

Kit Includes	Quantity	Applications	Reactivity	MW (kDa)	Isotype
PKCα Antibody #2056	40 μ l	W IP IF-IC F	H M R Mk (Dg)	80	Rabbit
PKCζ (C24E6) Rabbit mAb #9368	40 μ l	W	H M R Mk	78	Rabbit IgG
PKD/PKCμ Antibody #2052	40 μ l	W F	H M R Mk	115	Rabbit
PKCδ (D10E2) Rabbit mAb #9616	40 μ l	W IP	H M R Mk (X) (B) (Dg) (Hr)	78	Rabbit IgG
Anti-rabbit IgG, HRP-linked Antibody #7074	100 μ l				Goat

Applications Key: W=Western Blotting IP=Immunoprecipitation IF-IC=Immunofluorescence (Immunocytochemistry) F=Flow Cytometry

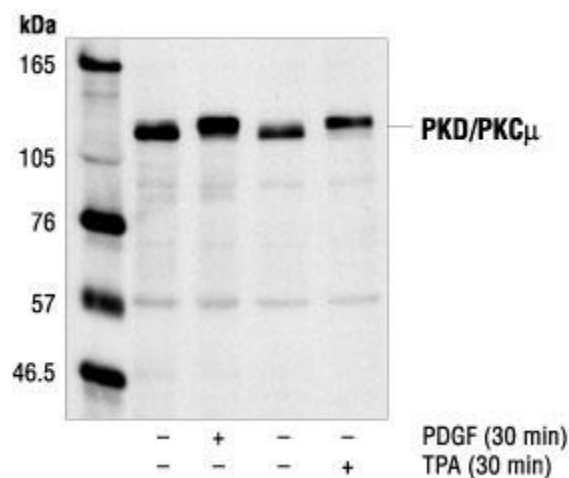
Reactivity Key: H=Human M=Mouse R=Rat Mk=Monkey X=Xenopus B=Bovine Dg=Dog Hr=Horse

Species enclosed in parentheses are predicted to react based on 100% sequence homology.

Specificity / Sensitivity

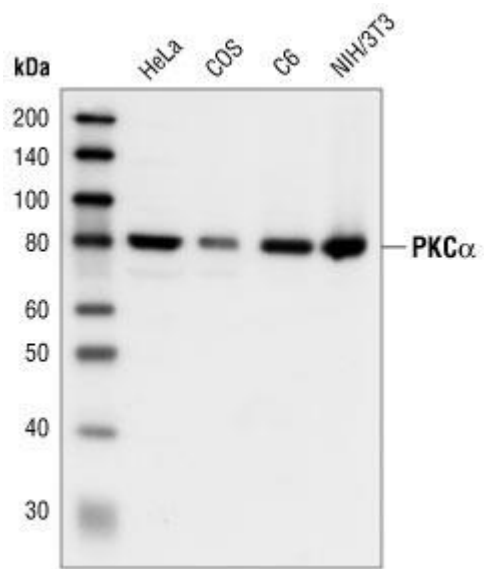
All antibodies in this kit detect endogenous levels of total protein from their respective targets. All antibodies only recognize their specified isoform and do not cross-react with other PKC isoforms.

Western Blotting



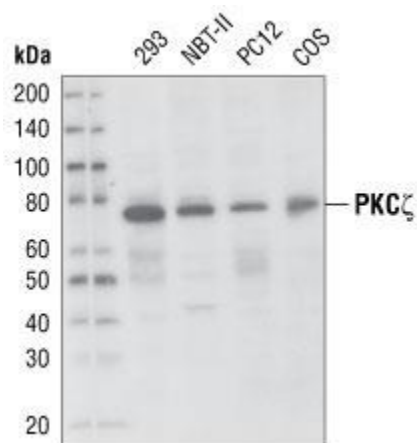
Western blot analysis of extracts from NIH/3T3 cells, untreated, PDGF-treated (50 ng/ml) or TPA-treated (0.2 μ M), using PKD/PKC μ Antibody #2052.

Western Blotting

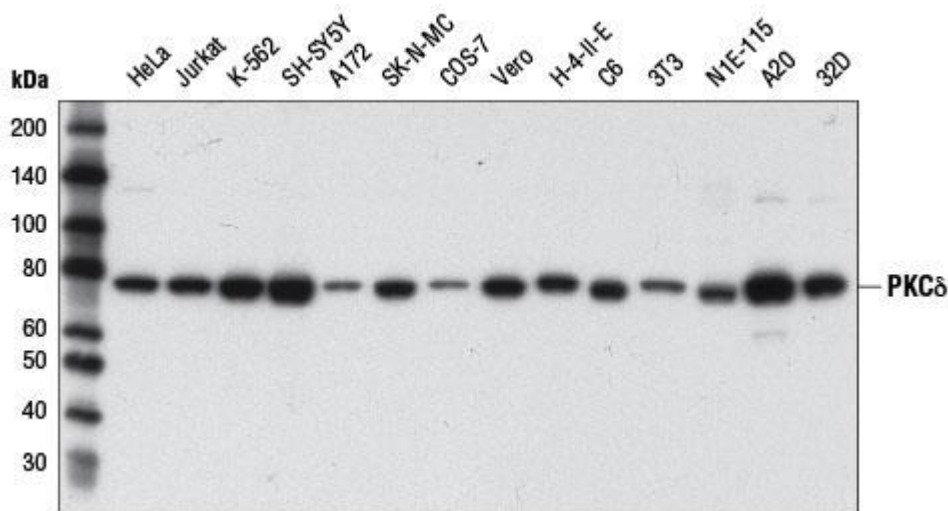


Western blot analysis of extracts of HeLa, COS, C6 and NIH/3T3 cells using PKC α Antibody #2056.

Western Blotting



Western blot analysis of extracts from 293, NBT-II, PC12 and COS cells using PKC ζ (C24E6) Rabbit mAb #9368.



Western blot analysis of extracts from various cell lines using PKCδ (D10E2) Rabbit mAb #9616.

Description

PKC Antibody Sampler Kit contains reagents to examine the total protein levels of various PKC isoforms. The kit contains enough primary and secondary antibodies to perform four Western blots per primary antibody.

Source / Purification

Monoclonal antibody is produced by immunizing animals with a synthetic peptide corresponding to human PKCζ and to residues surrounding Arg216 of human PKCδ protein. Polyclonal antibodies are produced by immunizing animals with synthetic peptides corresponding to the sequence of the human protein PKCα and mouse protein PKD (PKCμ).

Antibodies are purified by protein A and peptide affinity chromatography.

Background

Activation of protein kinase C (PKC) is one of the earliest events in a cascade that controls a variety of cellular responses, including secretion, gene expression, proliferation, and muscle contraction (1,2). PKC isoforms belong to three groups based on calcium dependency and activators. Classical PKCs are calcium-dependent via their C2 domains and are activated by phosphatidylserine (PS), diacylglycerol (DAG), and phorbol esters (TPA, PMA) through their cysteine-rich C1 domains. Both novel and atypical PKCs are calcium-independent, but only novel PKCs are activated by PS, DAG, and phorbol esters (3-5). Members of these three PKC groups contain a pseudo-substrate or autoinhibitory domain that binds to substrate-binding sites in the catalytic domain to prevent activation in the absence of cofactors or activators. Control of PKC activity is regulated through three distinct phosphorylation events. Phosphorylation occurs *in vivo* at Thr500 in the activation loop, at Thr641 through autophosphorylation, and at the carboxy-terminal hydrophobic site

Ser660 (2). Atypical PKC isoforms lack hydrophobic region phosphorylation, which correlates with the presence of glutamic acid rather than the serine or threonine residues found in more typical PKC isoforms. The enzyme PDK1 or a close relative is responsible for PKC activation. A recent addition to the PKC superfamily is PKC μ (PKD), which is regulated by DAG and TPA through its C1 domain. PKD is distinguished by the presence of a PH domain and by its unique substrate recognition and Golgi localization (6). PKC-related kinases (PRK) lack the C1 domain and do not respond to DAG or phorbol esters. Phosphatidylinositol lipids activate PRKs, and small Rho-family GTPases bind to the homology region 1 (HR1) to regulate PRK kinase activity (7).

1. [Nishizuka, Y. \(1984\) *Nature* 308, 693-698.](#)
2. [Keranen, L.M. et al. \(1995\) *Curr. Biol.* 5, 1394-1403.](#)
3. [Mellor, H. and Parker, P.J. \(1998\) *Biochem J.* 332 \(Pt 2\), 281-292.](#)
4. [Ron, D. and Kazanietz, M.G. \(1999\) *FASEB J.* 13, 1658-1676.](#)
5. [Moscat, J. and Diaz-Meco, M.T. \(2000\) *EMBO Rep.* 1, 399-403.](#)
6. [Baron, C.L. and Malhotra, V. \(2002\) *Science* 295, 325-328.](#)
7. [Flynn, P. et al. \(2000\) *J. Biol. Chem.* 275, 11064-11070.](#)