

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**

# Anti-Erythropoietin

Developed in Rabbit IgG Fraction of Antiserum

Product No. E0271

## **Product Description**

Anti-Erythropoietin is developed in rabbit using as immunogen recombinant human erythropoietin (EPO), expressed in CHO cells. The antibody is purified by Protein A affinity chromatography. Rabbit Anti-Human EPO is provided lyophilized from phosphate buffered saline, to which no preservatives have been added.

Anti-Erythropoietin reacts with human erythropoietin by various immunochemical techniques including immunoblotting, indirect ELISA, and neutralization.

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.<sup>2</sup> 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

Anti-Erythropoietin is supplied lyophilized from a 0.2  $\mu$ m filtered solution of phosphate buffered saline.

# 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. Avoid repeated freezing and thawing.

# **Preparation Instructions**

To one vial of lyophilized powder, add 1 ml of 0.2  $\mu$ m-filtered PBS to produce a 1.0 mg/ml stock solution of Anti-Human EPO. If aseptic technique is used, no further filtration should be needed for use in cell culture environments.

### **Product Profile**

Anti-Erythropoietin is tested for its ability to neutralize the biological activity of recombinant human EPO on the human cell line TF-1. The  $ND_{50}$  of the antibody is defined as the concentration of antibody resulting in a one-half maximal inhibition of bioactivity of recombinant human EPO, when recombinant human EPO is present at a concentration just high enough to elicit a maximum response.

By indirect ELISA, a working antibody concentration of approximately 1  $\mu g/ml$  detects 3 ng/well of recombinant human EPO.

By indirect immunoblotting, a working antibody concentration of approximately 2 μg/ml detects recombinant human EPO at 100 ng/lane under both reducing and non-reducing conditions.

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

Endotoxin: <10 ng/vial by LAL method

#### References

- 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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