#12686 Store at -20°C

Phospho-MCM3 (Ser112) (D3S4M) Rabbit mAb

☑ 100 µl
(10 western blots)

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rev. 09/23/13

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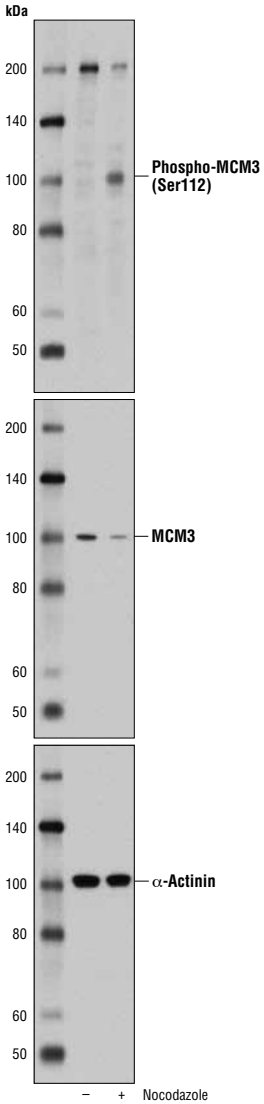
Entrez Gene ID #4172
UniProt ID #P25205

Applications	Species Cross-Reactivity*	Molecular Wt.	Isotype
W, IP Endogenous	H, (M, R, Hm, Z, Dg)	100 kDa	Rabbit IgG**

Background: The minichromosome maintenance (MCM) 2-7 proteins are a family of six related proteins required for initiation and elongation of DNA replication. MCM2-7 bind together to form the heterohexameric MCM complex that is thought to act as a replicative helicase at the DNA replication fork (1-5). This complex is a key component of the pre-replication complex (pre-RC) (reviewed in 1). Cdc6 and CDT1 recruit the MCM complex to the origin recognition complex (ORC) during late mitosis/early G1 phase forming the pre-RC and licensing the DNA for replication (reviewed in 2). Licensing of the chromatin permits the DNA to replicate only once per cell cycle, thereby helping to ensure that genetic alterations and malignant cell growth do not occur (reviewed in 3). Phosphorylation of the MCM2, MCM3, MCM4, and MCM6 subunits appears to regulate MCM complex activity and the initiation of DNA synthesis (6-8). CDK1 phosphorylation of MCM3 at serine 112 during late mitosis/early G1 phase has been shown to initiate complex formation and chromatin loading *in vitro* (8). MCM proteins are removed during DNA replication, causing chromatin to become unlicensed through inhibition of pre-RC reformation. Studies have shown that the MCM complex is involved in checkpoint control by protecting the structure of the replication fork and assisting in restarting replication by recruiting checkpoint proteins after arrest (reviewed in 3,9).

Specificity/Sensitivity: Phospho-MCM3 (Ser112) (D3S4M) Rabbit mAb recognizes endogenous levels of MCM3 protein only when phosphorylated at Ser112. This antibody also recognizes a 200 kDa band of unknown origin in both interphase and mitotic cells.

Source/Purification: Monoclonal antibody is produced by immunizing animals with a synthetic phosphopeptide corresponding to residues surrounding Ser112 of human MCM3 protein.



Western blot analysis of HeLa cell extracts, untreated (-) or treated with Nocodazole #2190 (100 ng/ml, 24 hr; +), using Phospho-MCM3 (Ser112) (D3S4M) Rabbit mAb (upper), MCM3 (D47B6) Rabbit mAb #4003 (middle) and α-Actinin (D6F6) XP® Rabbit mAb #6487 (lower).

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.

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	1:200

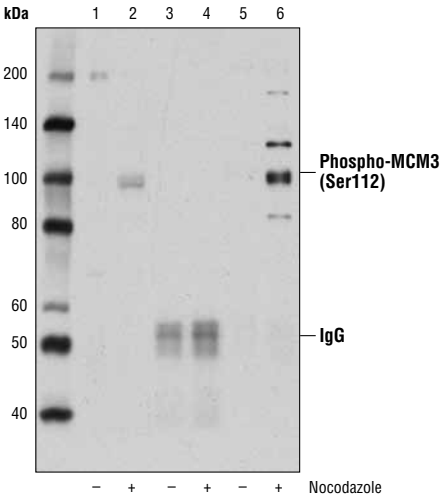
For product specific protocols please see the web page for this product at www.cellsignal.com.

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

Background References:

(1) Lei, M. and Tye, B.K. (2001) *J Cell Sci* 114, 1447-54.
(2) Lygerou, Z. and Nurse, P. (2000) *Science* 290, 2271-3.
(3) Forsburg, S.L. (2004) *Microbiol Mol Biol Rev* 68, 109-31.
(4) Tye, B.K. and Sawyer, S. (2000) *J Biol Chem* 275, 34833-6.
(5) Labib, K. et al. (2000) *Science* 288, 1643-7.
(6) Charych, D.H. et al. (2008) *J Cell Biochem* 104, 1075-86.
(7) Masai, H. et al. (2006) *J Biol Chem* 281, 39249-61.
(8) Lin, D.I. et al. (2008) *Proc Natl Acad Sci USA* 105, 8079-84.
(9) Bailis, J.M. et al. (2008) *Mol Cell Biol* 28, 1724-38.

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.



Immunoprecipitation of Phospho-MCM3 (Ser112) from HeLa cell extracts, untreated (-) or treated with Nocodazole #2190 (100 ng/ml, 24 hr; +), using Rabbit (DA1E) mAb IgG XP® Isotype Control #3900 (lane 3 and 4) or Phospho-MCM3 (Ser112) (D3S4M) Rabbit mAb (lane 5 and 6). Lanes 1 and 2 are 10% input. Western blot analysis was performed using Phospho-MCM3 (Ser112) (D3S4M) Rabbit mAb.