Anti-rhesus macaque IL-18 Antibody

ORDERING INFORMATION

Catalog Number: AF2548

Lot Number: VVS01

Size: 100 µg

Formulation: 0.2 µm filtered solution in PBS with 5% trehalose

Storage: -20° C

Reconstitution: sterile PBS

Specificity: rhesus macaque IL-18

Immunogen: *E. coli*-derived rrmIL-18

Ig Type: goat IgG

Applications: Neutralization of bioactivity
Western blot
Immunocytochemistry
Direct ELISA

Preparation
Produced in goats immunized with purified, *E. coli*-derived, recombinant rhesus macaque interleukin 18 (rrmIL-18; R&D Systems' Catalog # 2548-RM). Rhesus macaque IL-18 specific IgG was purified by rhesus macaque IL-18 affinity chromatography.

Formulation
Lyophilized from a 0.2 µm filtered solution in phosphate-buffered saline (PBS) with 5% trehalose.

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 rhesus macaque IL-18 bioactivity.

Applications
Neutralization of Rhesus Macaque IL-18 Bioactivity - The exact concentration of antibody required to neutralize rrmIL-18 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-rhesus macaque IL-18 antibody was determined to be approximately 0.4 - 1.2 µg/mL in the presence of 10 ng/mL of rrmIL-18 and KG-1 cells at 1 x 10^6 cells/mL. The specific conditions are described in the figure legends.

Western blot - This antibody can be used at 0.1 - 0.2 µg/mL with the appropriate secondary reagents to detect rhesus macaque and human IL-18. The detection limit for rrmIL-18 and rhIL-18 is approximately 20 ng/lane and 5 ng/lane under non-reducing and reducing conditions, respectively. In this format, this antibody shows approximately 10% cross-reactivity with rmIL-18, rrIL-18 and rpIL-18.

Immunocytochemistry - This antibody will detect IL-18 in cells. The working dilution is 2 - 15 µg/mL. For chromogenic detection of labeling, use R&D Systems' Cell and Tissue Staining Kits (CTS Series).

Direct ELISA - This antibody can be used at 0.5 - 1.0 µg/mL with the appropriate secondary reagents to detect rhesus macaque and human IL-18. The detection limit for rrmIL-18 and rhIL-18 is approximately 0.5 ng/well.

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

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1-800-343-7475

Figure 1

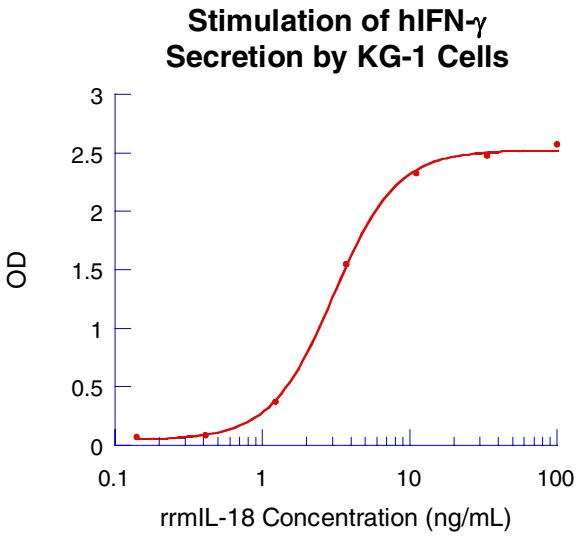

Figure 2

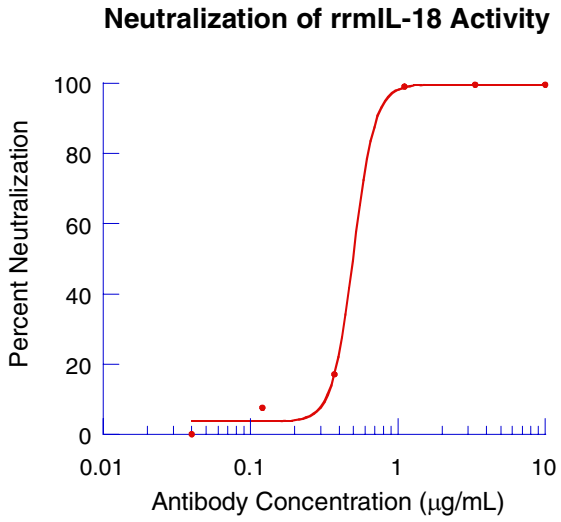

Figure 1
rrmIL-18 stimulates IFN-γ secretion by KG-1 cells (Novick, D. *et al.*, 1999, Immunity **10**(1):127 - 136). The ED_{50} for this effect is typically 1 - 4 ng/mL.

Figure 2
To measure the ability of the antibody to neutralize rhesus macaque IL-18 bioactivity on human KG-1 cells, various concentrations of the antibody were incubated with rrmIL-18 in a 96 well microplate for 1 hour at 37° C. Following this incubation period, rhTNF-α stimulated KG-1 cells were then added to the mixture. The assay mixture, in a total volume of 200 μL, containing antibody at the concentrations indicated, rrm IL-18 at 10 ng/mL, rhTNF-a at 20 ng/mL and cells at 1 x 10^6 cells/mL, was incubated at 37° C for 1 day in a humidified CO2 incubator. At the end of the incubation, 50 μL of supernatant was collected from each well, diluted 1:3 with ELISA diluent, and assayed for human IFN-γ levels using a human IFN-γ ELISA kit (R&D Systems, Catalog # DIF50). The ND_{50} of the antibody under these conditions is approximately 0.4 - 1.2 μg/mL.