

## Product Information

### MONOCLONAL ANTI-HUMAN CXCR-3 CLONE 49801.111 Purified Mouse Immunoglobulin

Product Number **C6473**

#### Product Description

Monoclonal Anti-Human CXCR-3 (mouse IgG1 isotype) is derived from the 49801.111 hybridoma produced by the fusion of mouse myeloma cells and splenocytes from a Balb/c mouse immunized with human CXCR-3 transfected NSO mouse myeloma cells. The antibody is purified from ascites fluid using protein G chromatography.

Monoclonal Anti-Human CXCR-3 reacts with CXCR-3 transfected cells and not with the parent cell line by flow cytometry. The antibody shows no cross-reactivity with human CXCR-1, CXCR-2 and CXCR-4.

Monoclonal Anti-Human CXCR-3 may be used detect CXCR-3 by flow cytometry.

Chemokines have been sub-divided into families on the basis of the relative position of their cysteine residues. The  $\alpha$ - and  $\beta$ - families, with four cysteine residues, are the largest and best characterized. In the  $\alpha$ -family, one amino acid separates the first two cysteine residues (CXC); in the  $\beta$ -family the two cysteine residues (CC) are adjacent to each other. The  $\alpha$ -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  $\beta$ -family category are MCP1-5 and RANTES. The chemokine lymphotactin belongs to the  $\gamma$ -family, with only two cysteines (C), and the recently described fractalkine or neurotactin is a member of the  $*$ -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<sub>3</sub>CR1) have been cloned to date. Expression of chemokine receptors can be restricted to some cell types (CXCR1 is expressed in neutrophils)

while others (such as CCR2) are expressed in a wide variety of cells.<sup>1</sup> 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<sub>3</sub>CR1, 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 the circulation, and heparan sulfates proteoglycans which may serve to establish an ECM concentration gradient.

CXCR-3, also known as the IP-10/Mig receptor, shares approximately 40% protein sequence identity with CXCR-1 and CXCR-2, and 34.2 - 36.9% amino acid sequence identity with the five known CC chemokine receptors 1-5.<sup>2</sup> CXCR-3 is highly expressed by IL-2-activated T lymphocytes (Th1), but not by resting T lymphocytes, B lymphocytes, monocytes or granulocytes. CXCR-3 binds IP-10 and Mig (but not PF4) with high affinity and mediates Ca<sup>2+</sup> mobilization and chemotaxis. CXCR-3 does not bind any of the CXC chemokines containing the ELR motif.

#### Reagents

The product is supplied lyophilized from a 0.2  $\mu$ m filtered solution in phosphate buffered saline. Endotoxin level is < 10 ng per mg antibody as determined by the LAL method.

**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**

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.

**Product Profile**

For flow cytometry, a working concentration of 5-10 µg/ml is determined using 10 µl (10<sup>5</sup>) cells.

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

**References**

1. Wells, N.C., et al., Trends Pharm. Sci., **19**, 376 (1998).
2. Loetscher, M. et al. (1996) J. Exp. Med. 184:963.

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