

# Monoclonal Anti-human Histamine H1 R-Phycoerythrin

Catalog Number: FAB4726P Lot Number: ABMJ01 100 Tests

## **Reagents Provided**

#### Phycoerythrin (PE)-conjugated mouse monoclonal anti-human

Histamine H1 R: Supplied as 25  $\mu g$  of antibody in 1 mL saline containing up to 0.5% BSA and 0.1% sodium azide.

Clone #: 480054

Isotype: mouse IgG,

#### **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  $2^{\circ}$  -  $8^{\circ}$  C.

#### Intended Use

Designed to quantitatively determine the percentage of cells bearing Histamine H1 R within a population and qualitatively determine the density of Histamine H1 R 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 cells transfected with human HRH1 (Accession # P35367). The IgG fraction of the tissue culture supernatant was purified by Protein G affinity chromatography. The purified antibody was then conjugated to PE fluorochrome. Cell surface expression of Histamine H1 R is determined by flow cytometry using 488 nm wavelength excitation and monitoring emitted fluorescence with a detector optimized to collect peak emissions at 565 - 605 nm.



A) U937 cells or B) U937 cells activated with PMA were stained with PE-conjugated anti-human Histamine H1 R (Catalog # FAB4726P, filled histograms) or PE-conjugated mouse isotype control (Catalog # IC002P, open histograms).

# **Background Information**

HRH1 (Histamine H1 R) is a 60 kDa, 487 amino acid (aa) G protein-coupled 7-transmembrane putative glycoprotein. It is implicated in the pathogenesis of asthma and is a major pharmaceutical target for antihistamines. It has been detected as a mixture of monomers and homodimers, with the highest concentration in the placenta, followed by brain, lung (produced by airway smooth muscle, bronchial epithelium and macrophages), and other tissues. Extracellular domains of human HRH1 share 72% aa sequence identity with corresponding regions of mouse or rat HRH1.

## **Flow Cytometry Validation**

This antibody has been tested for flow cytometry using U937 cells.

- 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.
- 2. After blocking, 10  $\mu$ L of conjugated antibody was added to up to 1 x 10<sup>6</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).
- 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 PE-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.