Revision 3

G9a/EHMT2 (C6H3) Rabbit mAb

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Applications:	Reactivity:	Sensitivity:	MW (kDa):	Source/Isotype:	UniProt ID:	Entrez-Gene Id:
W, IF-IC, ChIP	H M R Mk	Endogenous	160,180	Rabbit IgG	#Q96KQ7	10919

Product Usage Information

For optimal ChIP results, use 10 µl of antibody and 10 µg of chromatin (approximately 4 x 10^6 cells) per IP. This antibody has been validated using SimpleChIP® Enzymatic Chromatin IP Kits.

Application	Dilution
Western Blotting	1:1000
Immunofluorescence (Immunocytochemistry)	1:50
Chromatin IP	1:100

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.

Specificity/Sensitivity

G9a/EHMT2 (C6H3) Rabbit mAb detects endogenous levels of total G9a/EHMT2 protein (both the 165 kDa G9a-L and 140 kDa G9a-S isoforms). This antibody does not cross-react with other histone methyltransferases, including GLP.

Species predicted to react based on 100% sequence homology

Bovine, Pig, Horse

Source / Purification

Monoclonal antibody is produced by immunizing animals with a synthetic peptide corresponding to the carboxy terminus of the human G9a/EHMT2 protein.

Background

G9a, also known as Euchromatic histone-lysine N-methyltransferase 2 (EHMT2), is a member of a family of histone lysine methyltransferases, each of which contains a conserved catalytic SET domain originally identified in Drosophila Su[var]3-9, Enhancer of zeste, and Trithorax proteins (1). Recombinant G9a can mono-, di- and tri-methylate histone H3 on Lys9 and Lys27 *in vitro* (1,2). However, *in vivo* G9a forms a complex with GLP, a G9a-related histone methyltransferase, and together these proteins function as the major euchromatic histone H3 Lys9 mono- and di-methyltransferases, creating transcriptionally repressive marks that facilitate gene silencing (3,4). G9a methylates itself on Lys165, a modification that regulates the association of HP1 repressor proteins with the G9a/GLP complex (5). The G9a/GLP complex also contains Wiz, a zinc finger protein that is required for G9a/GLP hetero-dimerization and complex stability (6). Wiz contains two CtBP co-repressor binding sites, which mediate the association of the G9a/GLP with the CtBP co-repressor complex (6). In addition, G9a and GLP are components of other large transcriptional co-repressor complexes, such as those involving E2F6 and CDP/cut (7-9). G9a interacts with DNMT1, and both proteins are required for methylation of DNA and histone H3 (Lys9) at replication foci, providing a functional link between histone H3 Lys9 and CpG methylation during DNA replication (10). G9a activity is critical for meiotic prophase progression, as mutant mice deficient in germ line G9a show a large loss of mature gametes (11). In addition, G9a facilitates increased global levels of di-methyl histone H3 (Lys9) during hypoxic stress and increased G9a expression is associated with hepatocelluar carcinoma (12,13).

Background References

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4. Tachibana, M. et al. (2005) *Genes Dev* 19, 815-26.
5. Sampath, S.C. et al. (2007) *Mol Cell* 27, 596-608.
6. Ueda, J. et al. (2006) *J Biol Chem* 281, 20120-8.
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8. Shi, Y. et al. (2003) *Nature* 422, 735-8.
9. Nishio, H. and Walsh, M.J. (2004) *Proc Natl Acad Sci USA* 101, 11257-62.
10. Estève, P.O. et al. (2006) *Genes Dev* 20, 3089-103.
11. Tachibana, M. et al. (2007) *EMBO J* 26, 3346-59.
12. Kondo, Y. et al. (2007) *Hepatol Res* 37, 974-83.

Species Reactivity

Species reactivity is determined by testing in at least one approved application (e.g., western blot).

Western Blot Buffer

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

Applications Key

W: Western Blotting **IF-IC:** Immunofluorescence (Immunocytochemistry) **ChIP:** Chromatin IP

Cross-Reactivity Key

H: Human **M:** Mouse **R:** Rat **Mk:** Monkey

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