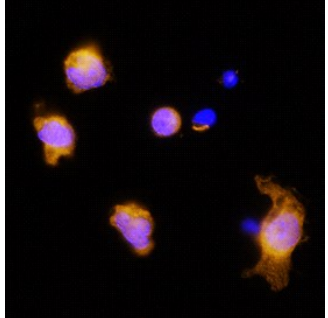
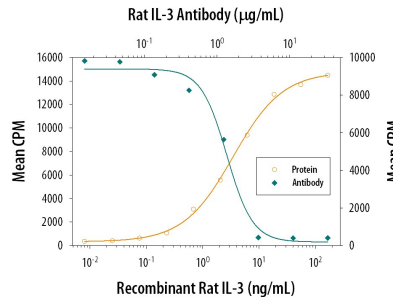


DESCRIPTION	
Species Reactivity	Rat
Specificity	Detects rat IL-3 in direct ELISAs and Western blots. In direct ELISAs and Western blots, approximately 5% cross-reactivity with recombinant mouse IL-3 and less than 1% cross-reactivity with recombinant human IL-3 is observed.
Source	Polyclonal Goat IgG
Purification	Antigen Affinity-purified
Immunogen	<i>E. coli</i> -derived recombinant rat IL-3 Ile27-Cys169 Accession # P97688
Endotoxin Level	<0.10 EU per 1 µg of the antibody by the LAL method.
Formulation	Lyophilized from a 0.2 µm filtered solution in PBS with Trehalose. See Certificate of Analysis for details. *Small pack size (-SP) is supplied as a 0.2 µm filtered solution in PBS.

APPLICATIONS		
Please Note: Optimal dilutions should be determined by each laboratory for each application. <i>General Protocols</i> are available in the <i>Technical Information</i> section on our website.		
	Recommended Concentration	Sample
Western Blot	0.1 µg/mL	Recombinant Rat IL-3 (Catalog # 2524-RL)
Immunocytochemistry	5-15 µg/mL	See Below
Neutralization	Measured by its ability to neutralize IL-3-induced proliferation in the M-NFS-60 mouse myelogenous leukemia lymphoblast cell line. Holmes, K.L. <i>et al.</i> (1985) <i>Proc. Natl. Acad. Sci. USA</i> 82 :6687. The Neutralization Dose (ND ₅₀) is typically 0.75-3 µg/mL in the presence of 0.5 ng/mL Recombinant Rat IL-3.	

DATA	
<p>Immunocytochemistry</p>  <p>IL-3 in Rat Splenocytes. IL-3 was detected in immersion fixed rat splenocytes using Goat Anti-Rat IL-3 Antigen Affinity-purified Polyclonal Antibody (Catalog # AF2524) at 15 µg/mL for 3 hours at room temperature. Cells were stained using the NorthernLights™ 557-conjugated Anti-Goat IgG Secondary Antibody (yellow, Catalog # NL001) and counterstained with DAPI (blue). Specific staining was localized to cytoplasm. View our protocol for Fluorescent ICC Staining of Non-adherent Cells.</p>	<p>Neutralization</p>  <p>Cell Proliferation Induced by IL-3 and Neutralization by Rat IL-3 Antibody. Recombinant Rat IL-3 (Catalog # 2524-RL) stimulates proliferation in the M-NFS-60 mouse myelogenous leukemia lymphoblast cell line in a dose-dependent manner (orange line). Proliferation elicited by Recombinant Rat IL-3 (0.5 ng/mL) is neutralized (green line) by increasing concentrations of Goat Anti-Rat IL-3 Antigen Affinity-purified Polyclonal Antibody (Catalog # AF2524). The ND₅₀ is typically 0.75-3 µg/mL.</p>

PREPARATION AND STORAGE	
Reconstitution	Reconstitute at 0.2 mg/mL in sterile PBS.
Shipping	The product is shipped at ambient temperature. Upon receipt, store it immediately at the temperature recommended below. *Small pack size (-SP) is shipped with polar packs. Upon receipt, store it immediately at -20 to -70 °C
Stability & Storage	Use a manual defrost freezer and avoid repeated freeze-thaw cycles. <ul style="list-style-type: none"> ● 12 months from date of receipt, -20 to -70 °C as supplied. ● 1 month, 2 to 8 °C under sterile conditions after reconstitution. ● 6 months, -20 to -70 °C under sterile conditions after reconstitution.

BACKGROUND

Rat interleukin-3 (IL-3; also multi-CSF) is a 26 kDa, variably glycosylated monomeric polypeptide that belongs to the α -helix family of hematopoietic cytokines (1, 2). IL-3 has pleiotrophic activities on a number of hematopoietic-related cells (1, 3). The rat molecule has two alternate splice forms. The first is termed IL-3 β and is synthesized as a 169 amino acid (aa) precursor that contains a 27 aa signal sequence and a 142 aa mature segment (1, 2). The second is called IL-3 α , and is identical to IL-3 β , save for a three amino acid (Tyr-Pro-Gln) deletion at positions 56-58 (1). The beta form is considered the most common form. Each form has an α -helical structure with two intrachain disulfide bonds and two potential N-linked glycosylation sites. Rat IL-3 is generally considered to be species-specific in its activity. In the mature region, rat IL-3 shares 55%, 30%, and 24% aa sequence identity with mouse, human, and bovine IL-3, respectively. Cells known to express IL-3 include connective tissue mast cells, astrocytes, microglia, megakaryocytes, eosinophils, T cells, keratinocytes and thymic epithelium.

IL-3 exerts its biological activities by binding to a 70 kDa, low affinity, ligand-binding IL-3 R α subunit, (6) which then recruits a 120 kDa, common β -chain, signal transducing subunit (7) to form a functional IL-3 receptor (1, 6, 7). Receptors for IL-3 are present on bone marrow progenitors, macrophages, mast cells, eosinophils, megakaryocytes, basophils, and various myeloid leukemic cells. IL-3 can stimulate the proliferation and differentiation of pluripotent hematopoietic stem cells as well as various lineage committed progenitors including those for neutrophils, macrophages, megakaryocytes, and erythroid cells. IL-3 can stimulate the growth of early B cells and mature macrophages, mast cells, eosinophils, basophils, and megakaryocytes. IL-3 augments the function activity of basophils, mast cells, eosinophils, and macrophages (1, 8). In combination with other molecules such as CD40L and/or IL-4, IL-3 can stimulate production of dendritic cells (1, 2, 9, 10).

References:

1. Martinez-Moczygemba, M. and D.P. Huston (2003) *J. Allergy Clin. Immunol.* **112**:653.
2. Mangi, M.H. and A.C. Newland (1999) *Cytokines Cell. Mol. Ther.* **5**:87.
3. Esandi, M. del C. *et al.* (1998) *Gene* **211**:151.
4. Cohen, D.R. *et al.* (1986) *Nucleic Acids Res.* **14**:3641.
5. Gebicke-Haerter, P.J. *et al.* (1994) *J. Neuroimmunol.* **50**:203.
6. Chritton, S.C. and M.H. Sheng (1997) GenBank Accession # NP_640353.
7. Appel, K. *et al.* (1995) *J. Neurosci.* **15**:5800.
8. Schrader, J.W. (2001) in *Cytokine Reference*, Oppenheim, J.J. and M. Feldmann (eds): Academic Press, p. 855.
9. Ebner, S. *et al.* (2002) *J. Immunol.* **168**:6199.
10. Buelens C. *et al.* (2002) *Blood* **99**:993.