### G9a/EHMT2 (D5R4R) XP® Rabbit mAb



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#### For Research Use Only. Not for Use in Diagnostic Procedures.

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

## Product Usage Information

For optimal ChIP results, use 10  $\mu$ l of antibody and 10  $\mu$ g of chromatin (approximately 4 x 10<sup>6</sup> cells) per IP. This antibody has been validated using SimpleChIP<sup>®</sup> Enzymatic Chromatin IP Kits.

The CUT&RUN dilution was determined using CUT&RUN Assay Kit #86652.

Application	Dilution
Western Blotting	1:1000
Immunoprecipitation	1:100
Immunofluorescence (Immunocytochemistry)	1:3200
Chromatin IP	1:50
CUT&RUN	1:50

#### Storage

Supplied in 10 mM sodium HEPES (pH 7.5), 150 mM NaCl,  $100 \mu g/ml$  BSA, 50% glycerol and less than 0.02% sodium azide. Store at -20°C. Do not aliquot the antibody.

# Specificity/Sensitivity Source / Purification

G9a/EHMT2 (D5R4R) XP® Rabbit mAb recognizes endogenous levels of total G9a protein.

Monoclonal antibody is produced by immunizing animals with recombinant protein specific to the amino terminus of human G9a 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 heterodimerization 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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- 3. Tachibana, M. et al. (2002) Genes Dev 16, 1779-91.
- 4. Tachibana, M. et al. (2005) *Genes Dev* 19, 815-26.
- 5. Sampath, S.C. et al. (2007) *Mol Cell* 27, 596-608.
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- 7. Ogawa, H. et al. (2002) Science 296, 1132-6.
- 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 nonfat

dry milk, 1X TBS, 0.1% Tween® 20 at 4°C with gentle shaking, overnight.

**Applications Key** W: Western Blotting IP: Immunoprecipitation IF-IC: Immunofluorescence (Immunocytochemistry)

ChIP: Chromatin IP C&R: CUT&RUN

Cross-Reactivity Key H: Human M: Mouse R: Rat Mk: Monkey

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