Anti-human CCL23/Ckβ8-1 Antibody

ORDERING INFORMATION

Catalog Number: AF508

Lot Number: DFB01

Size: 100 µg

Formulation: 0.2 µm filtered solution in PBS

Storage: -20° C

Reconstitution: sterile PBS

Specificity: human CCL23

Immunogen: *E. coli*-derived rhCCL23

Ig Type: goat IgG

Applications: Neutralization of bioactivity
Western blot
Direct ELISA

Preparation
Produced in goats immunized with purified, *E. coli*-derived, recombinant human CCL23 (rhCkβ8-1), a splice variant of CCL23/Ckβ8/MPIF-1. CCL23 specific IgG was purified by human CCL23 affinity chromatography.

Formulation
Lyophilized from a 0.2 µm filtered solution in phosphate-buffered saline (PBS).

Endotoxin Level
< 0.2 EU per 1 µg of the antibody as determined by the LAL method.

Reconstitution
Reconstitute with sterile PBS. If 1 mL of PBS is used, the antibody concentration will be 0.1 mg/mL.

Storage
Lyophilized samples are stable for twelve months from date of receipt when stored at -20° C to -70° C. Upon reconstitution, the antibody can be stored at 2° - 8° C for 1 month without detectable loss of activity. Reconstituted antibody can also be aliquotted and stored frozen at -20° C to -70° C **in a manual defrost freezer** for six months without detectable loss of activity. **Avoid repeated freeze-thaw cycles.**

Specificity
This antibody has been selected for its ability to neutralize the biological activity of rhCCL23. It will also neutralize the biological activity of rhMPIF-1 at a 10-fold higher Ig concentration. In direct ELISAs and western blots, this antibody shows approximately 20% cross-reactivity with rhMPIF-1. Additionally, in direct ELISAs, this antibody shows no cross-reactivity with other chemokines tested.[1]

Neutralization of Human CCL23 Bioactivity
The exact concentration of antibody required to neutralize rhCCL23 activity is dependent on the cytokine concentration, cell type, growth conditions and the type of activity studied. To provide a guideline, R&D Systems has determined the neutralization dose for this antibody under a specific set of conditions. The **Neutralization Dose$_{50}$ (ND$_{50}$)** for this antibody is defined as that concentration of antibody 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.

The ND$_{50}$ for this lot of anti-human CCL23 antibody was determined to be approximately 0.15 - 0.75 µg/mL in the presence of 0.04 µg/mL of rhCCL23, using the BaF/3 CCR1 transfected cell line. The specific conditions are described in the figure legends.

Additional Applications
Direct ELISA - This antibody can be used at 0.5 - 1.0 µg/mL with the appropriate secondary reagents to detect human CCL23. The detection limit for rhCCL23 is approximately 0.3 ng/well.

Western blot - This antibody can be used at 0.1 - 0.2 µg/mL with the appropriate secondary reagents to detect human CCL23. The detection limit for rhCCL23 is approximately 2 ng/lane under non-reducing and reducing conditions.

Optimal dilutions should be determined by each laboratory for each application.

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1-800-343-7475

Figure 1

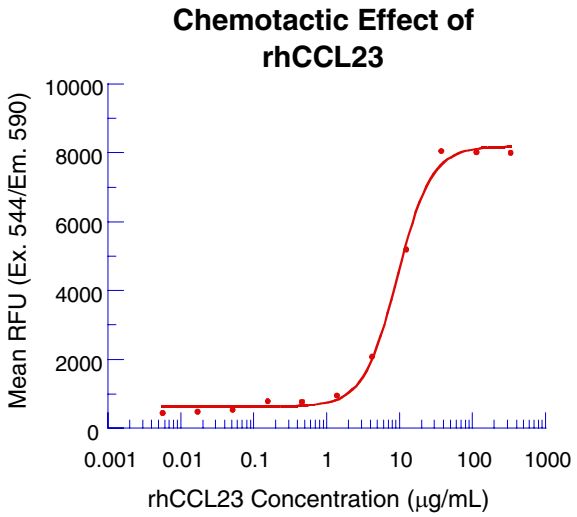

Figure 2

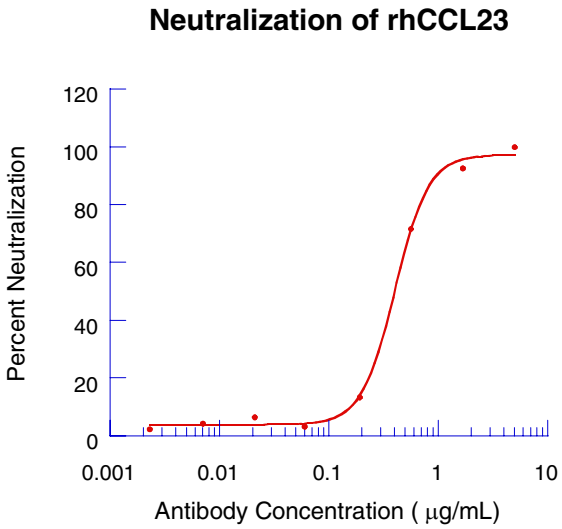

Figure 1
Human CCL23 chemoattracts hCCR1 transfected BaF/3 cells. The number of cells that have migrated through to the lower chamber are quantitated using Resazurin dye (R&D Systems, Catalog # AR002). The ED_{50} for this effect is typically 2 - 10 μg/mL.

Figure 2
To measure the ability of the antibody to neutralize the chemoattractant activity of rhCCL23 for BaF/3 hCCR1 cells, rhCCL23 was incubated with various concentrations of antibody for 30 minutes at room temperature in a 96-well microplate. Following this preincubation period, 75 μL of the cytokine-antibody solution (containing rhCCL23 at a final concentration of 0.04 μg/mL and antibody at the concentrations indicated) was transferred to the lower compartment of a 96-well chemotaxis chamber (NeuroProbe, Cabin John, MD). The chemotaxis chamber was then assembled using a PVP-free polycarbonate filter (5 micron pore size) and 0.25 x 10^6 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 using Resazurin dye (R&D Systems, Catalog # AR002). The relative fluorescence was then read in a fluorescent plate reader set at Ex. 544/Em. 590. As shown in Figure 2, the ND_{50} for this lot of antibody is approximately 0.15 - 0.75 μg/mL.

[1]rh6Ckine, rm6Ckine, rhBLC/BCA-1, rmBLC, rmC10, rrCINC-1, rrCINC-2α, rrCINC-2β, rmCRG-2, rhENA-78, rhEotaxin, rhEotaxin 2, rmEotaxin, rhFractalkine, rmFractalkine, rhGCP-2, rmGCP-2, rhGROα, rhGROβ, rhGROγ, rhHCC-4, rhI-309, rhIL-8, rhIP-10, rhI-TAC, rmJE, rmKC, rmLymphotactin, rmMarc, rhMCP-1, rhMCP-2, rhMCP-3, rhMCP-4, rmMCP-5, rhMDC, rmMDC, rhMIG, rmMIG, rhMIP-1α, rmMIP-1α, rhMIP-1β (rhACTII), rmMIP-1β, rmMIP-1γ, rmMIP-2, rhMIP-3α, rrMIP-3α, rhMIP-3β, rmMIP-3β, rvMIP-I, rvMIP-II, rvMIP-III, rhNAP-2, rhParc, rhRANTES, rmRANTES, rhSDF-1α, rmSDF-1α, rhSDF-1β, rhTARC, rmTCA-3, rmTeck, rhVIC