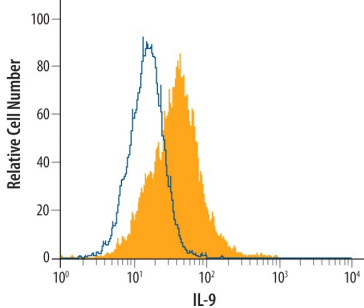
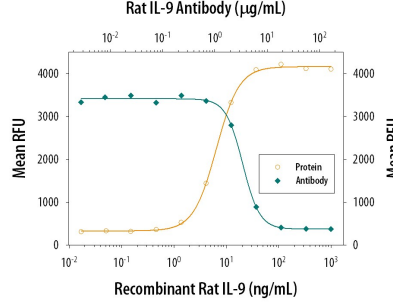


| DESCRIPTION               |  |
|---------------------------|--|
| <b>Species Reactivity</b> | Rat  |
| <b>Specificity</b>        | Detects rat IL-9 in direct ELISAs and Western blots. In direct ELISAs, approximately 30% cross-reactivity with recombinant mouse IL-9 is observed, and approximately 10% cross-reactivity with recombinant human IL-9 is observed. |
| <b>Source</b>             | Polyclonal Sheep IgG   |
| <b>Purification</b>       | Antigen Affinity-purified  |
| <b>Immunogen</b>          | Mouse myeloma cell line NS0-derived recombinant rat IL-9<br>Gln19-Ala144<br>Accession # NP_001099217   |
| <b>Endotoxin Level</b>    | <0.10 EU per 1 µg of the antibody by the LAL method.   |
| <b>Formulation</b>        | Lyophilized from a 0.2 µm filtered solution in PBS with Trehalose. See Certificate of Analysis for details.<br>*Small pack size (-SP) is supplied as a 0.2 µm filtered solution in PBS.  |

| APPLICATIONS   |  |
|--|--|
| <b>Please Note:</b> 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. |  |
|  | <b>Recommended Concentration      Sample</b>   |
| <b>Western Blot</b>  | 0.1 µg/mL      Recombinant Rat IL-9 (Catalog # 4134-RL)  |
| <b>Intracellular Staining by Flow Cytometry</b>  | 2.5 µg/10 <sup>6</sup> cells      See Below  |
| <b>CyTOF-ready</b>   | Ready to be labeled using established conjugation methods. No BSA or other carrier proteins that could interfere with conjugation.   |
| <b>Neutralization</b>  | Measured by its ability to neutralize IL-9-induced proliferation in the TS1 mouse helper T cell line. Moreau, F. J. <i>et al.</i> (1988) <i>Nature</i> <b>336</b> :690. The Neutralization Dose (ND <sub>50</sub> ) is typically 1.5-7.5 µg/mL in the presence of 10 ng/mL Recombinant Rat IL-9. |

| DATA   |  |
|--|--|
| <p><b>Intracellular Staining by Flow Cytometry</b></p>  <p><b>Detection of IL-9 in Rat Splenocytes by Flow Cytometry.</b> Rat splenocytes were treated for 24 hours with 50 ng/mL PMA and 500 ng/mL Ca<sup>2+</sup> ionomycin then stained with Sheep Anti-Rat IL-9 Antigen Affinity-purified Polyclonal Antibody (Catalog # AF4134, filled histogram) or control antibody (Catalog # 5-001-A, open histogram), followed by NorthernLights™ 637-conjugated Anti-Sheep IgG Secondary Antibody (Catalog # NL011). To facilitate intracellular staining, cells were fixed with paraformaldehyde and permeabilized with saponin.</p> | <p><b>Neutralization</b></p>  <p><b>Cell Proliferation Induced by IL-9 and Neutralization by Rat IL-9 Antibody.</b> Recombinant Rat IL-9 (Catalog # 4134-RL) stimulates proliferation in the TS1 mouse helper T cell line in a dose-dependent manner (orange line). Proliferation elicited by Recombinant Rat IL-9 (10 ng/mL) is neutralized (green line) by increasing concentrations of Sheep Anti-Rat IL-9 Antigen Affinity-purified Polyclonal Antibody (Catalog # AF4134). The ND<sub>50</sub> is typically 1.5-7.5 µg/mL.</p> |

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

**BACKGROUND**

Rat interleukin-9 (IL-9; also T-cell growth factor P40 and MEA) is a 20-30 kDa secreted glycoprotein that belongs to the IL-7/IL-9 family of hematopoietic cytokines. It is best considered an atypical Th2-type cytokine (1-4). Rat IL-9 was initially isolated from the 4437A rat thymoma cell line (5). The IL-9 precursor is 144 amino acids (aa) in length and contains an 18 aa signal sequence plus a 126 aa mature segment (5). An extended, 219 aa alternate precursor form is reported in GenBank (Accession # XP\_001068265). It shows a 113 aa substitution for the first 38 aa of the standard form. Its significance is unknown. Mature rat IL-9 is 75%, 74% and 58% aa identical to cotton rat, mouse and human IL-9, respectively (6-9). There is variability in cross-species reactivity. Canine and mouse IL-9 are purportedly active on human cells (9-11), while human IL-9 is inactive on mouse cells (9). Cells reported to express IL-9 include activated CD4+ T cells and eosinophils (12, 13). The receptor for rat IL-9 is composed of the 62 kDa rat IL-9Ra chain and the 64 kDa rat common cytokine receptor  $\gamma$ -chain (5, 14, 15). IL-9 is suggested to play a significant, but likely supporting role in allergy and asthma. Its synthesis is induced upon exposure to allergen (10, 13). Following release, it is known to upregulate IgE production by IL-4-stimulated B cells (16). It also stimulates mucin secretion and IL-13 production through a direct action on respiratory epithelium (10, 17). In the bone marrow, it promotes eosinophilia, likely through IL-5 induction (18, 19). And once produced, eosinophils are chemoattracted to the bronchi through the IL-9-induced secretion of eotaxin-1 by airway smooth muscle cells (20).

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