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Product Information

Anti-Interleukin-8
produced in goat, affinity isolated antibody

Catalog Number **I8026**

Product Description

Anti-Interleukin-8 (IL-8, CXCL8) is produced in goat using as immunogen a recombinant human IL-8 (rhIL-8) expressed in *E. coli*. The antibody is purified using human IL-8 affinity chromatography.

Anti-Interleukin-8 will neutralize the biological activity of both rhIL-8 and natural human IL-8. It will not neutralize the biological activity of rhGROα. The antibody may also be used in immunoblotting and immuno-histochemistry.

Interleukin 8 (IL-8), formerly called monocyte-derived neutrophil chemotactic factor, belongs to the α or C-X-C chemokine family.[1] The mature form of IL-8 has 4 cysteine residues, as do other members of the chemokine family, and the first two cysteine residues are separated by glutamine.[2] IL-8 genomic DNA consists of 4 exons and 3 introns with a single "TATA" and "CAT"-like sequence.[2] The mature form of human IL-8 consists of 72 amino acids with a molecular mass of 8,000 Daltons. IL-8 exhibits chemotactic activity *in vitro* for T cells,[3] basophils, and neutrophils.[2] IL-8 activates neutrophils to release lysosomal enzymes including myeloperoxidase, α-mannosidase, and β-glucuronidase.[2]

IL-1 will induce the production of IL-8 from fibroblasts, keratinocytes, endothelial cells, hepatoma cells, astrocytoma cells, glioblastoma cells, lung epithelial cells, synovial membrane cells, melanocytes, melanoma cells, and gastric cancer cells.[2] LPS will also stimulate IL-8 production in monocytes/macrophages.[2]

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 0.1 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.

Product Profile

Neutralization: the antibody has the ability to neutralize the biological activity of natural and recombinant IL-8.

To measure the ability of the antibody to neutralize the chemo-attractant activity of rhCXCL8 for BaF/3 cells transfected with hCXCR2 cells, rhIL-8 was preincubated with various dilutions of the antibody for 30 minutes at room temperature in a 96 well plate. Following this preincubation, 75 μL of the cytokine-antibody solution (containing rhIL-8 at a final concentration of 20 ng/mL and antibody at concentrations from 0.01-20 μg/mL) was transferred to the lower compartment of a 96 well chemotaxis chamber. The chamber was then assembled using a PVP-free polycarbonate filter (5 μm pore size) and 0.2×10^4 cells/well was added to the top chamber. After incubation for 3 hours at 37 °C in a 5% CO_2 humidified incubator, the chamber was disassembled and the cells that migrated through to the lower chamber were transferred to a working plate and quantitated overnight. The fluorescence was then read in a fluorescent plate reader (λ_{ex} = 544/λ_{em} = 590 nm).

The ND_{50} of the antibody is defined as the concentration of antibody resulting in a one-half maximal inhibition of bioactivity of rhIL-8 that is present at a concentration just high enough to elicit a maximum response.

Immunocytochemistry: 5-15 μg/mL may be used to detect human IL-8 (CXCL8).

Immunoblotting: 0.1 μg/mL detects rhIL-8 at 5 ng/lane under non-reducing and reducing conditions.

Immunohistochemistry: 5-15 μg/mL may be used to detect IL-8 in cultured cells or tissue sections.

Note: In order to obtain the best results in various techniques and preparations, it is recommended to determine optimal working dilutions by titration.

References

1. Yoshimura, T. et al., *Proc. Natl. Acad. Sci. USA*, **84**, 9233 (1987).
2. Mukaida, N. et al., *Microbiol. Immunol*., **36**, 773 (1992).
3. Larsen, C.G. et al., *Science*, **243**, 1464 (1989).
4. Schröder, J.M. et al., *J. Immunol*., **139**, 3474 (1987).

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