

DESCRIPTION	
Species Reactivity	Human
Specificity	Detects human CXCL1/GRO α /KC/CINC-1 in direct ELISAs and Western blots. In direct ELISAs, less than 50% cross-reactivity with recombinant human (rh) GRO β and rhGRO γ is observed, less than 25% cross-reactivity with recombinant rat (rr) CINC-1 is observed and less than 15% cross-reactivity with recombinant mouse (rm) KC is observed. In Western blots, no cross-reactivity with rhNAP-2 or rhPF-4 is observed.
Source	Polyclonal Goat IgG
Purification	Antigen Affinity-purified
Immunogen	<i>E. coli</i> -derived recombinant human CXCL1/GRO α /KC/CINC-1 (R&D Systems, Catalog # 275-GR) Ala35-Asn107 Accession # P09341
Endotoxin Level	<0.10 EU per 1 μ g of the antibody by the LAL method.
Formulation	Lyophilized from a 0.2 μ m filtered solution in PBS with Trehalose. See Certificate of Analysis for details. *Small pack size (-SP) is supplied as a 0.2 μ m filtered solution in PBS.

APPLICATIONS

Please Note: Optimal dilutions should be determined by each laboratory for each application. *General Protocols* are available in the *Technical Information* section on our website.

	Recommended Concentration	Sample
Western Blot	0.1 μ g/mL	See Below
Neutralization	Measured by its ability to neutralize CXCL1/GRO α /KC/CINC-1-induced chemotaxis in the BaF3 mouse pro-B cell line transfected with human CXCR2. The Neutralization Dose (ND ₅₀) is typically 0.03-0.15 μ g/mL in the presence of 0.01 μ g/mL Recombinant Human CXCL1/GRO α /KC/CINC-1.	

DATA

Western Blot

Detection of Recombinant Human CXCL1/GRO α /KC/CINC-1 by Western Blot.

Western blot shows 25 ng of Recombinant Human CXCL1/GRO α /KC/CINC-1 (Catalog # 275-GR), Recombinant Mouse CXCL1/GRO α /KC/CINC-1 aa 20-96 (Catalog # 453-KC), Recombinant Rat CXCL1/GRO α /KC/CINC-1 (Catalog # 515-CN), Recombinant Human CXCL2/GRO β /MIP-2/CINC-3 aa 35-107 (Catalog # 276-GB), Recombinant Human CXCL3/GRO γ /CINC-2/DCIP-1 (Catalog # 277-GG), Recombinant Human CXCL7/NAP-2 (Catalog # 393-NP), and Recombinant Human CXCL4/PF4 (Catalog # 795-P4), PVDF Membrane was probed with 0.1 μ g/mL of Goat Anti-Human CXCL1/GRO α /KC/CINC-1 Antigen Affinity-purified Polyclonal Antibody (Catalog # AF275) followed by HRP-conjugated Anti-Goat IgG Secondary Antibody (Catalog # HAF109). A specific band was detected for CXCL1/GRO α /KC/CINC-1 at approximately 11 kDa (as indicated). This experiment was conducted under reducing conditions and using Immunoblot Buffer Group 3.

Neutralization

Chemotaxis Induced by CXCL1/GRO α and Neutralization by Human CXCL1/GRO α Antibody.

Recombinant Human CXCL1/GRO α (Catalog # 275-GR) chemoattracts the BaF3 mouse pro-B cell line transfected with human CXCR2 in a dose-dependent manner (orange line). The amount of cells that migrated through to the lower chemotaxis chamber was measured by Resazurin (Catalog # AR002). Chemotaxis elicited by Recombinant Human CXCL1/GRO α (0.01 μ g/mL) is neutralized (green line) by increasing concentrations of Human CXCL1/GRO α Antigen Affinity-purified Polyclonal Antibody (Catalog # AF275). The ND₅₀ is typically 0.03-0.15 μ g/mL.

PREPARATION AND STORAGE

Reconstitution	Reconstitute at 0.2 mg/mL in sterile PBS.
Shipping	The product is shipped at ambient temperature. Upon receipt, store it immediately at the temperature recommended below. *Small pack size (-SP) is shipped with polar packs. Upon receipt, store it immediately at -20 to -70 °C
Stability & Storage	Use a manual defrost freezer and avoid repeated freeze-thaw cycles. <ul style="list-style-type: none"> ● 12 months from date of receipt, -20 to -70 °C as supplied. ● 1 month, 2 to 8 °C under sterile conditions after reconstitution. ● 6 months, -20 to -70 °C under sterile conditions after reconstitution.

BACKGROUND

The gene for CXCL1/GRO α was initially discovered in hamster cells, using subtractive hybridization techniques, as a message that is over-expressed in tumorigenic cells and in normal cells during growth stimulation. The hamster cDNA was cloned and used as a probe for the subsequent cloning of the human GRO cDNA. Independently, a cDNA encoding a secreted protein with melanoma growth stimulating activity (MGSA) was also cloned from a human melanoma cell line and found to be identical to GRO. In addition to the initially cloned GRO gene, now designated CXCL1, two additional GRO genes, GRO β or MIP-2 α and GRO γ or MIP-2 β , which shared 90% and 86% amino acid sequence homology, respectively, with CXCL1, have been identified. All three human GROs are members of the alpha (C-X-C) subfamily of chemokines. The three GRO cDNAs encode 107 amino acid precursor proteins from which the N-terminal 34 amino acid residues are cleaved to generate the mature GROs. There are no potential N-linked glycosylation sites in the amino acid sequences. GRO expression is inducible by serum or PDGF and/or by a variety of inflammatory mediators, such as IL-1 and TNF, in monocytes, fibroblasts, melanocytes, and epithelial cells. In certain tumor cell lines, GRO is expressed constitutively. Similar to other alpha chemokines, the three GRO proteins are potent neutrophil attractants and activators. In addition, these chemokines are also active toward basophils. All three GROs can bind with high affinity to the IL-8 receptor type B. The rat homolog of human CXCL1, CINC, is much more active than human CXCL1 on rat neutrophils, suggesting that this cytokine may have selective species specificity.