

# Monoclonal Anti-human CRTH-2-Fluorescein

Catalog Number: FAB3338F

Lot Number: ABGH01

100 Tests

## Reagents Provided

**Carboxyfluorescein (CFS)-conjugated mouse monoclonal anti-human CRTH-2:** Supplied as 50 µg of antibody in 1 mL saline containing up to 0.5% BSA and 0.1% sodium azide.

**Clone #:** 301108

**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 CRTH-2 within a population and qualitatively determine the density of CRTH-2 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 NS0 cell transfectants expressing human Chemoattractant receptor-homologous molecule on Th2 cells (rhCRTH-2; Accession # BAA74518). The IgG fraction of the tissue culture supernatant was purified by Protein G affinity chromatography. The purified antibody was then conjugated to CFS fluorochrome. Cell surface expression of CRTH-2 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

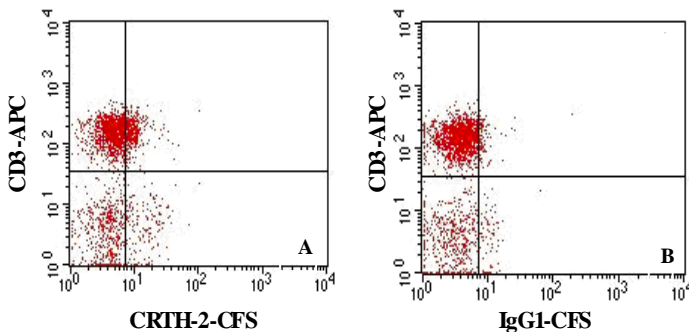
CRTH-2, also known as GPR44, is a seven transmembrane G protein-coupled receptor that is expressed by activated Th2 cells and eosinophils. CRTH-2 binds Prostaglandin D2 and induces chemotaxis of these cells in allergic and inflammatory immune responses. Human and mouse CRTH-2 share 78% amino acid sequence identity.

## Flow Cytometry Validation

This antibody has been tested for flow cytometry using human lymphocytes.

- Cells may be Fc-blocked with 1 µg of human IgG/10<sup>5</sup> cells for 15 minutes at room temperature. Do not wash excess blocking IgG from this reaction.
- After blocking, 10 µL of conjugated antibody was added to 1 - 2.5 x 10<sup>5</sup> 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).
- 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 CFS-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.



Human lymphocytes were stained with A) CFS-conjugated anti-human CRTH-2 (Catalog # FAB3338F) or B) isotype control (Catalog # IC002F) and APC-conjugated anti-human CD3 (Catalog # FAB100A).

2009/12/14

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