

Phospho-PKC (pan) (ζ Thr410) (190D10) Rabbit mAb

- Small 100 μ l (10 western blots)
- Large 300 μ l (30 western blots)

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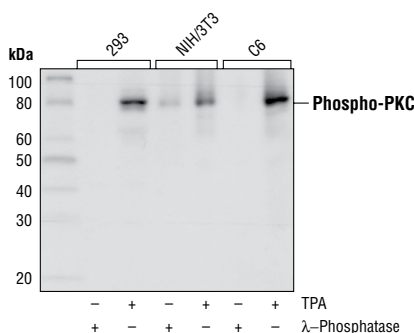
This product is intended for research purposes only. This product is not intended to be used for therapeutic or diagnostic purposes in humans or animals.

Applications	Species Cross-Reactivity	Molecular Wt.	Source	Isotype
W	H, M, R, Mk	76 to 85 kDa	Rabbit*	IgG

Background: Activation of PKC is one of the earliest events in a cascade leading to a variety of cellular responses such as secretion, gene expression, proliferation and muscle contraction (1,2). PKC isoforms have been classified into three groups: classical PKCs, which are Ca²⁺ dependent via their C2 domains and are activated by phosphatidylserine (PS), diacylglycerol (DAG) and phorbol esters (TPA or PMA) through their cysteine-rich C1 domains, novel PKCs, which are Ca²⁺ independent but are still regulated by PS, DAG and TPA and atypical PKCs, which are Ca²⁺ independent and do not require PS, DAG or TPA for their activation (3–7). Members of these three PKC groups contain a pseudo-substrate or autoinhibitory domain that binds to the substrate binding site in the catalytic domain, preventing its activation in the absence of cofactors or activators.

Other members have been recently added to the PKC superfamily based on homology within the catalytic domain. PKC μ , or PKD, is regulated by DAG and TPA through its C1 domain. However, PKD is distinguished by a PH domain, as well as by its unique substrate recognition and Golgi localization. The PKC-related kinases, or PRKs, lack a C1 domain and thus do not respond to DAG or phorbol esters. Instead, they can be activated by phosphatidylinositol lipids and their kinase activity is directly regulated by small GTPases of the Rho family through Rho binding to the homology region 1 (HR1).

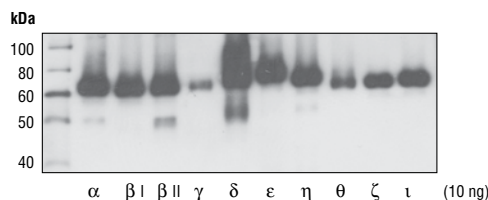
The activity of PKC is under the control of three distinct phosphorylation events. Specifically, Thr500 in the activation loop, the Thr641 autophosphorylation site and the Ser660 hydrophobic site at the carboxy-terminus of PKC β II are phosphorylated *in vivo* (2). For the atypical PKC isoforms, there is no phosphorylation in the hydrophobic region, which has a glutamic acid residue in place of the serine or threonine residue found in other PKC isoforms. The enzyme PDK1, or perhaps a close relative, is responsible for PKC activation.



Western blot analysis of extracts from 293, NIH/3T3 and C6 cells, treated with TPA or λ -phosphatase as indicated, using Phospho-PKC (pan) (ζ Thr410) (190D10) Rabbit mAb.

Specificity/Sensitivity: Phospho-PKC (pan) (ζ Thr410) (190D10) Rabbit mAb detects endogenous levels of PKC α , β I, β II, γ , δ , ϵ , ν , θ and ι isoforms only when phosphorylated at a residue homologous to threonine 410 of human PKC ζ .

Source/Purification: Monoclonal antibody is produced by immunizing animals with a synthetic phosphopeptide corresponding to residues surrounding Thr410 of human PKC ζ .



Western blot analysis of baculovirus-expressed PKC isoforms, using Phospho-PKC (pan) (ζ Thr410) (190D10) Rabbit mAb.

Storage: Supplied in 10 mM sodium HEPES (pH 7.5), 150 mM NaCl, 100 μ g/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

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.

Background References:

- (1) Nishizuka, Y. (1984) *Nature* 308, 693–698.
- (2) Keranen, L.M. et al. (1995) *Curr. Biol.* 5, 1394–1403.
- (3) Newton, A.C. (1995) *J. Biol. Chem.* 270, 28495–28498.
- (4) Mellor, H. and Parker, P.J. (1998) *Biochem. J.* 332, 281–292.
- (5) Ron, D. and Kazanietz, M.G. (1999) *FASEB J.* 13, 1658–1676.
- (6) Way, K.J. et al. (2000) *Trends Pharmacol. Sci.* 21, 181–187.
- (7) Moscat, J. and Diaz-Meco, M.T. (2000) *EMBO Rep.* 1, 399–403.
- (8) Good, J.A. et al. (1998) *Science* 281, 2042–2045.
- (9) Sonnenburg, E.D. (2001) *J. Biol. Chem.* 276, 45289–45297.

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.

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.