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#25520

Histone H3 Lysine Mutant-Specific Antibody Sampler Kit



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New 10/20

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

Products Included	Product #	Quantity	Mol. Wt.	Isotype/Source
Histone H3 (K9M Mutant Specific) (E2E9L) Rabbit mAb	70414	20 µl	17 kDa	Rabbit IgG
Histone H3 (K27M Mutant Specific) (D3B5T) Rabbit mAb	74829	20 µl	17 kDa	Rabbit IgG
Histone H3 (K36M Mutant Specific) Antibody	26218	20 µl	17 kDa	Rabbit
Histone H3 (D1H2) XP® Rabbit mAb	4499	20 µl	17 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 Histone H3 Lysine Mutant-Specific Antibody Sampler Kit provides an economical means of detecting Lys (K) to Meth (M) cancer driver mutations in histone H3. The kit includes enough antibodies to perform two western blot experiments with each primary antibody.

Background: Multiple exome sequencing analyses have uncovered a high frequency of histone H3 driver mutations in a number of different cancers, including diffuse intrinsic pontine glioma (DIPG), chondroblastoma, sarcomas, and HPV-negative head and neck squamous cell carcinoma. Previous studies have shown that lysine to methionine histone mutations in these cancers act as potent inhibitors of their respective lysine methyltransferases, resulting in gross alterations to the histone methylation landscape and deregulation of gene expression. In DIPG for example, the histone H3 K27M mutation is accompanied by a dramatic reduction in the levels of polycomb repressive complex 2 (PRC2)-mediated tri-methylation of histone H3 lysine 27, changes in the distribution of PRC2 on the genome, and altered expression of genes associated with various cancer pathways (1-3). In chondrocytomas, the histone H3 K36M mutation functions to inhibit the WHSC1 (MMSET) and SETD2 histone methyltransferases, resulting in a reduction in the levels of histone H3 lysine 36 tri-methylation and deregulation of a number of cancer-associated genes (4). Similar to the H3K27M and H3K36M mutations, the histone H3 K9M mutation has been shown to inhibit the H3K9-directed histone methyltransferase G9a, resulting in reduced levels of histone H3 lysine 9 trimethylation (5). Given the widespread role of G9a in the regulation of gene expression, it is likely that this K9M mutation also plays a role in cancer.

Specificity/Sensitivity: Histone H3 (K9M Mutant Specific) (E2E9L) Rabbit mAb recognizes endogenous levels of K9M mutant histone H3.1, H3.2, and H3.3 proteins. This antibody does not cross-react with wild-type histone H3.1, H3.2, or H3.3 proteins. Histone H3 (K27M Mutant Specific) (D3B5T) Rabbit mAb recognizes endogenous levels of K27M mutant histone H3.1, H3.2, and H3.3 proteins. This antibody may cross-react with wild-type histone H3.1, H3.2, and H3.3 when used at a high concentration. Careful titration of this antibody may be required to obtain optimal specificity. Histone H3 (K36M Mutant Specific) Antibody recognizes endogenous levels of K36M mutant histone H3.1, H3.2, and H3.3 proteins. This antibody may show slight cross-reactivity with wild-type histone H3.1, H3.2, or H3.3 when used at a high concentration. Careful titration of this antibody may be required to obtain optimal specificity. Histone H3 (D1H2) XP® Rabbit mAb detects endogenous levels of total Histone H3 protein, including isoforms H3.1, H3.2, and H3.3. This antibody also detects the Histone H3 variant CENP-A. This antibody does not cross-react with other core histones.

Source/Purification: Mutant-specific histone H3 monoclonal antibodies were produced by immunizing animals with synthetic peptides corresponding to K9M, K27M, or K36M mutant sequences of human histone H3.3 protein. Total histone H3 monoclonal antibody was produced by immunizing animals with a synthetic peptide corresponding to the carboxy terminus of human histone H3 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:

- (1) Chan, K.M. et al. (2013) *Genes Dev* 27, 985-90.
- (2) Lewis, P.W. et al. (2013) *Science* 340, 857-61.
- (3) Piunti, A. et al. (2017) *Nat Med* 23, 493-500.
- (4) Fang, D. et al. (2016) *Science* 352, 1344-8.
- (5) Jayaram, H. et al. (2016) *Proc Natl Acad Sci U S A* 113, 6182-7.

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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 S—S. cerevisiae Ce—C. elegans Hr—Horse All—all species expected **Species enclosed in parentheses are predicted to react based on 100% homology.**