

## Reagents Provided

**Peridinin-Chlorophyll-Protein-Complex (PerCP)-conjugated mouse monoclonal anti-human CCR6:** Supplied as 25 µg of antibody in 1 mL saline containing up to 0.5% BSA and 0.1% sodium azide.

Clone #: 53103

Isotype: mouse IgG<sub>2B</sub>

## Reagents Not Provided

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

## Storage

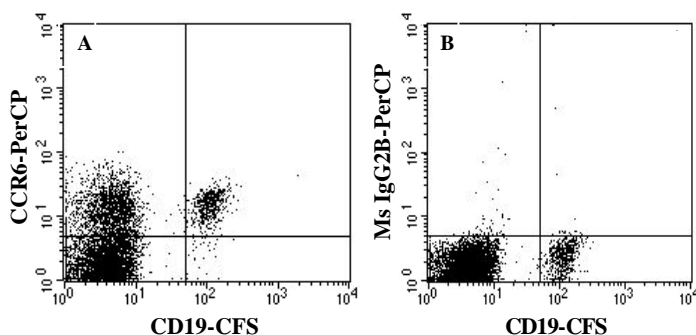
Reagents are stable for **twelve months** from the date of receipt when stored in the dark at 2° - 8° C.

## Intended Use

Designed to quantitatively determine the percentage of cells bearing CCR6 within a population and qualitatively determine the density of CCR6 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 transfected NS0 cells human CCR6 (rhCCR6; Accession # P51684). The IgG fraction of the ascites fluid was purified by Protein G affinity chromatography. The purified antibody was then conjugated to PerCP fluorochrome. Cell surface expression of CCR6 is determined by flow cytometry. PerCP has a maximum absorption of 482 nm and 564 nm and a maximum emission of 675 nm.



Whole blood lymphocytes were stained with CFS-conjugated anti-human CD19 (Catalog # FAB4867F). These cells were then double-stained with PerCP-conjugated anti-human CCR6 (Catalog# FAB195C, dot plot A) or isotype control (Catalog # IC0041C, dot plot B).

## Background Information

CCR6 is a seven transmembrane-spanning G protein-coupled receptor that binds the chemokine MIP-3α and mediates the chemoattraction of B and T lymphocytes.

## Flow Cytometry Validation

This antibody has been tested for flow cytometry using B cells in whole blood.

- 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 PerCP-labeled mouse IgG<sub>2B</sub> antibody. This procedure may need to be modified, depending upon cell type and final utilization. Individual users may need to titrate to determine 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.