#9814 Store at -20°C

Acetylated-Lysine (Ac-K-100) MultiMab™ Rabbit mAb mix

Cell Signaling TECHNOLOGY®

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rev. 04/27/16

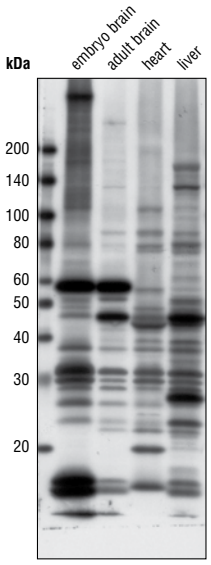
For Research Use Only. Not For Use In Diagnostic Procedures.

Applications	Species Cross-Reactivity*	Isotype	Motif
W, IP, E-P, ChIP	All	Rabbit IgG**	XXX(Kac)XXX
Endogenous			

Background: Acetylation of lysine, like phosphorylation of serine, threonine or tyrosine, is an important reversible modification controlling protein activity. The conserved amino-terminal domains of the four core histones (H2A, H2B, H3 and H4) contain lysines that are acetylated by histone acetyltransferases (HATs) and deacetylated by histone deacetylases (HDACs) (1). Signaling resulting in acetylation/deacetylation of histones, transcription factors and other proteins affects a diverse array of cellular processes including chromatin structure and gene activity, cell growth, differentiation and apoptosis (2-6). Recent proteomic surveys suggest that acetylation of lysine residues may be a widespread and important form of posttranslational protein modification that affects thousands of proteins involved in control of cell cycle and metabolism, longevity, actin polymerization and nuclear transport (7,8). The regulation of protein acetylation status is impaired in the pathologies of cancer and polyglutamine diseases (9), and HDACs have become promising targets for anti-cancer drugs currently in development (10).

Specificity/Sensitivity: Acetylated-Lysine (Ac-K-100) MultiMab™ Rabbit mAb mix recognizes proteins post-translationally modified by acetylation on the ε-amine groups of lysine residues. The antibody recognizes acetylated lysine in a wide range of sequence contexts. It has been demonstrated to recognize acetylated histones, p53, CBP, PCAF and chemically acetylated BSA. The antibody has been shown to react with as little as 0.04 ng of chemically acetylated BSA while not recognizing up to 25 µg of non-acetylated BSA. (U.S. Patent No's.: 6,441,140; 6,982,318; 7,259,022; 7,344,714; U.S.S.N. 11,484,485; and all foreign equivalents.)

Source/Purification: MultiMab™ rabbit monoclonal mix antibodies are prepared by combining individual rabbit monoclonal clones in optimized ratios for the approved applications. Each antibody in the mix is carefully selected based on motif recognition and performance in multiple assays. Each mix is engineered to yield the broadest possible coverage of the modification being studied while ensuring a high degree of specificity for the modification or motif



Western blot analysis of extracts from various mouse tissues using Acetylated-Lysine (Ac-K-100) MultiMab™ Rabbit mAb mix.

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.*

***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	1:100
Chromatin IP	1:50
ELISA-Peptide	1:1000

For application specific protocols please see the web page for this product at www.cellsignal.com.

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

Background References:

(1) Hassig, C.A. and Schreiber, S.L. (1997) *Curr Opin Chem Biol* 1, 300-8.

(2) Allfrey, V.G. et al. (1964) *Proc Natl Acad Sci USA* 51, 786-94.

(3) Liu, L. et al. (1999) *Mol Cell Biol* 19, 1202-9.

(4) Boyes, J. et al. (1998) *Nature* 396, 594-8.

(5) Polevoda, B. and Sherman, F. (2002) *Genome Biol* 3, reviews0006.

(6) Yoshida, M. et al. (2003) *Prog Cell Cycle Res* 5, 269-78.

(7) Kim, S.C. et al. (2006) *Mol Cell* 23, 607-18.

(8) Choudhary, C. et al. (2009) *Science* 325, 834-40.

(9) Hughes, R.E. (2002) *Curr Biol* 12, R141-3.

(10) Vigushin, D.M. and Coombes, R.C. (2004) *Curr Cancer Drug Targets* 4, 205-18.

U.S. Patent No. 5,675,063
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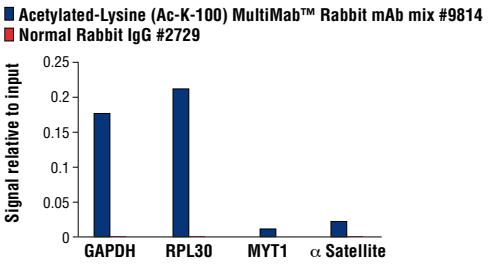
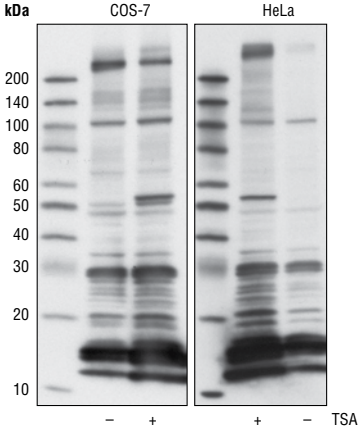
IMPORTANT: For western blots, incubate membrane with diluted antibody in 5% w/v BSA, 1X TBS, 0.1% Tween® 20 at 4°C with gentle shaking, overnight.

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.



Chromatin immunoprecipitations were performed with cross-linked chromatin from 4 x 10⁶ HeLa cells and either 10 µl of Acetylated-Lysine (Ac-K-100) MultiMab™ Rabbit mAb mix #9814 or 2 µl of Normal Rabbit IgG #2729, using SimpleChIP® Enzymatic Chromatin IP Kit (Magnetic Beads) #9003. The enriched DNA was quantified by real-time PCR using SimpleChIP® Human GAPDH Exon 1 Primers #5516, SimpleChIP® Human RPL30 Exon 3 Primers #7014, SimpleChIP® Human MYT-1 Exon 1 Primers #4493, and SimpleChIP® Human α Satellite Repeat Primers #4486. The amount of immunoprecipitated DNA in each sample is represented as signal relative to the total amount of input chromatin, which is equivalent to one.



Western blot analysis of extracts from COS-7 and HeLa cells, untreated (-) or treated with Trichostatin A (TSA) #9950 (1 µM, 6 hr; +), using Acetylated-Lysine (Ac-K-100) MultiMab™ Rabbit mAb mix.