Human IL-12/IL-35 p35 Antibody

Recombinant Monoclonal Rabbit IgG Clone # 2038C
Catalog Number: MAB15701

DESCRIPTION

Species Reactivity	Human
Specificity	Detects human IL-12/IL-35 p35 in direct ELISAs. Detects the IL-12/IL-35 p35 subunit either as part of the IL-12 p40/p35 heterodimer or as the IL-12/IL-35 p35 monomer. This antibody does not detect the IL-12/IL-23 p40 subunit.
Source	Recombinant Monoclonal Rabbit IgG Clone # 2038C
Purification	Protein A or G purified from cell culture supernatant
Immunogen	*S. frugiperda* insect ovarian cell line *Sf* 21-derived recombinant human IL-12 p40 and human IL-12 p35 Ile23-Ser328 of p40, Arg23-Ser219 of p35 Accession # P29460 (p40) & P29459 (P35)
Formulation	Lyophilized from a 0.2 μm filtered solution in PBS with Trehalose. See Certificate of Analysis for details. *Small pack size (-SP) is supplied either lyophilized or as a 0.2 μm filtered solution in PBS.

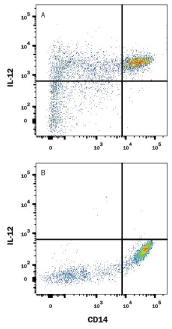
APPLICATIONS

Please Note: *Optimal dilutions should be determined by each laboratory for each application. General Protocols are available in the Technical Information section on our website.*

	Recommended Concentration	Sample
Immunocytochemistry	8-25 µg/mL	See Below
Intracellular Staining by Flow Cytometry	0.25 µg/10^6 cells	See Below

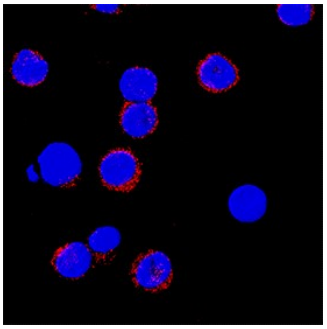
DATA

Intracellular Staining by Flow Cytometry



Detection of IL-12/IL-35 p35 in Human PBMCs treated with rhIFN- γ and LPS by Flow Cytometry. Human peripheral blood mononuclear cells (PBMCs), A) treated with recombinant human IFN- γ (Catalog # 285-IF, 75 ng/mL for 2 hours), then LPS (1 µg/mL for 12 hours) and lastly monensin (3 µM for 3 hours), or B) untreated, were stained with Mouse Anti-Human CD14 PE-conjugated Monoclonal Antibody (Catalog # FAB3832P) and Rabbit Anti-Human IL-12/IL-35 p35 Monoclonal Antibody (Catalog # MAB15701) followed by APC-conjugated Goat Anti-Rabbit Secondary Antibody (Catalog # F0111). Quadrant markers were set based on isotype control antibody (Catalog # MAB1050, data not shown). To facilitate intracellular staining, cells were fixed and permeabilized with FlowX FoxP3 Fixation & Permeabilization Buffer Kit (Catalog # FC012). View our protocol for Staining Intracellular Molecules.

Immunocytochemistry



IL-12/IL-35 p35 in Human PBMCs. IL-12/IL-35 p35 was detected in immersion fixed human peripheral blood mononuclear cells (PBMCs) using Rabbit Anti-Human IL-12/IL-35 p35 Monoclonal Antibody (Catalog # MAB15701) at 8 µg/mL for 3 hours at room temperature. Cells were stained using the NorthernLights™ 557-conjugated Anti-Rabbit IgG Secondary Antibody (red; Catalog # NL004) and counterstained with DAPI (blue). Specific staining was localized to cytoplasm. View our protocol for Fluorescent ICC Staining of Non-adherent Cells.

PREPARATION AND STORAGE

Reconstitution	Reconstitute at 0.5 mg/mL in sterile PBS.
Shipping	The product is shipped at ambient temperature. Upon receipt, store it immediately at the temperature recommended below. *Small pack size (-SP) is shipped with polar packs. Upon receipt, store it immediately at -20 to -70 °C
Stability & Storage	**Use a manual defrost freezer and avoid repeated freeze-thaw cycles.** ● 12 months from date of receipt, -20 to -70 °C as supplied. ● 1 month, 2 to 8 °C under sterile conditions after reconstitution. ● 6 months, -20 to -70 °C under sterile conditions after reconstitution.

BACKGROUND

Interleukin 12, also known as natural killer cell stimulatory factor (NKSF) or cytotoxic lymphocyte maturation factor (CLMF), is a pleiotropic cytokine originally identified in the medium of activated human B lymphoblastoid cell lines. The p40 subunit of IL-12 has been shown to have extensive amino acid sequence homology to the extracellular domain of the human IL-6 receptor while the p35 subunit shows distant but significant sequence similarity to IL-6, G-CSF, and chicken MGF. These observations have led to the suggestion that IL-12 might have evolved from a cytokine/soluble receptor complex. Human and murine IL-12 share 70% and 60% amino acid sequence homology in their p40 and p35 subunits, respectively. IL-12 apparently shows species specificity with human IL-12 reportedly showing minimal activity in the murine system.

IL-12 is produced by macrophages and B lymphocytes and has been shown to have multiple effects on T cells and natural killer (NK) cells. These effects include inducing production of IFN-γ and TNF by resting and activated T and NK cells, synergizing with other IFN-γ inducers at both the transcriptional and post-transcriptional levels. This interaction induces IFN-γ gene expression, enhancing the cytotoxic activity of resting NK and T cells, inducing and synergizing with IL-2 in the generation of lymphokine-activated killer (LAK) cells, acting as a co-mitogen to stimulate proliferation of resting T cells, and inducing proliferation of activated T and NK cells. Current evidence indicates that IL-12, produced by macrophages in response to infectious agents, is a central mediator of the cell-mediated immune response by its actions on the development, proliferation, and activities of TH1 cells. In its role as the initiator of cell-mediated immunity, it has been suggested that IL-12 has therapeutic potential as a stimulator of cell-mediated immune responses to microbial pathogens, metastatic cancers, and viral infections such as AIDS.

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