

#9127 Store at -20°C

# Phospho-MEK1 (Thr286) Antibody

✓ 100 µl  
(10 western blots)



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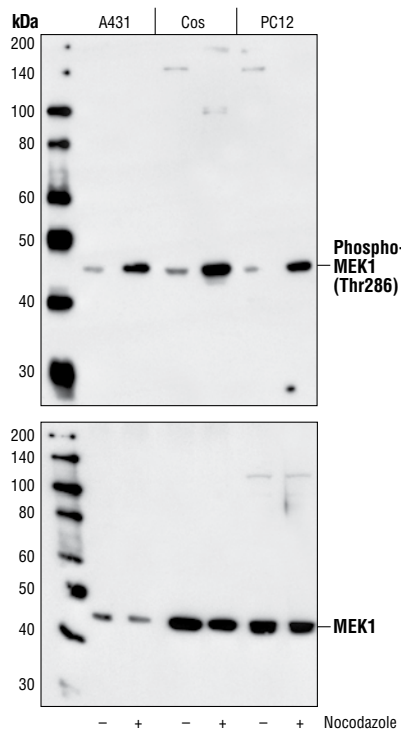
This product is intended for research purposes only. This product is not intended to be used for therapeutic or diagnostic purposes in humans or animals.

Applications	Species Cross-Reactivity*	Molecular Wt.	Source
W, IP, IF-IC, F Endogenous	H, R, Mk, (M)	45 kDa	Rabbit**

**Background:** MEK1 and MEK2, also called MAPK or Erk kinases, are dual-specificity protein kinases that function in a mitogen activated protein kinase cascade controlling cell growth and differentiation (1–3). Activation of MEK1 and MEK2 occurs through phosphorylation of two serine residues at positions 217 and 221 (in the activation loop of subdomain VIII) by Raf-like molecules. MEK1/2 is activated by a wide variety of growth factors and cytokines and also by membrane depolarization and calcium influx (1–4). Constitutively active forms of MEK1/2 are sufficient for the transformation of NIH/3T3 cells or the differentiation of PC12 cells (4). MEK activates p44 and p42 MAP kinase by phosphorylating both threonine and tyrosine residues at sites located within the activation loop of kinase subdomain VIII.

MEK1 is phosphorylated at Ser298 by PAK1, which facilitates signal transduction from Raf to MEK1 and Erk2 (5–7). MEK1 is also phosphorylated by cdk5 at Thr286 in mitotic cells, causing negative feedback of the p44/42 MAP kinase pathway (8).

**Specificity/Sensitivity:** Phospho-MEK1 (Thr286) Antibody detects endogenous levels of MEK1 phosphorylated at threonine 286. This antibody does not cross-react with phosphorylated MEK2.



Western blot analysis of extracts from A431, COS and PC12 cells, untreated or nocodazole-treated, using Phospho-MEK1 (Thr286) Antibody (upper) or MEK1 Antibody #9124 (lower).

**Source/Purification:** Polyclonal antibodies are produced by immunizing animals with a synthetic phospho-peptide (KLH-coupled) corresponding to residues surrounding Thr286 of human MEK1. Antibodies are purified by protein A and peptide affinity chromatography.

Entrez-Gene ID #5604  
Swiss-Prot Acc. #Q02750

**Storage:** Supplied in 10 mM sodium 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.

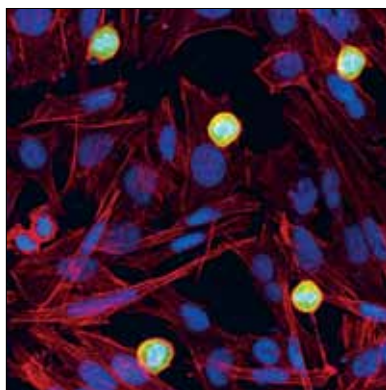
\*\*Anti-rabbit secondary antibodies must be used to detect this antibody.

**Recommended Antibody Dilutions:**

Western blotting	1:1000
Immunoprecipitation	1:50
Immunofluorescence (IF-IC)	1:100
Flow Cytometry	1:50

For application specific protocols please see the web page for this product at [www.cellsignal.com](http://www.cellsignal.com).

Please visit [www.cellsignal.com](http://www.cellsignal.com) for a complete listing of recommended companion products.



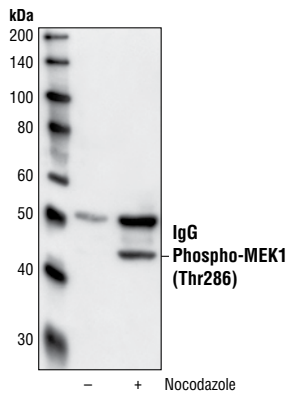
Confocal immunofluorescent analysis of mitotic HeLa cells labeled with Phospho-MEK1 (Thr286) Antibody (green). Actin filaments have been labeled with Alexa Fluor<sup>®</sup> 555 phalloidin (red). Blue pseudocolor = DRAQ5<sup>®</sup> #4084 (fluorescent DNA dye).

**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.

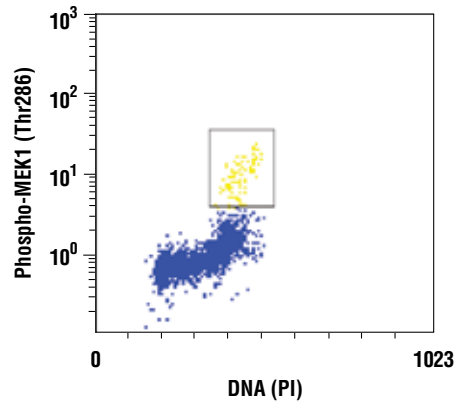
DRAQ5<sup>®</sup> is a registered trademark of Biostatus Limited.

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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 All—all species expected Species enclosed in parentheses are predicted to react based on 100% homology.



Immunoprecipitation followed by Western blot analysis of extracts from COS cells, untreated or nocodazole-treated, using Phospho-MEK1 (Thr286) Antibody.



Flow cytometric analysis of untreated Jurkat cells, using Phospho-MEK1 (Thr286) Antibody versus propidium iodide (DNA content). The box indicates Phospho-MEK1 positive cells.

**Background References:**

- (1) Crews, C.M. et al. (1992) *Science* 258, 478–480.
- (2) Alessi, D.R. et al. (1994) *EMBO J.* 13, 1610–1619.
- (3) Rosen, L.B. et al. (1994) *Neuron* 12, 1207–1221.
- (4) Cowley, S. et al. (1994) *Cell* 77, 841–852.
- (5) Xu, B. et al. (1999) *J. Biol. Chem.* 274, 34029–34035.
- (6) Coles, L.C. and Shaw, P.E. (2002) *Oncogene* 21, 2236–2244.
- (7) Eblen, S.T. et al. (2002) *Mol. Cell. Biol.* 22, 6023–6033.
- (8) Sharma, P. et al. (2002) *J. Biol. Chem.* 277, 528–534.