

## Technical Data Sheet

## Recombinant Rat MCP-1

## Product Information

<b>Material Number:</b>	<b>555110</b>
<b>Size:</b>	5 µg
<b>Concentration:</b>	0.1 mg/ml
<b>Reactivity:</b>	QC Testing: Rat
<b>Storage Buffer:</b>	Frozen aqueous buffered solution containing BSA and glycerol.

## Description

Rat Monocyte Chemoattractant Protein (MCP-1) is a member of the C-C chemokine superfamily and a homolog of human MCP-1. MCP-1 is expressed by a variety of cell types including monocyte/macrophages, endothelial cells and mesangial cells.<sup>3</sup> MCP-1 has chemoattractant activity for monocytes, lymphocytes and basophils but is not active for neutrophils. Recombinant rat MCP-1 is a highly glycosylated protein ranging in size from 27 - 30 kD as measured by SDS-PAGE analysis. Furthermore, there is evidence from experimental animal models that MCP-1 can suppress tumor formation by attracting monocytes to the tumor site. Recombinant rat MCP-1 (MN 555110) is supplied as a frozen liquid comprised of 0.22 µm sterile-filtered aqueous buffered solution containing glycerol and 1.0 mg/ml bovine serum albumin, with no preservatives. Recombinant rat MCP-1 is ≥ 95% pure as determined by SDS-PAGE, and an absorbance assay based on the Beers-Lambert law. The endotoxin level is ≤ 0.1 ng per µg of rat MCP-1, as measured in a chromogenic LAL assay.

## Preparation and Storage

Store product at -80°C prior to use or for long term storage of stock solutions.

Rapidly thaw and quick-spin product prior to use.

Avoid multiple freeze-thaws of product.

This preparation contains no preservatives, thus it should be handled under aseptic conditions.

## Application Notes

## Application

ELISA Standard	Routinely Tested
Bioassay	Tested During Development
Intracellular block/flow cytometry	Tested During Development

## Recommended Assay Procedure:

Upon initial thawing, recombinant rat MCP-1 (MN 555110) should be aliquoted into polypropylene microtubes and frozen at -80°C for future use. Alternatively, the product can be diluted in sterile neutral buffer containing not less than 0.5 - 10 mg/mL carrier protein, such as human or bovine albumin, aliquoted and stored at -80°C. For *in vitro* biological assay use, carrier-protein concentrations of 0.5 - 1 mg/mL are recommended. For use as an ELISA standard, carrier-protein concentrations of 5 - 10 mg/mL are recommended. Failure to add carrier protein or store at indicated temperatures may result in a loss of activity. Carrier proteins should be pre-screened for possible effects in each investigator's experimental system. Carrier proteins may have an undesired influence on experimental results due to toxicity, high endotoxin levels or possible blocking activity.

**ELISA Standard:** Recombinant rat MCP-1 (MN 555110) can be useful as a quantitative standard for measuring rat MCP-1 protein levels using sandwich ELISA with the purified C4 antibody (Cat. No. 555072) as a capture antibody and the biotinylated B4 antibody (Cat. No. 555074) as the detection antibody. To obtain linear standard curves, investigators may want to consider using doubling dilutions of recombinant rat MCP-1 from 2000-5 pg/mL to be included for each ELISA plate. For measuring rat MCP-1 in serum or plasma, investigators are highly encouraged to use the BD OptEIA™ Rat MCP-1 Set (Cat. No. 555130).

**Bioassay:** Investigators are advised that the Bioassay application is not routinely tested for this material and are highly encouraged to both titrate this material and include appropriate controls in relevant experiments. An activity range encompassing an ED<sub>50</sub> = 60 - 600 ng/mL has previously been reported using THP-1 as indicator cells utilizing a calcium flux assay.

**Ligand Blocking Control for Immunofluorescent Staining of Cytokines:** Recombinant rat MCP-1 can be used as a blocking control to demonstrate the specificity of MCP-1 staining by the PE-conjugated format (Cat. No. 554443) of the 2H5 Anti-Rat MCP-1 antibody. Investigators are advised that this blocking application is not routinely tested for this material. The use of staining controls for the immunofluorescent staining and flow cytometric analysis of cytokine producing cells has been previously described (Prussin *et al.*).

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## Suggested Companion Products

Catalog Number	Name	Size	Clone
555072	Purified Mouse Anti-Rat MCP-1	0.5 mg	C4
555074	Biotin Mouse Anti-Rat MCP-1	0.5 mg	B4
555130	Rat MCP-1 ELISA Set	20 plates	(none)
554058	Avidin-Horseradish Peroxidase (HRP)	1.0 ml	(none)
555214	TMB Substrate Reagent Set	each	(none)
554443	PE Hamster Anti-Mouse/Rat MCP-1	0.1 mg	2H5

## Product Notices

1. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
2. Source of all serum proteins is from USDA inspected abattoirs located in the United States.
3. Please refer to [www.bdbiosciences.com/pharming/en/protocols](http://www.bdbiosciences.com/pharming/en/protocols) for technical protocols.

## References

Capsoni F, Minonzio F, Ongari AM, Zanussi C. A new simplified single-filter assay for 'in vitro' evaluation of chemotaxis of <sup>51</sup>Cr-labeled polymorphonuclear leukocytes. *J Immunol Methods*. 1989; 120(1):125-131. (Methodology)

Haelens A, Wuyts A, Proost P, Struyf S, Opdenakker G, van Damme J.. Leukocyte migration and activation by murine chemokines. *Immunobiology*. 1996; 195(4-5):499-521. (Biology)

Prussin C, Metcalfe DD. Detection of intracytoplasmic cytokine using flow cytometry and directly conjugated anti-cytokine antibodies. *J Immunol Methods*. 1995; 188(1):117-128. (Methodology)

Rollins BJ, Sunday ME. Suppression of tumor formation in vivo by expression of the JE gene in malignant cells.. *Mol Cell Biol*. 1991; 11(6):3125-3131. (Biology)

Yoshimura T, Takeya M, Takahashi K. Molecular cloning of rat monocyte chemoattractant protein-1 (MCP-1) and its expression in rat spleen cells and tumor cell lines. *Biochem Biophys Res Commun*. 1991; 174(2):504-509. (Biology)

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