

DESCRIPTION

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|---------------------------|---|
| Species Reactivity | Human |
| Specificity | Detects human CXCL9/MIG in ELISAs and Western blots. In ELISAs, does not cross-react with recombinant mouse (rm) CXCL9, recombinant human CXCL10. |
| Source | Monoclonal Mouse IgG ₁ Clone # 49106 |
| Purification | Protein A or G purified from hybridoma culture supernatant |
| Immunogen | <i>E. coli</i> -derived recombinant human CXCL9/MIG Thr23-Thr125 Accession # Q07325 |
| Endotoxin Level | <0.10 EU per 1 µg of the antibody by the LAL method. |
| 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. |

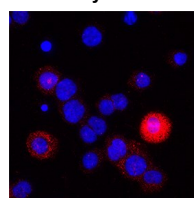
APPLICATIONS

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.

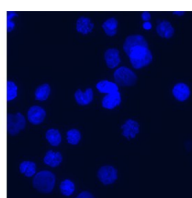
| | Recommended Concentration | Sample |
|---|--|---|
| Western Blot | 1 µg/mL | Recombinant Human CXCL9/MIG (Catalog # 392-MG) under non-reducing conditions only |
| Immunocytochemistry | 8-25 µg/mL | See Below |
| Intracellular Staining by Flow Cytometry | 2.5 µg/10 ⁶ cells | THP-1 cells treated with Recombinant Human IFN-γ (Catalog # 285-IF), fixed with paraformaldehyde, and permeabilized with saponin |
| Human CXCL9/MIG Sandwich Immunoassay | | Reagent |
| ELISA Capture | 2-8 µg/mL | Human CXCL9/MIG Antibody (Catalog # MAB392) |
| ELISA Detection Standard | 0.1-0.4 µg/mL | Human CXCL9/MIG Biotinylated Antibody (Catalog # BAF392) Recombinant Human CXCL9/MIG (Catalog # 392-MG) |
| CyTOF-ready | Ready to be labeled using established conjugation methods. No BSA or other carrier proteins that could interfere with conjugation. | |
| Neutralization | Measured by its ability to neutralize CXCL9/MIG-induced chemotaxis in the BaF3 mouse pro-B cell line transfected with mouse CXCR3. The Neutralization Dose (ND ₅₀) is typically 0.5-4 µg/mL in the presence of 0.25 µg/mL Recombinant Human CXCL9/MIG. | |

DATA

Immunocytochemistry



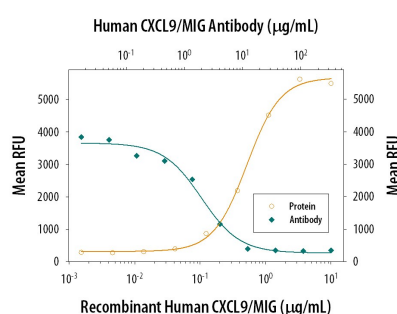
THP-1 cells + IFN gama



THP-1 cells

CXCL9/MIG in THP-1 Human Cell Line. CXCL9/MIG was detected in immersion fixed THP-1 human acute monocytic leukemia cell line treated with IFN gamma (positive staining) and THP-1 human acute monocytic leukemia cell line (untreated; negative staining) using Mouse Anti-Human CXCL9/MIG Monoclonal Antibody (Catalog # [MAB392](#)) at 25 µg/mL for 3 hours at room temperature. Cells were stained using the NorthernLights™ 557-conjugated Anti-Mouse IgG Secondary Antibody (red; Catalog # [NL007](#)) and counterstained with DAPI (blue). Specific staining was localized to cytoplasm. Staining was performed using our protocol for Fluorescent ICC Staining of Non-adherent Cells.

Neutralization



Chemotaxis Induced by CXCL9/MIG and Neutralization by Human CXCL9/MIG Antibody. Recombinant Human CXCL9/MIG (Catalog # [392-MG](#)) chemoattracts the BaF3 mouse pro-B cell line transfected with mouse CXCR3 in a dose-dependent manner (orange line). The amount of cells that migrated through to the lower chemotaxis chamber was measured by Resazurin (Catalog # [AR002](#)). Chemotaxis elicited by Recombinant Human CXCL9/MIG (0.25 µg/mL) is neutralized (green line) by increasing concentrations of Mouse Anti-Human CXCL9/MIG Monoclonal Antibody (Catalog # [MAB392](#)). The ND₅₀ is typically 0.5-4 µg/mL.

PREPARATION AND STORAGE

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|--------------------------------|---|
| 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 | <p>Use a manual defrost freezer and avoid repeated freeze-thaw cycles.</p> <ul style="list-style-type: none"> • 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

CXCL9, a member of the α subfamily of chemokines that lack the ELR domain, was initially identified as a lymphokine-activated gene in mouse macrophages. Human CXCL9 was subsequently cloned using mouse MIG cDNA as a probe. The CXCL9 gene is induced in macrophages and in primary glial cells of the central nervous system specifically in response to IFN- γ . CXCL9 has been shown to be a chemoattractant for activated T-lymphocytes and TIL but not for neutrophils or monocytes. The human CXCL9 cDNA encodes a 125 amino acid (aa) residue precursor protein with a 22 aa residue signal peptide that is cleaved to yield a 103 aa residue mature protein. CXCL9 has an extended carboxy-terminus containing greater than 50% basic aa residues and is larger than most other chemokines. The carboxy-terminal residues of CXCL9 are prone to proteolytic cleavage resulting in size heterogeneity of natural and recombinant CXCL9. CXCL9 with large carboxy-terminal deletions have been shown to have diminished activity in the calcium flux assay. A chemokine receptor (CXCR3) specific for CXCL9 and IP-10 has been cloned and shown to be highly expressed in IL-2-activated T-lymphocytes. The *E. coli*-expressed CXCL9 preparations produced at R&D Systems have been shown to contain greater than 80% full length CXCL9.

References:

1. Loetscher, M. *et al.* (1996) J. Exp. Med. **184**:963.
2. Liao, F. *et al.* (1995) J. Exp. Med. **182**:1301.
3. Vanguri, P. (1995) J. Neuroimmunol. **56**:35.