3050 Spruce Street
Saint Louis, Missouri 63103 USA
Telephone 800-325-5832 • (314) 771-5765
Fax (314) 286-7828
email: techserv@sial.com
sigma-aldrich.com

ProductInformation

Monoclonal Anti-Interleukin-9 Receptor
Clone 33423
Purified Mouse Immunoglobulin

Product Number **I 3157**

Product Description

Monoclonal Anti-Human Interleukin-9 Receptor (clone 33423) (mouse IgG1) is produced from a mouse hybridoma elicited from a mouse immunized with purified recombinant human soluble interleukin-9 receptor (IL-9 R) expressed in insect *Sf* 21 cells as immunogen. The antibody is purified by Protein A affinity chromatography.

Monoclonal Anti-Human Interleukin-9 Receptor recognizes recombinant human IL-9 R by ELISA, neutralization, and immunoblotting (predicted molecular mass 26 kDA, observed molecular mass approximately 29 kDa as a result of glycosylation).

Human Interleukin-9 (IL-9) was first identified based on its proliferative activity on a human myeloid leukemic cell line.[1] Human IL-9 stimulates the proliferation of the human megakaryocytic leukemic cell line M07e.[2] Human IL-9 supports erythroid colony formation and synergizes with IL-4 in the production of IgE and IgG.[3, 4]

The interleukin-9 receptor is a member of the hematopoietin receptor superfamily. The cDNAs encoding mouse and human IL-9 receptors have been isolated.[5, 6] The deduced mouse and human transmembrane proteins, sharing 53% amino acid sequence identity, contain 468 and 533 amino acid residues, respectively. A number of isoforms of IL-9 R, including a putative soluble form, have also been identified. The IL-9 receptor is expressed in a variety of hematopoietic cells including T cells, neutrophils, mast cells, and macrophages.[1]

Reagent

Monoclonal Anti-Human Interleukin-9 Receptor is supplied as approximately 500 µg of antiserum lyophilized from a 0.2 µm filtered solution of phosphate buffered saline (PBS) containing 5% trehalose.

Preparation Instructions

To one vial of lyophilized powder, add 1 ml of sterile phosphate buffered saline to produce a 0.5 mg/ml stock solution of antibody.

Storage/Stability

Prior to reconstitution, store at –20 °C. Reconstituted product may be stored at 2-8 °C for up to one month. For prolonged storage, freeze in working aliquots at –20 °C. Avoid repeated freezing and thawing. Do not store in frost-free freezer.

Product Profile

Anti-Human Interleukin-9 Receptor has the ability to block the cell surface human IL-9 R mediated bioactivities induced by recombinant human IL-9 response on human MO7e cells. Various concentrations of the antibody are incubated with MO7e cells for 1 hour at 37 °C in a 96 well plate. Following this pre-incubation, recombinant human IL-9 is added to the wells. The assay mixture in a total volume of 100 µl, containing antibody at concentrations of 0.01-200 µg/ml, recombinant human IL-9 at 2 ng/ml, and cells at 1 x10^5 cells/ml, is incubated at 37 °C for 72 hours in a humidified 5% CO_2 incubator. The mixture is pulsed with ^{3}H-thymidine during the final 4 hours. The cells are detached and harvested onto glass fiber filters, and the ^{3}H-thymidine incorporated into the DNA is measured.

The Neutralization Dose$_{50}$ (ND_{50}) for anti-human IL-9 R is approximately 2-4 µg/ml in the presence of 2 ng/ml of recombinant human IL-9 (Prod. No. I 3394) using human MO7e cells.

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 that cytokine is present at a concentration just high enough to elicit a maximum response.

The exact concentration of antibody required to neutralize the human cell surface IL-9 R mediated bioactivity is dependent on the IL-9 concentration, as well as on the number and types of IL-9 receptors present on the cell surface (a function of cell type and culture conditions).

For immunoblotting, a working antibody concentration of 1-2 µg/ml is recommended. The detection limit for human IL-9 receptor is approximately 25 ng/lane under non-reducing and reducing conditions.

For ELISAs, a working antibody concentration of 0.5-1.0 µg/ml is recommended. The detection limit for soluble human IL-9 receptor is approximately 6 ng/well.

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

Endotoxin level is < 0.1 EU (endotoxin units) of antibody as determined by the LAL (*Limulus* amebocyte lysate) method.

References

1. Yang, Y., Leukemia and Lymphoma, **8**, 441 (1992).
2. Yang, Y., et al., Blood, **74**, 1880 (1989).
3. Donahue, R., et al., Blood, **75**, 2271 (1990).
4. Petit-Frere, C., et al., Cytokine, **3**, 466 (1991).
5. Renauld, J., et al., J. Immunol., **144**, 4235 (1990).
6. Chang, M.S., et al., Blood, **83**, 3199-3205 (1994).

kaa 02/03