Anti-rat LIX Antibody

ORDERING INFORMATION

Catalog Number: AF543

Lot Number: DVY01

Size: 100 μg

Formulation: 0.2 μm filtered solution in PBS

Storage: -20° C

Reconstitution: sterile PBS

Specificity: rat LIX

Immunogen: *E. coli*-derived rrLIX

Ig Type: rat LIX specific goat IgG

Applications: Neutralization of bioactivity
Western blot
ELISA

Preparation
Produced in goats immunized with purified, *E. coli*-derived, recombinant rat LIX (rrLIX). Rat LIX specific IgG was purified by rat LIX affinity chromatography.

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

Endotoxin Level
< 0.1 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 rrLIX in bioactivity.

Neutralization of Rat LIX Bioactivity
The exact concentration of antibody required to neutralize rrLIX 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-rat LIX antibody was determined to be approximately 0.3 - 1.5 μg/mL in the presence of 0.2 μg/mL of rrLIX, using BaF/3 cells transfected with rhCXCR-2. 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 rat LIX. The detection limit for rrLIX is approximately 0.2 ng/well. In this format, this antibody shows approximately 30% cross-reactivity with rmGCP-2, less than 5% cross-reactivity with rhGCP-2 and no cross-reactivity with other chemokines tested[1].

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

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

[1]rh6Ckine, rm6Ckine, rhBLC/BCA-1, rmBLC, rmC10, rrCINC-1, rrCINC-2α, rrCINC-2β, rrCINC-3, rhCkβ8-1, rvCMV UL146, rmCRG-2, rhENA-78, rhEotaxin, rmEotaxin, rhEotaxin-2, rmEotaxin-2, rhEotaxin-3, rhFractalkine, rmFractalkine, rrFractalkine, rhGROα, rhGROβ, rhHCC-1, rhHCC-4, rhI-309, rhIL-8, rpIL-8, rhIP-10, rhI-TAC, rmJE, rmKC, rhLeukotactin-1, rhLymphotactin, rmLymphotactin, rmMARC, rhMCP-1, rhMCP-2, rhMCP-3, rhMCP-4, rmMCP-5, rhMDC, rmMDC, rhMIG, rmMIG, rhMIP-1α, rmMIP-1α, rhMIP-1β, rmMIP-1β, rhMIP-1δ, rmMIP-1γ, rvMIP-I, rmMIP-2, rvMIP-II, rhMIP-3α, rmMIP-3α, rrMIP-3α, rhMIP-3β, rmMIP-3β, rvMIP-III, rhMPIF-1, rhNAP-2, rhPARC, rhRANTES, rmRANTES, rhSDF-1α, rmSDF-1α, rhSDF-1β, rhTarc, rmTarc, rmTCA-3, rhTeck, rmTeck

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1-800-343-7475

Figure 1

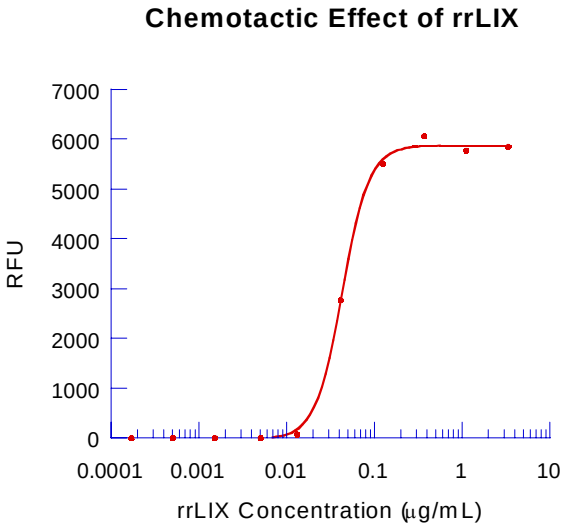

Figure 2

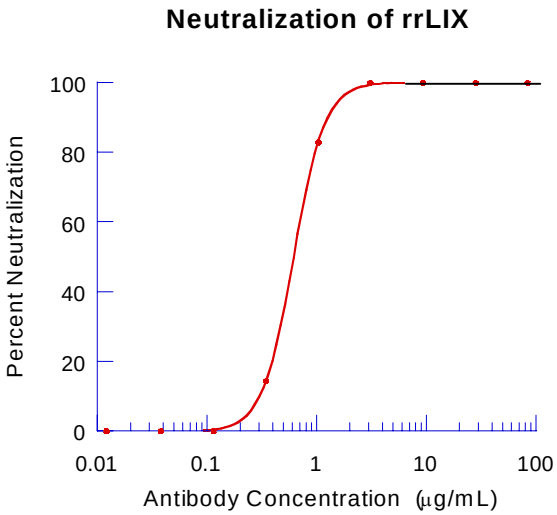

Figure 1
Recombinant rat LIX chemoattracts BaF/3 cells transfected with rhCXCR-2. The ED_{50} for this effect is typically 30 - 150 ng/mL.

Figure 2
To measure the ability of the antibody to neutralize the chemoattractant activity of rrLIX for BaF/3 hCXCR-2 transfected cells, rrLIX was incubated with various concentrations of the antibody for 15 minutes at room temperature in a 96 well microplate. Following this preincubation period, 75 µL of the cytokine-antibody solution (containing rrLIX at a final concentration of 0.2 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 0.2 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 carefully disassembled. The cells that migrate through to the lower chamber were transferred to a 96 well plate. Chemotaxis was measued by Resazurin (R&D Systems, Catalog # AR002) staining of cells that have migrated through the filter. As shown in figure 2, the ND_{50} for this lot of antibody is approximately 0.3 – 1.5 µg/mL.