Recombinant Mouse CCL2/JE/MCP-1

Catalog Number: 479-JE

DESCRIPTION

Source	*E. coli*-derived Gln24-Arg96 Accession # Q5SVU3
N-terminal Sequence Analysis	Gln24
Predicted Molecular Mass	8.5 kDa

SPECIFICATIONS

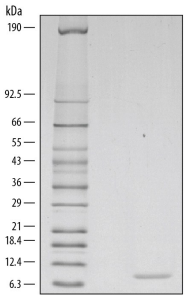
Activity	Measured by its ability to chemoattract BaF3 mouse pro-B cells transfected with human CCR2A. The ED_{50} for this effect is typically 2-10 ng/mL. Measured by its ability to chemoattract 2-day cultured human monocytes. Matsushima, K. *et al.* (1989) J. Exp. Med. **169**:1485. The ED_{50} for this effect is typically 5-20 ng/mL.
Endotoxin Level	<0.01 EU per 1 µg of the protein by the LAL method.
Purity	>97%, by SDS-PAGE visualized with Silver Staining and quantitative densitometry by Coomassie® Blue Staining.
Formulation	Lyophilized from a 0.2 µm filtered solution in PBS with BSA as a carrier protein. See Certificate of Analysis for details.

PREPARATION AND STORAGE

Reconstitution	Reconstitute at 100 µg/mL in sterile PBS containing at least 0.1% human or bovine serum albumin.
Shipping	The product is shipped at ambient temperature. Upon receipt, store it immediately at the temperature recommended below.
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. ● 3 months, -20 to -70 °C under sterile conditions after reconstitution.

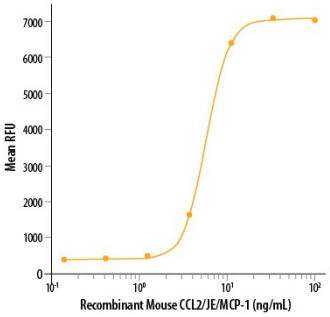
DATA

SDS-PAGE



1 µg/lane of Recombinant Mouse CCL2/JE/MCP-1 was resolved with SDS-PAGE under reducing (R) conditions and visualized by silver staining, showing a band at 8 kDa.

Bioactivity



Recombinant Mouse CCL2/JE/MCP-1 (Catalog # 479-JE) chemoattracts the BaF3 mouse pro-B cell line transfected with human CCR2A. The ED_{50} for this effect is typically 2-10 ng/mL.

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BACKGROUND

CCL2, also called monocyte chemotactic protein-1 (MCP-1) or JE, is a member of the C-C or β chemokine family that is best known as a chemotactic agent for mononuclear cells (1, 2). Mouse CCL2 cDNA encodes a 148 amino acid (aa) precursor protein with a 23 aa signal peptide and a 125 aa mature protein (1). Removal of the first 5 aa of the mature protein, including the N-terminal pyrrolidone carboxylic acid-modified glutamine, occurs naturally by metalloproteinase cleavage and downregulates activity but not receptor binding (3). Mouse CCL2 forms a broad band around 25 kDa on SDS-PAGE due to non-covalent dimerization and variable carbohydrate content (1). Mouse and rat express a form of CCL2 that is extended by 49 aa compared to other species. Mature mouse CCL2 shares 82% amino acid (aa) identity with rat CCL2 over the entire sequence, and 58%, 56%, 55%, 53% and 53% aa identity with human, equine, porcine, bovine and canine CCL2, respectively, over aa 24 - 101. Human CCL2 can, however, induce a limited response in rodent cells, and mouse CCL2 has full activity on human cells (2, 4). Fibroblasts, glioma cells, smooth muscle cells, endothelial cells, lymphocytes and mononuclear phagocytes can produce CCL2 either constitutively or upon mitogenic stimulation, but monocytes and macrophages appear to be the major source (1, 2). In addition to its chemotactic activity, CCL2 induces enzyme and cytokine release by monocytes, NK cells and lymphocytes, and histamine release by basophils that express its receptor, CCR2 (2). Additionally, it promotes Th2 polarization in $CD4^+$ T cells (5). CCL2-mediated recruitment of monocytes to sites of inflammation is proposed to play a role in the pathology of atherosclerosis, multiple sclerosis and allergic asthma (6, 7). When a DNA sequence encoding the 125 aa residue of the mature CCL2 protein was expressed in *E. coli* at R&D Systems, the purified protein had the predicted N-terminus but a mass of 8525 Da. The truncation of most of the C-terminal extension could be due either to purification artifact or to post-translational modification. The truncated recombinant CCL2 has a potency similar to that of human MCP-1 in the monocyte chemotaxis assay.

References:

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6. Daly, C. *et al.* (2003) Microcirculation **10**:247.
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