# p-PKC θ (Ser 695): sc-33025



The Power to Question

## **BACKGROUND**

Members of the protein kinase C (PKC) family play a key regulatory role in a variety of cellular functions, including cell growth and differentiation, gene expression, hormone secretion and membrane function. PKCs were originally identified as serine/threonine protein kinases whose activity was dependent on calcium and phospholipids. Diacylglycerols (DAG) and tumor promoting phorbol esters bind to and activate PKC. PKCs can be subdivided into at least two major classes, including conventional (c) PKC isoforms ( $\alpha$ ,  $\beta$ I,  $\beta$ II and  $\gamma$ ) and novel (n) PKC isoforms ( $\delta$ ,  $\epsilon$ ,  $\omega$ ,  $\eta$  and  $\theta$ ). PKC isoforms can be activated through tyrosine phosphorylation and catalytically activated upon treatment with  $H_2O_2$ . The Tyr 155, 525, 523 and 565 residues in the catalytic domain are crucial for activation of these enzymes. The residue Ser 643 appears to be an autophosphorylation site. PKC  $\theta$  can undergo autophosphorylation on Serine 676 (Ser 676) in the turn loop and Serine 695 (Ser 695) in the hydrophobic loop. Phosphorylation of Serine 676 may negatively regulate activation of NFκB. Ser 695 is crucial to activate the phosphorylation threonine 692 (Thr 692) and Threonine 703 (Thr 703) residues, both of which are necessary for mobility shift.

# **REFERENCES**

- 1. Takai, Y., et al. 1979. Calcium-dependent activation of a multifunctional protein kinase by membrane phospholipids. J. Biol. Chem. 254: 3692-3695.
- Castagna, M., et al. 1982. Direct activation of calcium-activated, phospholipid-dependent protein kinase by tumor-promoting phorbol esters. J. Biol. Chem. 257: 7847-7851.
- Kikkawa, U., et al. 1983. Protein kinase C as a possible receptor of tumorpromoting phorbol esters. J. Biol. Chem. 258: 11442-11445.
- Nishizuka, Y. 1984. The role of protein kinase C in cell surface signal transduction and tumour promotion. Nature 308: 693-698.
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- 6. Osada, S., et al. 1992. A new member of the protein kinase C family, nPKC  $\theta$ , predominantly expressed in skeletal muscle. Mol. Cell. Biol. 12: 3930-3938.
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- 8. Parekh, D., et al. 1999. Mammalian TOR controls one of two kinase pathways acting upon nPKC  $\delta$  and nPKC  $\epsilon$ . J. Biol. Chem. 274: 34758-34764.
- 9. Konishi, H., et al. 2001. Phosphorylation sites of protein kinase C  $\delta$  in  $H_2O_2$ -treated cells and its activation by tyrosine kinase *in vitro*. Proc. Natl. Acad. Sci. USA 98: 6587-6592.

## CHROMOSOMAL LOCATION

Genetic locus: PRKCQ (human) mapping to 10p15.1; Prkcq (mouse) mapping to 2 A1.

## SOURCE

p-PKC  $\theta$  (Ser 695) is a rabbit polyclonal antibody raised against a short amino acid sequence containing Ser 695 phosphorylated PKC $\theta$  mouse origin.

#### **PRODUCT**

Each vial contains 200  $\mu g$  IgG in 1.0 ml of PBS with <0.1% sodium azide and 0.1% gelatin.

Blocking peptide available for competition studies, sc-33025 P, (100  $\mu$ g peptide in 0.5 ml PBS containing < 0.1% sodium azide and 0.2% BSA).

## **APPLICATIONS**

p-PKC  $\theta$  (Ser 695) is recommended for detection of Ser 695 phosphorylated p-PKC  $\theta$  of mouse, rat and human origin by Western Blotting (starting dilution 1:200, dilution range 1:100-1:1000), immunofluorescence (starting dilution 1:50, dilution range 1:50-1:500) and solid phase ELISA (starting dilution 1:30, dilution range 1:30-1:3000).

p-PKC  $\theta$  (Ser 695) is also recommended for detection of correspondingly phosphorylated PKC  $\theta$  in additional species, including equine and canine.

Suitable for use as control antibody for PKC  $\theta$  siRNA (h): sc-36252, PKC  $\theta$  siRNA (m): sc-36247, PKC  $\theta$  siRNA (r): sc-270095, PKC  $\theta$  shRNA Plasmid (h): sc-36252-SH, PKC  $\theta$  shRNA Plasmid (m): sc-36247-SH, PKC  $\theta$  shRNA Plasmid (r): sc-270095-SH, PKC  $\theta$  shRNA Lentiviral Particles (h): sc-36252-V, PKC  $\theta$  shRNA (m) Lentiviral Particles: sc-36247-V and PKC  $\theta$  shRNA (r) Lentiviral Particles: sc-270095-V.

Molecular Weight of p-PKC  $\theta$ : 82 kDa.

Positive Controls: Jurkat + anti-CD3 cell lysate: sc-24710 or Jurkat + PMA cell lysate: sc-24718.

## **RECOMMENDED SECONDARY REAGENTS**

To ensure optimal results, the following support (secondary) reagents are recommended: 1) Western Blotting: use goat anti-rabbit IgG-HRP: sc-2004 (dilution range: 1:2000-1:100,000) or Cruz Marker™ compatible goat anti-rabbit IgG-HRP: sc-2030 (dilution range: 1:2000-1:5000), Cruz Marker™ Molecular Weight Standards: sc-2035, TBS Blotto B Blocking Reagent: sc-2335 (use 50 mM NaF, sc-24988, as diluent), Western Blotting Luminol Reagent: sc-2048 and Lambda Phosphatase: sc-200312A. 2) Immunofluorescence: use goat anti-rabbit IgG-FITC: sc-2012 (dilution range: 1:100-1:400) or goat anti-rabbit IgG-TR: sc-2780 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941.

## **SELECT PRODUCT CITATIONS**

 Shyu, Y.C., et al. 2014. Tight regulation of a timed nuclear import wave of EKLF by PKCθ and FOE during Pro-E to Baso-E transition. Dev. Cell 28: 409-422.

#### **STORAGE**

Store at  $4^{\circ}$  C, \*\*DO NOT FREEZE\*\*. Stable for one year from the date of shipment. Non-hazardous. No MSDS required.

# **RESEARCH USE**

For research use only, not for use in diagnostic procedures.

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