

Phospho-Threonine-X-Arginine Antibody

100 µl
 (10 Western mini-blots)



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This product is for *in vitro* research use only and is not intended for use in humans or animals.
This product is not intended for use as a therapeutic or in diagnostic procedures.

Applications	Species Cross-Reactivity*	Source	Motif
W, IHC-P, E-P	All	Rabbit**	T*XR

Background: Some signaling molecules can be regulated by phosphorylation at a specific threonine followed by arginine or lysine at the +2 position. For example, conventional PKC isozymes phosphorylate substrates containing serine or threonine with Arg or Lys at the -3, -2 and +2 positions (1-2). c-Raf, a mitogen-activated protein kinase and the main effector recruited by GTP-bound Ras, is phosphorylated at Thr481 and Thr491 followed by Lys at the +2 position (3). Phosphorylation of these sites is important for enzyme activities. To determine the phosphorylation state of Thr in the Thr-X-Arg motif, and to identify potential new phosphorylation sites with this motif, CST has developed a Phospho-Threonine X-Arginine Antibody that recognizes phosphorylated Thr followed by Arg or Lys at the +2 position.

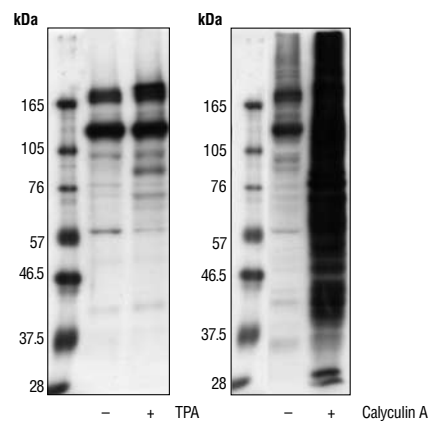
Specificity/Sensitivity: Phospho-Threonine-X-Arginine Antibody detects endogenous levels of proteins containing the phospho-Thr-X-Arg motif. This antibody detects phosphorylated Thr followed by Arg or Lys at the +2 position, though its reactivity is lower for Lys compared to Arg at the +2 position. The antibody does not cross-react with nonphospho-Thr or phospho-Ser in the same motif. It recognizes phospho-Thr in the FFT*R motif in PKCβ II but does not recognize phospho-Thr in other motifs that lack Lys or Arg at +2. (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.)

Source/Purification: Polyclonal antibodies are produced by immunizing animals with a synthetic peptide (KLH-coupled) containing the phospho-Thr-X-Arg motif. Antibodies are purified by protein A and peptide affinity chromatography.

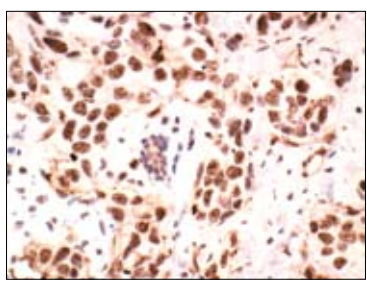
- Background References:**
 (1) Nishikawa, K. et al. (1997) *J. Biol. Chem.* 272, 952-960.
 (2) Pearson, R.B. and Kemp, B.E. (1991) *Methods Enzymol.* 200, 62-81.
 (3) Zhang, B. and Guan, K. (2000) *EMBO J.* 19, 5429-5439.

Phospho-Threonine-X-Arginine Antibody ELISA: Signal-to-noise ratio of phospho- versus nonphospho-peptides containing the phospho-threonine-X-arginine motif. (T denotes phosphorylated threonine.)*

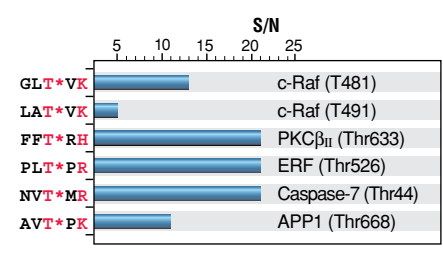
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Western blot analysis of extracts from Jurkat cells, untreated, TPA-treated or calyculin A-treated, using Phospho-Threonine-X-Arginine Antibody.



Immunohistochemical analysis of paraffin-embedded human breast carcinoma, showing staining of proteins containing phosphorylated threonine-X-arginine motifs, using Phospho-Threonine-X-Arginine Antibody.



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
 Immunohistochemistry (Paraffin) 1:200
IHC protocol: Unmasking buffer/Antibody diluent Citrate/TBST-5%NGS
 ELISA-Peptide 1:1000

- Companion Products:**
 Phospho-Akt Substrate (RXRXXS/T) (110B7E) Rabbit mAb #9614
 Phospho-PKA Substrate (RRXS/T) (100G7E) Rabbit mAb #9624
 Phospho-(Ser) PKC Substrate Antibody #2261
 Phospho-MAPK Substrates (PXTTP) (46G11) Rabbit mAb #4391
 Phospho-MAPK/CDK Substrates (PXSP or SPXR/K) (34B2) Rabbit mAb #2325
 Phospho-(Thr) MAPK/CDK Substrate Mouse mAb #2321
 Phospho-(Ser) CDKs Substrate Antibody #2324
 Phospho-(Ser/Thr) ATM/ATR Substrate (4F7) Rabbit mAb #2909
 Phospho-(Ser) Arg-X-Tyr/Phe-X-pSer Motif Antibody #2981
 Phospho-(Tyr) p85 PI3K Binding Motif Antibody #3821
 Phospho-Threonine Antibody (P-Thr-Polyclonal) #9381
 Phospho-Tyrosine Mouse mAb (P-Tyr-100) #9411
 Anti-rabbit IgG, HRP-linked Antibody #7074
 Phototope®-HRP Western Blot Detection System, Anti-rabbit IgG, HRP-linked Antibody #7071
 Prestained Protein Marker, Broad Range (Premixed Format) #7720
 Biotinylated Protein Ladder Detection Pack #7727
 20X LumiGLO® Reagent and 20X Peroxide #7003

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

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.