3050 Spruce Street, St. Louis, MO 63103 USA
Tel: (800) 521-8956 (314) 771-5765 Fax: (800) 325-5052 (314) 771-5757
email: techservice@sial.com sigma-aldrich.com

Product Information

ANTI-MACROPHAGE-DERIVED CHEMOKINE (MDC), HUMAN
Developed in Chicken,
Affinity Isolated Antibody

Product Number **M9307**

Product Description

Anti-Human Macrophage-derived Chemokine (MDC) is developed in chicken using purified recombinant human MDC, expressed in *E. coli*, as immunogen. The specific IgY antibody is purified using human MDC affinity chromatography.

Anti-Human MDC may be used to neutralize the bioactivity of recombinant human MDC. The antibody may also be used for immunoblotting and ELISA. By Immunoblotting and ELISA the antibody shows approximately 20% cross-reactivity with recombinant human MPIF-1, 10% cross-reactivity with recombinant human MIP-1δ and 1% cross-reactivity with recombinant mouse MDC, recombinant human HCC-1, recombinant human I-309, recombinant mouse MIP-1α, recombinant viral MIP-1, recombinant mouse MIP-1β and recombinant mouse MIP-1γ.

MDC is a member of the β (CC) chemokine family and induces chemotaxis or calcium mobilization in dendritic cells, IL-2 activated cells and activated T lymphocytes. MDC, also named stimulated T cell chemotactic protein (STCP-1), was isolated from clones of monocyte-derived macrophages and binds to CC chemokine receptor 4 (CCR4). T. MDC shows less than 35% identity to other C-C chemokine family members. It is expressed in dendritic cells, macrophages and activated monocytes. Expression is also detected in the tissues of thymus, lymph node and appendix. NH_2-terminally truncated forms of natural MDC were isolated from a CD8+ T cell clone and have been reported to inhibit HIV-1 infection.

Reagents

Anti-Human MDC is supplied lyophilized from a 0.2 μm filtered solution of phosphate buffered saline with 5% trehalose. Endotoxin level is <10 ng per mg antibody to <0.10EU/ug as determined by the LAL method.

Preparation Instructions

To one vial of lyophilized powder, add 1 ml of 0.2 μm-filtered PBS with 5% trehalose to produce a 0.1 mg/ml stock solution of antibody. If aseptic technique is used, no further filtration should be needed for use in cell culture environments.

Precautions and Disclaimer

This product is for R&D use only, not for drug, household, or other uses.

Storage/Stability

Prior to reconstitution, store at −20°C. Reconstituted product may be stored at 2-8°C for at least one month. For prolonged storage, freeze in working aliquots at −20°C. Avoid repeated freezing and thawing

Procedure

Neutralization of Bioactivity
To measure the ability of the antibody to neutralize the bioactivity of human MDC, recombinant human MDC was incubated with various concentrations of the antibody for 30 minutes in a microwell plate. Following preincubation, 75 μl of cytokine-antibody solution (containing antibody at concentrations of 0.01-100 μg/ml and recombinant human MDC at 0.05 μg/ml) was transferred to the lower compartment of a 96-well chemotaxis chamber. The chamber was assembled using a PVP-free polycarbonate filter and 2 x 10^6 cells/well were added to the top chamber. After the plate was incubated at 37°C for 3 hours in a humidified CO_2 incubator, the cells that migrated through to the lower chamber were stained with Alamar blue and the relative fluorescence was measured
The ND_{50} is the concentration of antibody required to yield one-half maximal inhibition of the cytokine activity on a responsive cell line, when the cytokine is present at a concentration just high enough to elicit a maximum response.

Product Profile

For neutralization, a working concentration of 0.5-1.5 :g/ml of Anti-Human MDC will neutralize 50% of the

bioactivity due to 50 ng/ml recombinant human MDC using CEM-NK^R.

For indirect immunoblotting, a working concentration of 0.1-0.2 μg/ml is determined using human MDC at 5 ng/lane and 20 ng/ml under non-reducing and reducing conditions, respectively.

Note: In order to obtain best results in different techniques and preparations we recommend determining optimal working dilutions by titration test.

References

1. Howell, D.N., et al., Natural killing target antigens as inducers of interferon: studies with an immunoselected, natural killing-resistant human T lymphoblastoid cell line. J. Immunol., **134**, 971-976 (1985).
2. Godiska, R., et al., Human macrophage-derived chemokine (MDC), a novel chemoattractant for monocytes, monocyte-derived dendritic cells, and natural killer cells. J. Exp. Med., **185**, 1595-1604 (1997).
3. Chang, M., et al., Molecular cloning and functional characterization of a novel CC chemokine, stimulated T cell chemotactic protein (STCP-1) that specifically acts on activated T lymphocytes. J. Biol. Chem., **272**, 25229-25237 (1997).
4. Pal, R., et al., Inhibition of HIV-1 infection by the β-chemokine MDC. Science, **278**, 695-698 (1997).
5. Pal, R., et al., Comments, Science, **281**, 487 (1998).
6. Imai, T., et al., Macrophage-derived chemokine is a functional ligand for the CC chemokine receptor 4. J. Biol. Chem., **273**, 1764-1768 (1998).

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