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#81490

Human Exhausted T Cell Antibody Sampler Kit



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New 01/21

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

Products Included	Product #	Quantity	Mol. Wt.	Isotype/Source
Tox/Tox2 (E6G50) Rabbit mAb	36778	20 µl	60-80 kDa	Rabbit IgG
TCF1/TCF7 (C63D9) Rabbit mAb	2203	20 µl	48, 50 kDa	Rabbit IgG
EOMES (D8D1R) Rabbit mAb	81493	20 µl	75, 85 kDa	Rabbit IgG
PD-1 (D4W2J) XP® Rabbit mAb	86163	20 µl	52-65 kDa	Rabbit IgG
CTLA-4 (E1V6T) Rabbit mAb	96399	20 µl	25-30 kDa	Rabbit IgG
TIGIT (E5Y1W) XP® Rabbit mAb	99567	20 µl	18, 30-40 kDa	Rabbit IgG
LAG3 (D2G40™) XP® Rabbit mAb	15372	20 µl	60-80 kDa	Rabbit IgG
TIM-3 (D5D5R™) XP® Rabbit mAb	45208	20 µl	45-70 kDa	Rabbit IgG
2B4/SLAMF4/CD244 (D5J9D) Rabbit mAb	54560	20 µl	70-120 kDa	Rabbit IgG
Anti-rabbit IgG, HRP-linked Antibody	7074	100 µl		Goat

See www.cellsignal.com for individual component applications, species cross-reactivity, dilutions and additional application protocols.

Description: The Human Exhausted T Cell Antibody Sampler Kit provides an economical means of characterizing the extent of exhaustion in T cells. This kit includes enough antibodies to perform two western blot experiments with each primary antibody.

Background: Tox, TCF1/TCF7, and EOMES play key roles in T cell development. Tox is also induced by high antigen stimulation during chronic viral infection or cancer, regulating T cell persistence and exhaustion. TCF1/TCF7 preserves the effector function of exhausted T cells during viral infection or cancer. EOMES is a key transcription factor for memory T cells and for full effector differentiation of CD8⁺ T cells. Expression of EOMES is induced in CD8⁺ T cells following viral infection and bacterial infection, and high levels of EOMES promotes T cell exhaustion. The dynamic expression of these transcription factors help characterize the extent to which a T cell is exhausted and will respond to antigen stimulation (1-5)

PD-1 (PDCD1, CD279), CTLA-4 (CD152), TIGIT (VSI9G, VSTM3), TIM-3 (HAVCR2), LAG3 (CD223), and 2B4 (SLAMF4, CD244) are immune cell co-inhibitory receptors (also known as immune checkpoints) that negatively regulate T cell function and dampen the immune response to pathogens and cancer. In addition to activated T cells, PD-1 is expressed by activated B cells and monocytes. Following interaction with its ligands, PD-L1 and PD-L2, PD-1 is phosphorylated at ITIM and ITSM motifs leading to recruitment of protein tyrosine phosphatases SHP-1 and SHP-2 and suppression of TCR signaling. CTLA-4 protein is primarily expressed on T cells, including CD8⁺ cytotoxic T cells, CD4⁺ helper T cells, and CD4⁺/FoxP3⁺ regulatory T cells. CTLA-4 protein competes with CD28 for B7.1 (CD80) and B7.2 (CD86) binding at the cell surface, resulting in downregulation of T cell activity. TIGIT is expressed at low levels on subsets of T cells and natural killer (NK) cells, and is upregulated at the protein level following activation of these cells. TIGIT marks exhausted T cells in the tumor microenvironment and during human immunodeficiency virus (HIV) infection. TIM-3 is ex-

pressed by exhausted T cells in the settings of chronic infection and cancer. Tumor-infiltrating macrophages and dendritic cells also express TIM-3. LAG3 is primarily expressed by activated CD4⁺ T cells, CD8⁺ T cells, FoxP3⁺ T regulatory cells (Tregs), and NK cells. 2B4 is a heterophilic cell surface receptor expressed on a variety of immune cells, including NK cells, T cells, eosinophils, mast cells, and dendritic cells. 2B4 has been shown to have both immune stimulatory and inhibitory effects on cells. Co-expression of multiple immune checkpoints help characterize the extent to which a T cell is exhausted and will respond to antigen stimulation. Therapeutic blockade of several of these immune checkpoint receptors is a promising strategy for neoplastic intervention by enabling anti-tumor immune responses (6-13).

Specificity/Sensitivity: Each antibody in the Human Exhausted T Cell Antibody Sampler Kit detects endogenous levels of its target human protein. Tox/Tox2 (E6G50) Rabbit mAb does not cross-react with Tox3 or Tox4 proteins, but does cross-react with an unidentified protein of 50 kDa in some murine cell extracts. TCF1/TCF7 (C63D9) Rabbit mAb does not recognize the dominant negative isoforms of TCF1/TCF7 lacking the amino-terminal β-catenin binding domain and does not cross-react with LEF1. TIGIT (E5Y1W) XP® Rabbit mAb cross-reacts with an unidentified protein of 42 kDa in some cell extracts. 2B4/SLAMF4/CD244 (D5J9D) Rabbit mAb cross-reacts with an unidentified protein of 18 kDa in some cell extracts.

Source/Purification: Monoclonal antibodies are produced by immunizing animals with a synthetic peptide corresponding to residues surrounding Gln435 of human Tox protein, Pro96 of human TCF1/TCF7 protein, Pro180 of human EOMES protein, Ala274 of human PD-1 protein, Leu95 of human CTLA-4 protein, and Asp367 of human 2B4/SLAMF4/CD244 protein. Monoclonal antibodies are produced by immunizing animals with a recombinant protein specific to the carboxy terminus of human TIGIT protein, the extracellular domain of human TIM-3 protein, and the amino terminus of human LAG3 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 antibodies.

Please visit www.cellsignal.com for validation data and a complete listing of recommended companion products.

Background References:

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- (2) Yao, C. et al. (2019) *Nat Immunol* 20, 890-901.
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- (4) Wang, Y. et al. (2019) *Front Immunol* 10, 169.
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- (6) Schildberg, F.A. et al. (2016) *Immunity* 44, 955-72.
- (7) Anderson, A.C. et al. (2016) *Immunity* 44, 989-1004.
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U.S. Patent No. 7,429,487, foreign equivalents, and child patents deriving therefrom.

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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.**