

Product Information

Anti-Leukemia Inhibitory Factor

produced in goat, IgG fraction of antiserum

Catalog Number **L9152**

Product Description

Anti-Leukemia Inhibitory Factor (LIF) is produced in goat using recombinant mouse leukemia inhibitory factor, expressed in *E. coli*, as the immunogen. The product is purified by Protein G affinity chromatography.

Anti-Leukemia Inhibitory Factor recognizes recombinant mouse LIF by neutralization, immunoblotting, and immunohistochemistry. The antibody will neutralize the biological activity of recombinant mouse LIF and also neutralize the biological activity of recombinant human LIF at a 10-fold higher IgG concentration. By immunoblotting, the antibody shows <10% cross-reactivity with recombinant human LIF.

Leukemia inhibitory factor is a multifunctional glycoprotein that induces macrophage differentiation and suppresses the proliferation of the murine M1 myeloid cell line.¹

Reagent

Supplied lyophilized from a 0.2 µm filtered solution in phosphate buffered saline containing 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 sterile-filtered phosphate buffered saline to produce a 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 2-8 °C for up to one month. For prolonged storage, freeze in working aliquots at -20 °C. Avoid repeated freezing and thawing.

Procedure

In this bioassay, recombinant mouse LIF was pre-incubated with various dilutions of the antibody for 1 hour at 37 °C in a 96-well plate. Then, DA-1a cells were added to each well to give a final concentration of 5×10^4 cells/mL in 0.1 mL containing 0.5 ng/mL recombinant human LIF. This was incubated for 68 hours at 37 °C in a 5% CO₂ humidified incubator. Tetrazolium salt (MTT) was added for the final 4 hours. The dark blue formazan crystals produced by active cells were then solubilized with 0.1 mL of 20% (w/v) SDS in 50% DMF (v/v), pH 4.7, and absorption at 570 nm was measured.

Product Profile

Neutralization: The antibody was tested for its ability to neutralize the bioactivity of recombinant mouse LIF in a cell proliferation assay using the murine leukemic cell line DA-1a.² The ND₅₀ of the antibody is defined as the concentration of antibody resulting in a one-half maximal inhibition of cytokine activity on a responsive cell line, when that cytokine is present at a concentration just high enough to elicit a maximum response.

Immunoblotting: a working concentration of 1-2 µg/mL is recommended. The detection limit for recombinant mouse LIF is ~20 ng/lane under both non-reducing and reducing conditions. Because this antibody preparation is a total IgG fraction, complete monospecificity cannot be assumed.

Immunohistochemistry: this antibody may be used with the appropriate secondary reagents to detect mouse LIF. Staining may be done on tissues dissected from animals that were fixed by vascular perfusion with 4% paraformaldehyde/PBS (pH 7.4) and followed by perfusion with a 10% sucrose solution in 0.1 M phosphate buffer (pH 7.2). A working dilution of 10 µg/mL of the primary antibody is recommended for 5-15 µm thick cryostat sections fixed on slides. For free-floating section, primary antibodies should be diluted to 1-2 µg/mL to reduce background staining. Staining on cells and tissues may be done by employing either fluorescent or chromogenic labels.

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

Endotoxin: <10 ng/mg of the antibody as determined by the LAL method.

References

1. Gearing, D., et al., *EMBO J.*, **6**, 3995 (1987).
2. Moreau, F.J., et al., *Nature*, **336**, 690 (1988).

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