



Anti-human PD-ECGF Neutralizing Antibody

ORDERING INFORMATION

Catalog Number: AB-229-NA

Lot Number: EV04

Size: 1 mg

Formulation: sterile solution in PBS

Storage: -20° C

Reconstitution: sterile PBS

Specificity: rhPD-ECGF

Antigen: Sf 21-derived rhPD-ECGF

Ig class: goat IgG

Applications: Neutralization of bioactivity
Western blot
ELISA

Preparation

Produced in goats immunized with purified, insect cell line Sf 21-derived, recombinant human platelet-derived endothelial cell growth factor (PD-ECGF). Total IgG was purified by Protein G affinity chromatography.

Formulation

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

Endotoxin Level

< 10 ng per 1 mg 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 1 mg/mL.

Storage

Lyophilized samples are stable for greater than 6 months at -20° C to -70° C. Reconstituted antibody is stable for at least 1 month at 2° - 4° C or 3 months at -20° C to -70° C under sterile conditions. **Avoid repeated freeze-thaw cycles.**

Specificity

This antibody has been selected for its ability to neutralize the biological activity of rhPD-ECGF. Based on direct ELISA this antibody shows no cross-reactivity with other cytokines tested.¹

Neutralization of Human PD-ECGF Bioactivity

The exact concentration of antibody required to neutralize rhPD-ECGF bioactivity 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 Dose₅₀ (ND₅₀)** 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.

As shown in figures 1 and 2 on the next page, the ND₅₀ for this lot of anti-human PD-ECGF antibody was determined to be approximately 10 - 25 µg/mL in the presence of 150 ng/mL of rhPD-ECGF, using PD-ECGF responsive HUVE cells. The experimental details and specific conditions are described in the figure legends.

Additional Applications

For direct ELISAs, the antibody can be used at 0.5 - 1.0 µg/mL with the appropriate secondary reagents to detect rhPD-ECGF. The detection limit for rhPD-ECGF is approximately 0.3 ng/well.

For western blot analysis, the antibody can be used at 1 - 2 µg/mL with the appropriate secondary reagents to detect rhPD-ECGF. The detection limit for rhPD-ECGF is approximately 5 ng/lane under both non-reducing and reducing conditions. Because this antibody preparation is a total IgG fraction, complete monospecificity cannot be assumed.

Figure 1

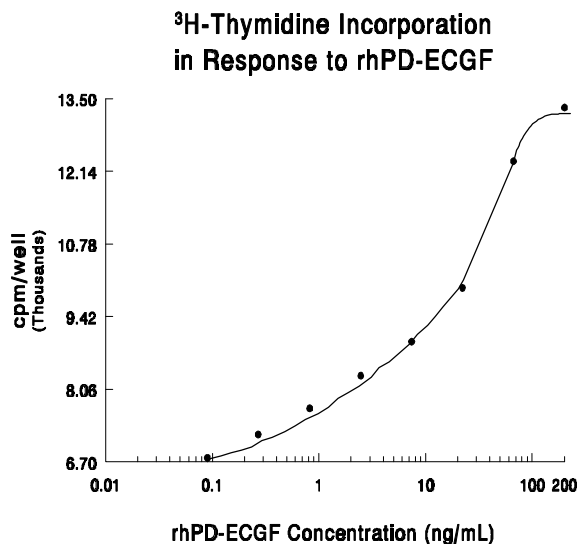


Figure 2

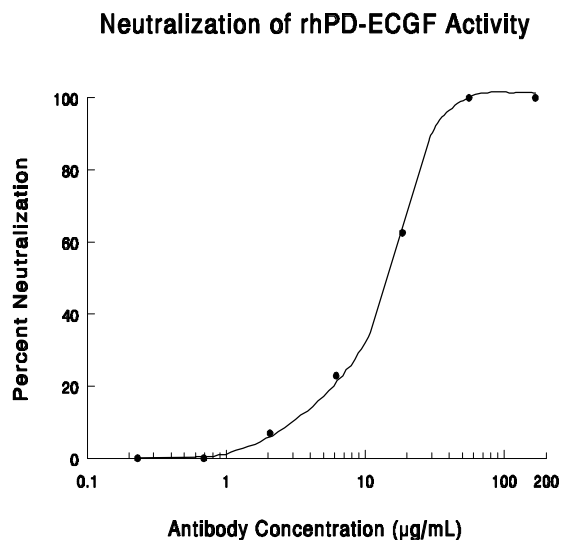


Figure 1

Human PD-ECGF stimulates the ³H-thymidine incorporation by human umbilical vein endothelial cells (Usuki, K., *et al.*, 1990, Cell Regulation 1:577). The ED₅₀ for this effect is typically 20 - 40 ng/mL.

Figure 2

To measure the ability of the antibody to neutralize rhPD-ECGF response, rhPD-ECGF was added to a 96 well plate containing various concentrations of antibody and preincubated for 1 hour at room temperature. Following this preincubation, HUVE cells were added. The assay mixture in a total volume of 100 µL, containing antibody at the concentrations indicated, rhPD-ECGF at 150 ng/mL and cells at 1 x 10⁵ cells/mL, was incubated at 37° C for 72 hours in a humidified CO₂ incubator. ³H-thymidine was added during the final 20 hours of incubation. The cells were subsequently detached and harvested onto glass fiber filters and the ³H-thymidine incorporated into DNA was determined. The ND₅₀ of the antibody under these conditions is approximately 10 - 25 µg/mL.

¹rhANG, rhAR, rhβ-Cellulin, rhβ-NGF, rmC10, rhCNTF, rrCNTF, rhEGF, rhENA-78, rhEpo, rhFGF acidic, rhFGF basic, rhFGF-3, rhFGF-4, rhFGF-5, rhFGF-6, rhFGF-7, rhG-CSF, rhGDNF, rhGM-CSF Rα, rmGM-CSF, rhGROα, rhGROβ, rhGROγ, rhHB-EGF, rhHGF, rhIL-309, rhIFN-γ, rhIGF-I, rhIGF-1 RI, rhIGF II, rhIL-1α, rhIL-1 RI, rhIL-1 RII, rmIL-1α, rhIL-1β, rmIL-1β, rhIL-1ra, rhIL-2, rhIL-2 sRα, rhIL-2 sRβ, rhIL-3, rhIL-3 sRα, rmIL-3, rhIL-4, rhIL-4 sR, rmIL-4, rhIL-5, rhIL-5 sRα, rhIL-5 sRβ, rmIL-5, rhIL-6, rhIL-6 sR, rmIL-6, rhIL-7, rhIL-7 R, rmIL-7, rhIL-8, rhIL-9, rmIL-9, rhIL-10, rmIL-10, rhIL-11, rhIL-12, rhIL-13, rmIL-13, rhIP-10, rmJE, rhLIF, rhLIF R, rmLIF, rhM-CSF, rmM-CSF, rhMCP-1, rhMCP-1 R, rhMidkine, rhMIP-1α, rmMIP-1α, rhMIP-1β, rmMIP-1β, rhNT-4, rhOSM, hPDGF, pPDGF, rhPDGF-AA, rhPDGF-AB, rhPDGF-BB, rhP/GF, rhPTN, rhRANTES, rhSCF, rmSCF, rhsgp130, rhSLPI, hTfR, rhTGF-α, rhTGF-β1, rhTGF-β2, rhTGF-β3, raTGF-β5, rhLAP (TGF-β1), rhLatent TGF-β1, rhTGF-β sRII, rhTGF-β sRIII, rhTNF-α, rmTNF-α, rhTNF-β, rhTNF RI, rhTNF RII, rhVEGF