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Phospho-HDAC4 (Ser632)/HDAC5 (Ser661)/HDAC7 (Ser486) Antibody



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rev. 01/11/16

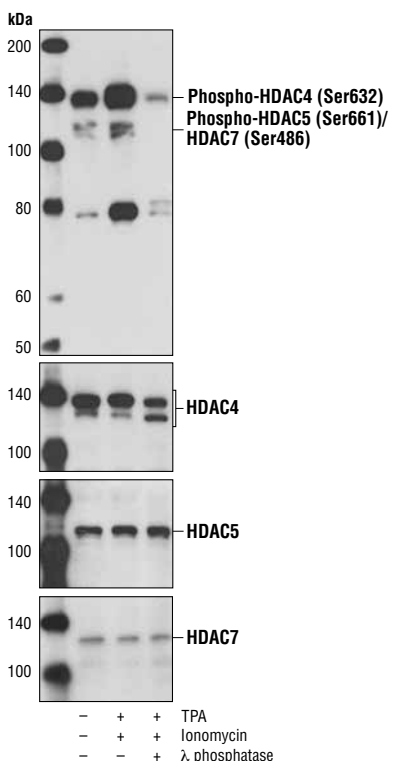
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Applications W, IP Endogenous	Species Cross-Reactivity*		Molecular Wt. 140, 124, 120 kDa	Source Rabbit**
	H	M		

Background: Acetylation of the histone tail causes chromatin to adopt an "open" conformation, allowing increased accessibility of transcription factors to DNA. The identification of histone acetyltransferases (HATs) and their large multiprotein complexes has yielded important insights into how these enzymes regulate transcription (1,2). HAT complexes interact with sequence-specific activator proteins to target specific genes. In addition to histones, HATs can acetylate nonhistone proteins, suggesting multiple roles for these enzymes (3). In contrast, histone deacetylation promotes a "closed" chromatin conformation and typically leads to repression of gene activity (4). Mammalian histone deacetylases can be divided into three classes on the basis of their similarity to various yeast deacetylases (5). Class I proteins (HDACs 1, 2, 3, and 8) are related to the yeast Rpd3-like proteins, those in class II (HDACs 4, 5, 6, 7, 9, and 10) are related to yeast Hda1-like proteins, and class III proteins are related to the yeast protein Sir2. Inhibitors of HDAC activity are now being explored as potential therapeutic cancer agents (6,7).

Histone deacetylases (HDACs) interact with an increasing number of transcription factors, including myocyte enhancer factor 2 (MEF2), to negatively regulate gene expression. HDACs are regulated in part by shuttling between the nucleus and cytoplasm, where export to the cytoplasm facilitates gene activation by removing HDACs from their target genes (8,9). The cytoplasmic export is facilitated by 14-3-3 proteins, which bind to specific phospho-serine residues on the HDAC proteins (8,9). These phospho-serine 14-3-3 binding modules are highly conserved between HDAC proteins, allowing for their collective regulation in response to specific cell stimuli. For example, the highly conserved HDAC 4 Ser632, HDAC 5 Ser498 and HDAC 7 Ser486 residues are all phosphorylated by CAMK and PKD kinases in response to multiple cell stimuli, including VEGF-induced angiogenesis in endothelial cells, B cell and T cell activation, and differentiation of myoblasts into muscle fiber (10-14).

Specificity/Sensitivity: Phospho-HDAC4 (Ser632)/HDAC5 (Ser661)/HDAC7 (Ser486) Antibody detects endogenous levels of HDAC4, HDAC5 and HDAC7 proteins only when phosphorylated on Ser632, Ser661 and Ser486, respectively. The antibody also crossreacts with an unidentified protein at 80 kDa.



Western blot analysis of extracts from DO11.10 thymocyte hybridoma cells, untreated (-) or treated with TPA (12-O-Tetradecanoylphorbol-13-Acetate) #4174 (0.2 μM, 1 hr; +) and Ionomycin, Calcium Salt #9995 (0.33 μM, 1 hr; +), using Phospho-HDAC4 (Ser632)/HDAC5 (Ser661)/HDAC7 (Ser486) Antibody (upper). Phospho-specificity of the antibody was determined by treating cell extracts with λ phosphatase. Total HDAC proteins were detected using Histone Deacetylase 4 (HDAC4) Antibody #2072, Histone Deacetylase 5 (HDAC5) Antibody #2082 and Histone Deacetylase 7 (HDAC7) Antibody #2882.

Source/Purification: Polyclonal antibodies are produced by immunizing animals with a synthetic peptide corresponding to human HDAC7 protein phosphorylated on Ser486. Antibodies are purified by protein A and peptide affinity chromatography.

Entrez Gene ID #9759, 10014, 51564
UniProt ID #P56524, Q9UQL6, Q8WUI4

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

***Species cross-reactivity is determined by western blot.**
****Anti-rabbit secondary antibodies must be used to detect this antibody.**

Recommended Antibody Dilutions:

Western Blotting	1:1000
Immunoprecipitation	1:25

Background References:

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IMPORTANT: For western blots, incubate membrane with diluted antibody in 5% w/v BSA, 1X TBS, 0.1% Tween® 20 at 4°C with gentle shaking, overnight.

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Applications Key: W—Western IP—Immunoprecipitation IHC—Immunohistochemistry ChIP—Chromatin Immunoprecipitation IF—Immunofluorescence F—Flow cytometry E-P—ELISA-Peptide
Species Cross-Reactivity Key: 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.