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#37495

# Mouse Immune Cell Phenotyping IHC Antibody Sampler Kit



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**For Research Use Only. Not For Use In Diagnostic Procedures.**

Products Included	Product #	Quantity	Isotype/Source
CD3-epsilon (D4V8L) Rabbit mAb	99940	20 µl	Rabbit IgG
CD4 (D7D2Z) Rabbit mAb	25229	20 µl	Rabbit IgG
CD8-α (D4W2Z) XP® Rabbit mAb (Mouse Specific)	98941	20 µl	Rabbit IgG
FoxP3 (D6O8R) Rabbit mAb	12653	20 µl	Rabbit IgG
F4/80 (D2S9R) XP® Rabbit mAb	70076	20 µl	Rabbit IgG
CD19 (D4V4B) XP® Rabbit mAb	90176	20 µl	Rabbit IgG
CD11c (D1V9Y) Rabbit mAb	97585	20 µl	Rabbit IgG
Granzyme B (D6E9W) Rabbit mAb	46890	20 µl	Rabbit IgG

See [www.cellsignal.com](http://www.cellsignal.com) for individual component applications, species cross-reactivity, dilutions, and additional application protocols.

**Description:** The Mouse Immune Cell Phenotyping IHC Antibody Sampler Kit provides an economical means of detecting the accumulation of immune cell types in formalin-fixed, paraffin-embedded tissue samples.

**Background:** Cluster of Differentiation 3 (CD3) is a multiunit protein complex expressed on the surface of T-cells that directly associates with the T-cell receptor (TCR). CD3 is composed of four polypeptides: ζ, γ, ε and δ. Engagement of TCR complex with antigens presented in Major Histocompatibility Complexes (MHC) induces tyrosine phosphorylation in the immunoreceptor tyrosine-based activation motif (ITAM) of CD3 proteins. CD3 phosphorylation is required for downstream signaling through ZAP-70 and p85 subunit of PI-3 kinase, leading to T cell activation, proliferation, and effector functions (1). Cluster of Differentiation 8 (CD8) is a transmembrane glycoprotein expressed primarily on cytotoxic T cells, but has also been described on a subset of dendritic cells in mice (2,3). On T cells, CD8 is a co-receptor for the TCR, and these two distinct structures are required to recognize antigen bound to MHC Class I (2). Cluster of Differentiation 4 (CD4) is expressed on the surface of T helper cells, regulatory T cells, monocytes, macrophages, and dendritic cells, and plays an important role in the development and activation of T cells. On T cells, CD4 is the co-receptor for the TCR, and these two distinct structures recognize antigen bound to MHC Class II. CD8 and CD4 co-receptors ensure specificity of the TCR-antigen interaction, prolong the contact between the T cell and the antigen presenting cell, and recruit the tyrosine kinase Lck, which is essential for T cell activation (2). Granzyme B is a serine protease expressed by CD8<sup>+</sup> cytotoxic T lymphocytes and natural killer (NK) cells and is a key component of the

immune response to pathogens and transformed cancer cells (4). Forkhead box P3 (FoxP3) is crucial for the development of T cells with immunosuppressive regulatory properties and is a well-established marker for T regulatory cells (Tregs) (5). CD19 is a co-receptor expressed on B cells that amplifies the signaling cascade initiated by the B cell receptor (BCR) to induce activation. It is a biomarker of B lymphocyte development, lymphoma diagnosis, and can be utilized as a target for leukemia immunotherapies (6,7). F4/80 (EMR1) is a heavily glycosylated G-protein-coupled receptor and is a well-established marker for mouse macrophages (8). CD11c (integrin αX, ITGAX) is a transmembrane glycoprotein highly expressed by dendritic cells, and has also been observed on activated NK cells, subsets of B and T cells, monocytes, granulocytes, and some B cell malignancies including hairy cell leukemia (9,10).

**Specificity/Sensitivity:** Each antibody included in the Mouse Immune Cell Phenotyping IHC Antibody Sampler Kit recognizes endogenous levels of its target mouse protein. Non-specific staining has been observed in mouse pancreas with CD3ε (D4V8L) Rabbit mAb, in mouse kidney and liver with CD4 (D7D2Z) Rabbit mAb, and in the sweat glands with Granzyme B (D6E9W) Rabbit mAb.

**Source/Purification:** Monoclonal antibodies are produced by immunizing animals with synthetic peptides corresponding to residues surrounding Val31 of mouse CD3ε protein, Asp42 of mouse CD8α protein, Ala232 of mouse CD4 protein, Pro44 of mouse FoxP3 protein, Leu427 of human CD19 protein, Ala1153 of mouse CD11c protein, or with recombinant proteins specific to human Granzyme B protein or mouse F4/80 protein.

**Storage:** Supplied in 10 mM sodium HEPES (pH 7.5), 150 mM NaCl, 100 µg/ml BSA, 50% glycerol and less than 0.02% sodium azide. Store at -20°C. Do not aliquot the antibody.

#### Background References:

- (1) Kuhns, M.S. et al. (2006) *Immunity* 24, 133-9.
- (2) Zamoyska, R. (1994) *Immunity* 1, 243-6.
- (3) Shortman, K. and Heath, W.R. (2010) *Immunol Rev* 234, 18-31.
- (4) Trapani, J.A. (2001) *Genome Biol* 2, REVIEWS3014.
- (5) Ochs, H.D. et al. (2007) *Immunol Res* 38, 112-21.
- (6) Tedder, T.F. et al. (1997) *Immunity* 6, 107-18.
- (7) Scheuermann, R.H. and Racila, E. (1995) *Leuk Lymphoma* 18, 385-97.
- (8) McKnight, A.J. et al. (1996) *J Biol Chem* 271, 486-9.
- (9) Kohrgruber, N. et al. (1999) *J Immunol* 163, 3250-9.
- (10) Qualai, J. et al. (2016) *PLoS One* 11, e0154253.

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**Applications:** W—Western IP—Immunoprecipitation IHC—Immunohistochemistry ChIP—Chromatin Immunoprecipitation IF—Immunofluorescence F—Flow cytometry E-P—ELISA-Peptide **Species Cross-Reactivity:** H—human M—mouse R—rat Hm—hamster Mk—monkey Mi—mink C—chicken Dm—D. melanogaster X—Xenopus Z—zebrafish B—bovine Dg—dog Pg—pig Sc—S. cerevisiae Ce—C. elegans Hr—Horse All—all species expected **Species enclosed in parentheses are predicted to react based on 100% homology.**