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ProductInformation

ANTI-INTERLEUKIN-18 (IL-18) RECEPTOR, HUMAN
Developed in Goat,
Affinity Isolated Antibody

Product Number **I4278**

Product Description

Anti-Interleukin-18 receptor (IL-18 R), Human is developed in goat using purified recombinant human IL-18 R, expressed in mouse NSO cells, as immunogen. The antibody is purified using human IL-18 R affinity chromatography.

Anti-Human IL-18 R may be used to neutralize the bioactivity of recombinant human IL-18 R. The antibody may also be used for immunoblotting and ELISA. By ELISA, the antibody show 5 % cross-reactivity with recombinant mouse IL-18 R.

Interleukin 18, also known as interferon-gamma-inducing factor (IGIF) and IL-1γ, shares some biologic activities with IL-12 and structural similarities with the IL-1 family of proteins. It plays an important role in the T cell helper type 1 (Th1) response through its ability to induce IFN-γ production in T cells and NK cells. IL-18 is related to the IL-1 family in terms of structure, receptor family and function. Structurally, IL-18 and IL-1β share primary amino acids sequences (signature sequence motif) and are similarly folded as all-β pleated sheet molecules. Similar to IL-1β, production of IL-18 requires caspase-1 (interleukin-1β converting enzyme, ICE) to cleave the pro-domain from the precursor protein to produce active mature IL-18. Gene expression and synthesis of TNF, IL-1, Fas ligand and several chemokines are induced by IL-18. As with IL-1, IL-18 participates in both innate and acquired immunity. IL-18 actions are mediated by an IL-18 receptor (IL-18 R) complex comprised of a binding chain (IL-18 Rα, also called IL-1 receptor-related protein, IL-1 Rrp) and a signal-transducing chain. Both chains are members of the IL-1 receptor family. The IL-18 R complex recruits the IL-1 R-activating kinase (IRAK) and TNFR-associated factor-6 (TRAF-6) which phosphorylates NF-κB-inducing kinase (NIK) with subsequent activation of NF-κB.

Reagents

Anti-Human IL-18 R is supplied lyophilized from a 0.2 μm filtered solution of phosphate buffered saline. Endotoxin level is < 10 ng per mg antibody as determined by the LAL method.

Preparation Instructions

To one vial of lyophilized powder, add 1 ml of 0.2 μm-filtered PBS to produce a 0.1 mg/ml stock solution of antibody. If aseptic technique is used, no further filtration should be needed for use in cell culture environments.

Storage/Stability

Prior to reconstitution, store at –20 °C. Reconstituted product may be stored at 2 °C to 8 °C for at least one month. For prolonged storage, freeze in working aliquots at –20 °C. Avoid repeated freezing and thawing

Procedure

Neutralization of Bioactivity

To measure the ability of the antibody to neutralize the bioactivity of human IL-18 R, various concentrations of the antibody were incubated with TNF-α stimulated human KG-1 cells at 2 x 10^5 cells/well for 1 hour at 37°C in a 96 well plate. Following preincubation, recombinant human IL-18 was added at 40 ng/ml. Each well contained antibody at concentrations of 0.01-10 μg/ml, recombinant human IL-18 at 40 ng/ml, TNF-α at 20 ng/ml, and cells at 1x10^6 cells/ml. Each well had a total volume of 200 μl/well and the plate was incubated at 37°C for 1 day in a humidified CO_2 incubator. After incubation, 100 μl of supernatant was collected from each well, diluted 1:2 with phosphate buffered saline and tested for IFN-γ levels using an IFN-γ ELISA kit. The ND_{50} is the concentration of antibody required to yield one-half maximal inhibition of the cytokine activity on a responsive cell line, when the cytokine is present at a concentration just high enough to elicit a maximum response.

Product Profile

For neutralization, a working concentration of 0.1-0.4 µg/ml of Anti-Human IL-18 R will neutralize 50% of the bioactivity due to 40 ng/ml recombinant human IL-18 using stimulated KG-1 cells.

For indirect ELISA, a working concentration of 0.5-1.0 µg/ml is determined to detect a limit of ~0.6 ng/well of recombinant human IL-18 R.

For indirect immunoblotting, a working concentration of 0.1-0.2 µg/ml is determined using human IL-18 R at 5 ng/lane under non-reducing and reducing conditions.

Note: In order to obtain best results in different techniques and preparations we recommend determining optimal working dilutions by titration test.

References

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