5</sup> cells for 15 minutes at room temperature. Do not wash excess blocking IgG from this reaction.
- 2. After blocking, 10  $\mu$ L of conjugated antibody was added to up to 1 x 10 $^6$  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 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 APC-labeled mouse IgG<sub>1</sub> 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.