

Product Information

Monoclonal Anti-Erythropoietin

Clone 148438

produced in rat, purified immunoglobulin

Catalog Number **E1407**

Product Description

Monoclonal Anti-Erythropoietin (Epo) (rat IgG2A isotype) is purified from a hybridoma resulting from the fusion of a mouse myeloma with B cells obtained from a rat immunized with purified, CHO cell-derived, recombinant mouse erythropoietin (GenelD 13856). The IgG fraction of the tissue culture supernatant was purified by Protein G affinity chromatography.

Monoclonal Anti-Erythropoietin can be used to neutralize recombinant mouse Erythropoietin.

Erythropoietin has been cloned from various species including human, murine, canine, etc. The mature proteins from the various species are highly conserved, exhibiting greater than 80% amino acid sequence identity. Erythropoietin, a glycoprotein produced primarily by the kidney and at lower levels by the liver, is the primary regulatory factor of erythropoiesis.¹ Epo promotes the proliferation, differentiation, and survival of the erythroid progenitors. Epo stimulates erythropoiesis by inducing growth and differentiation of burst forming units and colony forming units, into mature red blood cells.² Epo produced by kidney cells is increased in response to hypoxia or anemia. The biological effects of erythropoietin are mediated by the erythropoietin receptor, which binds Epo with high affinity and is a potent Epo antagonist. When Epo is present at low concentrations, the Epo receptor initiates prolongation of G1 in the cell cycle and sends a differentiation signal; whereas at high Epo concentrations, a proliferation signal is generated and the G1 is shortened.³

Reagent

Supplied lyophilized from a 0.2 µm filtered solution of phosphate buffered saline with 5% trehalose.

Precautions and Disclaimer

This product is for R&D use only, not for drug, household, or other uses. Please consult the Material Safety Data Sheet for information regarding hazards and safe handling practices.

Preparation Instructions

To one vial of lyophilized powder, add 1 mL of 0.2 µm filtered PBS to produce a 0.5 mg/mL stock solution. If aseptic technique is used, no further filtration should be necessary for use in cell culture environments.

Storage/Stability

Prior to reconstitution, store at -20 °C. Reconstituted product may be stored at 2-8 °C for up to one month. For extended storage, freeze in working aliquots at -20 °C. Repeated freezing and thawing, or storage in "frost-free" freezers, is not recommended.

Product Profile

Neutralization: To measure the ability of the antibody to neutralize the bioactivity of recombinant mouse Epo on TF-1 cells, rmEPO was incubated with various concentrations of antibody for 1 hour at 37° C in a 96 well microplate. Following this preincubation period, TF-1 cells were added. The assay mixture, in a total volume of 100 µL/well, containing antibody at concentrations of 0.001-100 µg/mL, rmEpo at a concentration of 40 ng/mL, and cells at 1×10^5 cells/mL, was incubated for 72 hours in a 5% CO₂ humidified incubator and pulsed with ³H-thymidine for the final four hours. The cells were then harvested onto glass fiber filters and the ³H-thymidine incorporated into DNA was determined.

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.

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

Endotoxin: < 0.1 EU/µg antibody as determined by the LAL method.

References

1. Lacombe, C., and Maeux, O., *Haematologica*, **83**, 724-732 (1998).
2. Egrie, J., et al., Human Cytokines, Aggarwal, B., et al., (eds.), Blackwell Scientific Publications, Boston, 383 (1992).
3. Carroll, M., et al., *Proc. Natl. Acad. Sci. USA*, **92**, 2869-2873 (1995).

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