

#9386 Store at -20°C

# Phospho-Threonine (42H4) Mouse mAb

✓ 100 µl (10 western blots)



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rev 11/02/10

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*	Isotype
W, IP, E-P Endogenous	All	Mouse IgM**

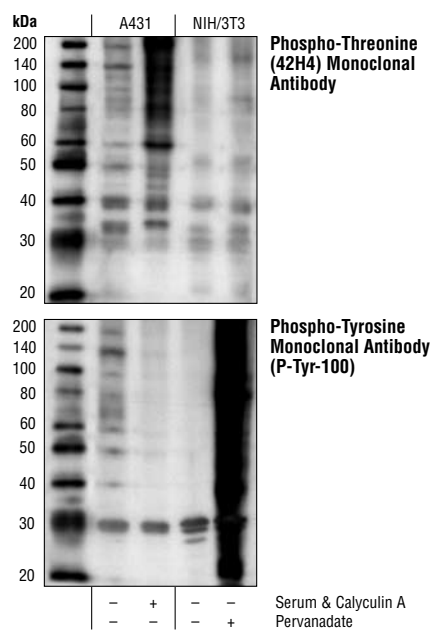
**Background:** Much of the dynamic behavior of cellular proteins, including the regulation of molecular interactions (1), subcellular localization (2) and transcriptional regulation (3) is controlled by a variety of posttranslational modifications (4). Antibodies specific for these posttranslational modifications are invaluable tools in the quest to understand normal and pathogenic molecular and cellular behavior.

General protein modification antibodies are designed to react with modified amino acid residues (e.g., phospho-threonine, phospho-tyrosine, acetyl-lysine, nitro-tyrosine) independently of the sequence in which they are embedded. This ability to recognize modified residues in a "context independent" fashion gives these antibodies broad reactivities, presumably conferring upon them the ability to react with hundreds of distinct proteins. This broad pattern of reactivity makes these antibodies especially valuable in multiplex analyses and target discovery programs.

Protein kinases are among the most abundant eukaryotic regulatory proteins; over 500 separate kinase genes are encoded in mammalian genomes (5,6). In spite of the importance of kinases in eukaryotic biology, relatively few of their physiological targets are known. Phospho-Threonine Antibody (P-Thr-Polyclonal) #9381 and Phospho-Threonine (42H4) Monoclonal Antibody #9386 provide powerful tools for discovering targets of serine/threonine kinases, for monitoring and characterizing *in vitro* threonine phosphorylation reactions as well as for high throughput Ser/Thr kinase drug discovery.

**Specificity/Sensitivity:** Phospho-Threonine (42H4) Mouse mAb binds phosphorylated threonine residues in a manner largely independent of the surrounding amino acid sequence. The antibody is phospho-specific but does not cross-react with phospho-tyrosine-containing sequences. It does show slight cross-reactivity with a few phosphoserine-containing peptides. By ELISA, it recognizes a wide variety of threonine-phosphorylated peptides. (U.S. Patent No.'s.: 6,441,140; 6,982,318; 7,259,022; 7,344,714; U.S.S.N. 11,484,485; and all foreign equivalents.)

**License/Use Restrictions:** Use of CST Motif Antibodies within certain methods (e.g., U.S. Patent No.'s 7,198,896 & 7,300,753) may require a license from CST. For information regarding academic licensing terms please have your technology transfer office contact CST Legal Department at CST\_ip@cellsignal.com. For information regarding commercial licensing terms please contact CST Pharma Services Department at ptmscan@cellsignal.com.



Western blot analysis of extracts from A431 cells, untreated or treated with calyculin A, a threonine phosphatase inhibitor, or extracts from NIH/3T3 cells, untreated or treated with pervanadate, a tyrosine phosphatase inhibitor, using Phospho-Threonine (42H4) Mouse mAb (upper) or Phospho-Tyrosine Monoclonal Antibody (P-Tyr-100) #9411 (lower).

**Source/Purification:** Monoclonal antibody is produced by immunizing animals with phospho-Thr-containing peptides.

**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-mouse secondary antibodies must be used to detect this antibody.

**Recommended Antibody Dilutions:**

Western Blotting	1:1000
Immunoprecipitation	1:50
ELISA (Peptide)	1:2000

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.

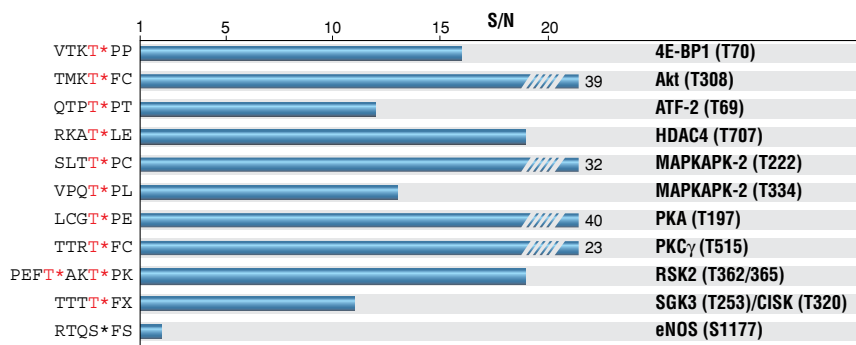
**Background References:**

- (1) Yaffe, M.B. and Elia, A.E. (2001) *Curr. Opin. Cell Biol.* 13, 131–138.
- (2) Appella, E. and Anderson, C.W. (2001) *J. Biochem. (Tokyo)* 268, 2764–2772.
- (3) Jenuwein, T. and Allis, C.D. (2001) *Science* 293, 1074–1080.
- (4) Krishna, R.G. and Wold, F. (1993) *Adv. Enzymol. Relat. Areas Mol. Biol.* 67, 265–298.
- (5) Venter, J.C. et al. (2000) *Science* 291, 1304–1351.
- (6) Manning, G. et al. (2002) *Science* 298, 1912–1934.

**IMPORTANT: For western blots, incubate membrane with diluted antibody in 5% w/v nonfat dry milk, 1X TBS, 0.1% Tween-20 at 4°C with gentle shaking, overnight.**

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



Phospho-Threonine (42H4) Mouse mAb ELISA Assay: Signal-to-noise ratio of phospho- versus nonphospho-peptides. (T\* denotes phosphorylated threonine.)