

SignalSlide® Phospho-p44/42 MAPK (Thr202/Tyr204) IHC Controls

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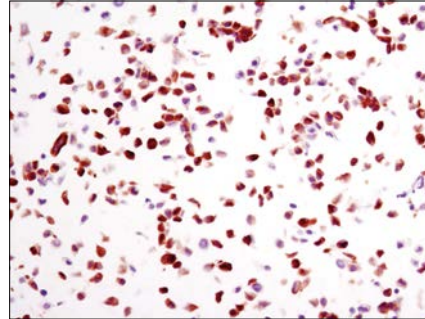
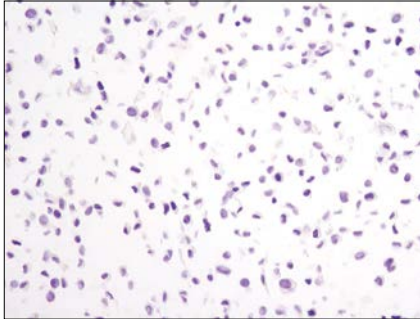
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Immunohistochemical analysis of paraffin-embedded NIH/3T3 cells, treated with U0126 #9903 (left) or TPA (12-O-Tetradecanoylphorbol-13-Acetate) #4174 (right), using Phospho-p44/42MAPK (Erk1/2) (Thr202/Tyr204) (D13.14.4E) XP™ Rabbit mAb #4370.

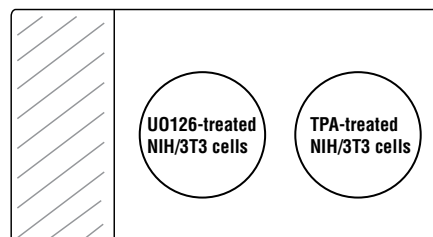
Description: Each control slide contains formalin fixed, paraffin-embedded NIH/3T3 cells, treated with either U0126 (1,4-diamino-2,3-dicyano-1,4-bis[2-aminophenylthio] butadiene) #9903 or TPA (12-O-Tetradecanoylphorbol-13-Acetate) #4174, that serve as a control for phospho-p44/42 MAPK (Thr202/Tyr204) immunostaining. U0126 has been shown to be a highly selective inhibitor of MEK1 and MEK2. TPA induces phosphorylation of p44/42 MAPK. Western blot analysis was performed on extracts derived from the same cells to verify the efficacy of the U0126 and TPA treatments.

Background: Mitogen-activated protein kinases (MAPKs) are a widely conserved family of serine/threonine protein kinases involved in many cellular programs such as cell proliferation, differentiation, motility, and death. The p44/42 MAPK (Erk1/2) signaling pathway can be activated in response to a diverse range of extracellular stimuli including mitogens, growth factors, and cytokines (1-3) and is an important target in the diagnosis and treatment of cancer (4). Upon stimulation, a sequential three-part protein kinase cascade is initiated, consisting of a MAP kinase kinase kinase (MAPKKK or MAP3K), a MAP kinase kinase (MAPKK or MAP2K), and a MAP kinase (MAPK). Multiple p44/42 MAP3Ks have been identified, including members of the Raf family as well as Mos and Tpl2/Cot. MEK1 and MEK2 are the primary MAPKKs in this pathway (5,6). MEK1 and MEK2 activate p44 and p42 through phosphorylation of activation loop residues Thr202/Tyr204 and Thr185/Tyr187, respectively. Several downstream targets of p44/42 have been identified, including p90RSK (7) and the transcription factor Elk-1 (8,9). p44/42 are negatively regulated by a family of dual-specificity (Thr/Tyr) MAPK phosphatases, known as DUSPs or MKPs (10), along with MEK inhibitors such as U0126 and PD98059.

Applications: These slides are intended for use in immunohistochemical assays.

Background References:

- (1) Roux, P.P. and Blenis, J. (2004) *Microbiol Mol Biol Rev* 68, 320-44.
- (2) Baccarini, M. (2005) *FEBS Lett* 579, 3271-7.
- (3) Meloche, S. and Pouyssegur, J. (2007) *Oncogene* 26, 3227-39.
- (4) Roberts, P.J. and Der, C.J. (2007) *Oncogene* 26, 3291-310.
- (5) Rubinfeld, H. and Seger, R. (2005) *Mol Biotechnol* 31, 151-74.
- (6) Murphy, L.O. and Blenis, J. (2006) *Trends Biochem Sci* 31, 268-75.
- (7) Dalby, K.N. et al. (1998) *J Biol Chem* 273, 1496-505.
- (8) Marais, R. et al. (1993) *Cell* 73, 381-93.
- (9) Kortenjann, M. et al. (1994) *Mol Cell Biol* 14, 4815-24.
- (10) Owens, D.M. and Keyse, S.M. (2007) *Oncogene* 26, 3203-13.



Entrez-Gene ID # 5595, 5594

Swiss-Prot Acc. # P27361, P28482

Storage: Store at 4° C.

Optimal staining is achieved if slides are stained following CST's standard IHC protocols and are used within 8 weeks of assay date; however, signals may persist beyond two months.

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.