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Product Information

Monoclonal Anti-CXCR-4 (Fusin), Clone 44716
produced in mouse, purified immunoglobulin

Catalog Number **C6598**

Product Description

Monoclonal Anti-CXCR-4 (Fusin) (mouse IgG2b isotype) is derived from the 44716 hybridoma, which is produced by the fusion of mouse myeloma cells with B cells from a Balb/c mouse immunized with human CXCR-4 transfected mouse 3T3 cells. The antibody is purified from the tissue culture supernatant using protein G chromatography.

Monoclonal Anti-CXCR-4 reacts with CXCR-4 (fusin) transfected human cells and not with the parent cell line. The antibody also reacts with cells expressing feline CXCR-4, but not rat CXCR-4. The antibody shows no cross-reactivity with other chemokine receptors. The antibody will neutralize CXCL12/SDF-1α-induced response using human CXCR-4 transfected BaF3 cells in a chemotaxis assay.

Monoclonal Anti-CXCR-4 may be used detect CXCR-4 present on human cells by flow cytometry. The antibody may be used to neutralize human cell surface CXCR-4 mediated bioactivity. The antibody may also be used for immunohistology.

Chemokines have been sub-divided into families on the basis of the relative position of their cysteine residues. The α- and β- families, with four cysteine residues, are the largest and best characterized. In the α-family, one amino acid separates the first two cysteine residues (CXC); in the β-family the two cysteine residues (CC) are adjacent to each other. The α-chemokines that contain the N-terminal Glu-Leu-Arg amino acid sequence (ELR-motif) are chemotactic for neutrophils, such as IL-8, while those that do not, act on lymphocytes, such as IP-10 and MIG. Examples of chemokines under the β-family category are MCP1-5 and RANTES. The chemokine lymphotactin belongs to the γ-family, with only two cysteines (C), and the recently described fractalkine or neurotactin is a member of the δ-family and has the first two cysteine residues separated by three amino-acids (CXXXC).

Chemokines bind to specific G protein-coupled cell surface receptors on target cells. Five CXC receptors (CXCR1-5), nine CC receptors (CCR1-9) and one CXXXC receptor (CX_3CR1) have been cloned to date. Expression of chemokine receptors can be restricted to some cell types, e.g., CXCR1 is expressed in neutrophils, while others, such as CCR2, are expressed in a wide variety of cells.[1] Receptor expression has also been found to be constitutive (including down regulation), inducible or restricted to a cell state of activation. In addition, some chemokine receptors are also expressed in non-hematopoietic cells, such as nerve, endothelial and epithelial cells. This suggests that chemokines have other roles besides leucocyte chemotaxis. CX_3CR1, for example, is highly expressed in adult brain.

Chemokine receptors are linked to phospholipases through the Gi class of G proteins (inhibition by pertussis toxin). Receptor activation leads to a cascade of cellular events including generation of inositol triphosphate, calcium release and activation of protein kinase C. Chemokine receptors also activate small GTP-binding proteins of the Ras and Rho families, the latter being involved in cell motility events. In addition, chemokines bind to non-signaling molecules such as the Duffy antigen receptor for chemokines (DARC) which may act to remove chemokines from circulation, and heparan sulfates proteoglycans which may serve to establish an ECM concentration gradient.

CXCR-4, also known as fusin or LESTR,[2-3] was originally discovered as an orphan receptor with structural similarity to chemokine receptors. CXCR-4 was subsequently identified as a necessary cofactor for entry of T cell-tropic HIV viruses into $CD4^+$ cells.[2] The CXC chemokine PBSF/SDF-1 has now been shown to be the ligand for CXCR-4 and a powerful inhibitor of infection by T cell-tropic HIV-1 strains.[4-5]

Reagent

Lyophilized from 0.2 μm-filtered solution in phosphate buffered saline containing carbohydrates.

Precautions and Disclaimer
This product is for R&D use only, not for drug, household, or other uses. Please consult the Safety Data Sheet for information regarding hazards and safe handling practices.

Preparation Instructions
To one vial of lyophilized powder, add 1 mL of 0.2 μm-filtered PBS to produce a 0.5 mg/mL stock solution of antibody. If aseptic technique is used, no further filtration should be needed for use in cell culture environments.

Storage/Stability
For continuous use, store reconstituted material at 2–8 °C for up to one month. For extended storage, freeze in working aliquots. Repeated freezing and thawing, or storage in "frost-free" freezers, is not recommended. If slight turbidity occurs upon prolonged storage, clarify the solution by centrifugation before use. Working dilution samples should be discarded if not used within 12 hours.

Product Profile
Neutralization: a working antibody concentration of 2.5–12 μg/mL will block 50% of the bioactivity due to 1 ng/mL recombinant human CXCL12/SDF-1α in an assay measuring chemotaxis using mouse BaF3 cells transfected with human CXCR-4.

The ND_{50} of the antibody is defined as the concentration of antibody resulting in a one-half maximal inhibition of CXCL12/SDF-1α mediated chemotaxis on a responsive cell line.

Flow Cytometry: a working antibody concentration of 2.5 μg/10^6 cells is recommended.

Immunohistochemistry: a working antibody concentration of 8–25 μg/mL is recommended using cultured cells or tissue sections.

Note: In order to obtain the best results using various techniques and preparations, we recommend determining optimal working dilutions by titration.

Endotoxin level is <0.1 EU per μg antibody as determined by the LAL method.

References

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4. Bleul, C. et al., *Nature*, **382**, 829 (1996).
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