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# Product Information

**Monoclonal Anti-Interleukin-8**
**Clone 6217.11**
produced in mouse, purified immunoglobulin

Catalog Number **I2519**

## Product Description

Monoclonal Anti-Interleukin-8 (IL-8) (mouse IgG1) is produced from the 6217.11 hybridoma produced by the fusion of mouse myeloma cells and splenocytes from BALB/c mice using purified recombinant human interleukin-8 expressed in *E. coli* as immunogen. The antibody is purified by Protein A affinity chromatography.

Monoclonal Anti-Interleukin-8 recognizes recombinant human interleukin-8 by various immunochemical techniques including immunoblotting, neutralization, ELISA capture, immunocytochemistry, and flow cytometry. By immunoblotting, the antibody shows 100% cross-reactivity with recombinant porcine IL-8, and no cross-reactivity with recombinant rat CINC-2α.

Interleukin-8 (IL-8), also called CXCL8, formerly called monocyte-derived neutrophil chemotactic factor, belongs to the chemokine-α family.[1] IL-8 exhibits chemotactic activity *in vitro* for T cells,[2] basophils, and neutrophils.[3]

## 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 sterile phosphate buffered saline (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 up to one month. For prolonged storage, freeze in working aliquots at –20 °C. Avoid repeated freezing and thawing. Do not store in a frost-free freezer.

## Product Profile

The antibody has the ability to neutralize human IL-8 bioactivity. 0.08–0.4 μg/mL of the antibody will neutralize 50% of the bioactivity in the presence of 20 ng/mL of recombinant human IL-8, Catalog Number I1645,using hCXCR-2 transfected BaF/3 cells. The exact concentration of antibody required to neutralize recombinant human IL-8 activity is dependent on the cytokine concentration, cell type, growth conditions, and the type of activity.

The Neutralization $Dose_{50}$ ($ND_{50}$) for this antibody is defined as that concentration required to yield one-half maximal inhibition of the cytokine activity on a responsive cell line, when that cytokine is present at a concentration just high enough to elicit a maximum response.

Immunoblotting: a working concentration of 1–2 μg/mL is recommended. Using a colorimetric detection system, the detection limit for recombinant human IL-8 is ~5 ng/lane under non-reducing and reducing conditions in SDS-PAGE. A chemiluminescent detection substrate will increase the sensitivity by 5 to 50-fold.

For capture ELISAs, Monoclonal Anti-Interleukin-8 can be used as the capture antibody. A working concentration of 0.5 μg/mL (100 μL/well) in combination with a biotinylated detection antibody is recommended.

Immunocytochemistry: a working concentration of 25 μg/mL is recommended using calcium ionophore- and PMA-stimulated human peripheral blood mononuclear cells in a chromogenic detection system.

For flow cytometry (intracellular staining), the cells must first be fixed and permeabilized using 4% paraformaldehyde and 0.1% saponin. The cells ($1–5 \times 10^5$) are stained with the primary antibody using 10 µl of a 25 µg/mL stock solution. Following a 30 minute incubation, the cells should be washed with 0.1% saponin prior to using a fluorochrome conjugated secondary reagent for detection (typically, 250 ng of a polyclonal antibody per reaction may added to develop the staining). After incubation with the fluorochrome, the cells should be washed for a final time in 0.1% saponin prior to analysis.

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

Endotoxin: <0.1 EU (endotoxin units) per 1µg of antibody as determined by the *Limulus* amebocyte lysate (LAL) method.

**References**


1. Yoshimure, T. et al., *Proc. Natl. Acad. Sci. USA*, **84**, 9233 (1987).
2. Larsen, C. et al., *Science*, **243**, 1464 (1989).
3. Mukaida, N. et al, *Microbiol. Immunol*., Vol. 36, **8**, 773 (1992


KAA,PHC,TMS,MAM 06/16-1