

Technical Data Sheet

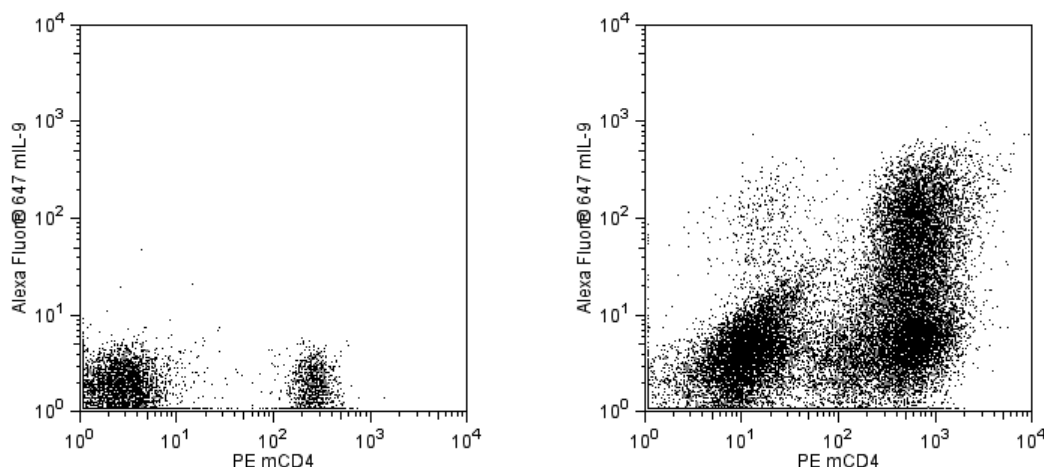
Alexa Fluor® 647 Armenian Hamster anti-Mouse IL-9

Product Information

Material Number:	561464
Alternate Name:	IL-9; Interleukin-9; MEA; P40; T-cell growth factor P40; TCGF III
Size:	0.1 mg
Concentration:	0.2 mg/ml
Clone:	D9302C12
Immunogen:	Mouse IL-9 Recombinant Protein
Isotype:	Armenian Hamster IgG2, κ
Reactivity:	QC Testing: Mouse
Storage Buffer:	Aqueous buffered solution containing $\leq 0.09\%$ sodium azide.

Description

The D9302C12 monoclonal antibody specifically binds to the multifunctional mouse cytokine, Interleukin-9 (IL-9). IL-9 is a 126 amino acid-long glycoprotein that is produced by various subsets of activated CD4⁺ T cells. IL-9 acts on target cells by binding to and signaling through the heterodimeric IL-9 receptor (IL-9R) complex that is comprised of transmembrane IL-9 receptor alpha (IL-9R α) and common gamma chain (γ c) subunits. IL-9 can promote the survival, growth, proliferation and/or differentiation of various cell types including thymocytes, T cells, B cells, mast cells, and hematopoietic progenitor cells. IL-9 can augment IL-4-induced IgE and IgG1 production from lipopolysaccharide-primed mouse B cells and induce granzyme and high-affinity IgE receptor gene expression by mouse T helper cell clones and mast cell lines. IL-9 plays an important role *in vivo* in helminth elimination. The D9302C12 antibody neutralizes mouse IL-9 bioactivity.



Multicolor flow cytometric analysis of IL-9 expression by unstimulated and activated mouse spleen cells. Mouse spleen cells were either unstimulated (Left Panel) or stimulated in a tissue culture plate coated with Anti-Mouse CD3 ϵ and soluble Anti-Mouse CD28 antibodies along with Recombinant Mouse IL-2, IL-4, and TGF- β proteins and Anti-Mouse IFN- γ antibody for 4 days. On day 4 the cells were harvested and restimulated with Phorbol 12-Myristate 13-Acetate (PMA; Sigma P-8139) plus Ionomycin (Sigma; I-0634) in the presence of BD GolgiStop™ Protein Transport Inhibitor for 5 hours (Right Panel). The cells were then fixed and permeabilized using BD Cytofix/Cytoperm™ Fixation/Permeabilization Solution Kit followed by staining with Alexa Fluor® 647 Armenian Hamster anti-Mouse IL-9 (Cat. No. 561463) and PE Rat Anti-Mouse CD4 (Cat. No. 553048). Two-color flow cytometric dot plots showing the correlated expression patterns of CD4 versus IL-9 were derived from gated events with the forward and side light-scatter characteristics of intact lymphocytes. Flow cytometry was performed using a BD™ LSR II Flow Cytometer System. Other compatible fixation and permeabilization treatments are listed in the Suggested Companion Products.

BD Biosciences

bdbiosciences.com

United States	Canada	Europe	Japan	Asia Pacific	Latin America/Caribbean
877.232.8995	888.268.5430	32.53.720.550	0120.8555.90	65.6861.0633	0800.771.7157

For country-specific contact information, visit bdbiosciences.com/how_to_order/

Conditions: The information disclosed herein is not to be construed as a recommendation to use the above product in violation of any patents. BD Biosciences will not be held responsible for patent infringement or other violations that may occur with the use of our products. Purchase does not include or carry any right to resell or transfer this product either as a stand-alone product or as a component of another product. Any use of this product other than the permitted use without the express written authorization of Becton Dickinson and Company is strictly prohibited.

For Research Use Only. Not for use in diagnostic or therapeutic procedures. Not for resale.

BD, BD Logo and all other trademarks are the property of Becton, Dickinson and Company. ©2011 BD



Preparation and Storage

The monoclonal antibody was purified from tissue culture supernatant or ascites by affinity chromatography.

The antibody was conjugated to Alexa Fluor® 647 under optimum conditions, and unreacted Alexa Fluor® 647 was removed.

Store undiluted at 4°C and protected from prolonged exposure to light. Do not freeze.

Application Notes

Application

Intracellular staining (flow cytometry)	Routinely Tested
---	------------------

Recommended Assay Procedure:

This fluorescent antibody is suitable for intracellular staining of mouse leukocytes using BD Cytotfix/Cytoperm™ Reagents or BD Phosflow™ Fix Buffer I and Perm/Wash Buffer I (please see *Suggested Companion Products*).

Suggested Companion Products

Catalog Number	Name	Size	Clone
554714	BD Cytotfix/Cytoperm™ Fixation/Permeabilization Kit	250 tests	(none)
554724	Protein Transport Inhibitor (Containing Monensin)	0.7 ml	(none)
554722	Fixation and Permeabilization Solution	125 ml	(none)
554655	Fixation Buffer	100 ml	(none)
554723	Perm/Wash Buffer	100 ml	(none)
554656	Stain Buffer (FBS)	500 ml	(none)
557870	Fix Buffer I	250 ml	(none)
557885	Perm/Wash Buffer I	125 ml	(none)
553048	PE Rat Anti-Mouse CD4	0.1 mg	RM4-5
553057	Purified NA/LE Hamster Anti-Mouse CD3e	0.5 mg	145-2C11
553294	Purified NA/LE Hamster Anti-Mouse CD28	0.5 mg	37.51
550069	Recombinant Mouse IL-2	20 µg	(none)
550067	Recombinant Mouse IL-4	10 µg	(none)
356039	Transforming Growth Factor-β (TGF-β), human natural, 1 X 5 µg	NA	(none)
554408	Purified NA/LE Rat Anti-Mouse IFN-γ	0.5 mg	XMGI.2

Product Notices

1. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
2. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.
3. Alexa Fluor® 647 fluorochrome emission is collected at the same instrument settings as for allophycocyanin (APC).
4. For fluorochrome spectra and suitable instrument settings, please refer to our Fluorochrome Web Page at www.bdbiosciences.com/colors.
5. The Alexa Fluor®, Pacific Blue™, and Cascade Blue® dye antibody conjugates in this product are sold under license from Molecular Probes, Inc. for research use only, excluding use in combination with microarrays, or as analyte specific reagents. The Alexa Fluor® dyes (except for Alexa Fluor® 430), Pacific Blue™ dye, and Cascade Blue® dye are covered by pending and issued patents.
6. Caution: Sodium azide yields highly toxic hydrazoic acid under acidic conditions. Dilute azide compounds in running water before discarding to avoid accumulation of potentially explosive deposits in plumbing.
7. Alexa Fluor® is a registered trademark of Molecular Probes, Inc., Eugene, OR.

References

Faulkner H, Humphreys N, Renauld JC, Van Snick J, Grecis R. Interleukin-9 is involved in host protective immunity to intestinal nematode infection. *Eur J Immunol.* 1997; 27(10):2536-2540. (Biology)

Hultner L, Druet C, Moeller J, et al. Mast cell growth-enhancing activity (MEA) is structurally related and functionally identical to the novel mouse T cell growth factor P40/TCGFIII (interleukin 9). *Eur J Immunol.* 1990; 20(6):1413-1416. (Biology)

Louahed J, Kermouni A, Van Snick J, Renauld JC. IL-9 induces expression of granzymes and high-affinity IgE receptor in murine T helper clones. *J Immunol.* 1995; 154(10):5061-5070. (Biology)

Petit-Frere C, Dugas B, Braquet P, Mencia-Huerta JM. Interleukin-9 potentiates the interleukin-4-induced IgE and IgG1 release from murine B lymphocytes. *Immunology.* 1993; 79(1):146-151. (Biology)

Renauld JC, Kermouni A, Vink A, Louahed J, Van Snick J. Interleukin-9 and its receptor: involvement in mast cell differentiation and T cell oncogenesis. *J Leukoc Biol.* 1995; 57(3):353-360. (Biology)

Suda T, Murray R, Fischer M, Yokota T, Zlotnik A. Tumor necrosis factor-α and P40 induce day 15 murine fetal thymocyte proliferation in combination with IL-2. *J Immunol.* 1990; 144(5):1783-1787. (Biology)

Van Snick J, Cayphas S, Vink A, Uytendhove C, Coulie PG, Rubira MR, Simpson RJ. Purification and NH2-terminal amino acid sequence of a T-cell-derived lymphokine with growth factor activity for B-cell hybridomas. *Proc Natl Acad Sci U S A.* 1986; 83(24):9679-9683. (Biology)

Van Snick J, Goethals A, Renauld JC, et al. Cloning and characterization of a cDNA for a new mouse T cell growth factor (P40). *J Exp Med.* 1989; 169(1):363-368. (Biology)