

## Rabbit (polyclonal) Anti-PKCε[<sup>729</sup>P] Phosphospecific Antibody, Unconjugated

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Catalog Number: 44977G

Store at 2 to 8°C (short-term), or -20°C (long-term)

**Clonality:** Polyclonal  
**Quantity:** 10 mini-blot size  
**Volume:** 100 µL

**Host/Class:** Rabbit  
**Reactivity:** Human and mouse PKCε

### Product Description

Protein Kinase Cε (PKCε is an 87 kDa (migrates predominantly as 95 kDa in quiescent cells) member of the novel group (nPKCs: sensitive to diacylglycerol, phosphatidylserine, and phorbol esters) of the PKC family of serine/threonine kinases that are involved in a wide range of physiological processes including mitogenesis, cell survival, metastasis and transcriptional regulation. PKCε is an actin cytoskeleton regulating protein whose overexpression confers an oncogenic phenotype by affecting the ras signaling cascade. PKCε is phosphorylated by PDK1 on threonine 566 and is then autophosphorylated on the turn motif threonine 710 and the hydrophobic motif serine 729. Catalytically inactive PKCε is not phosphorylated on serine 729. Phosphorylation of serine 729 is mediated by mTOR-dependent pathways, plays an important role in regulating PKCε subcellular localization, and is responsible for the appearance of the 92 kDa PKCε species. During development reactivity was confirmed with human and mouse PKCε. This antibody may partially cross react with βII [pS<sup>660</sup>] as determined by peptide competition experiments.

### Product Specifications

**Immunogen:** The antiserum was produced against a chemically synthesized phosphopeptide derived from a region of human PKCε that contains serine 729. The sequence is conserved in mouse and rat.

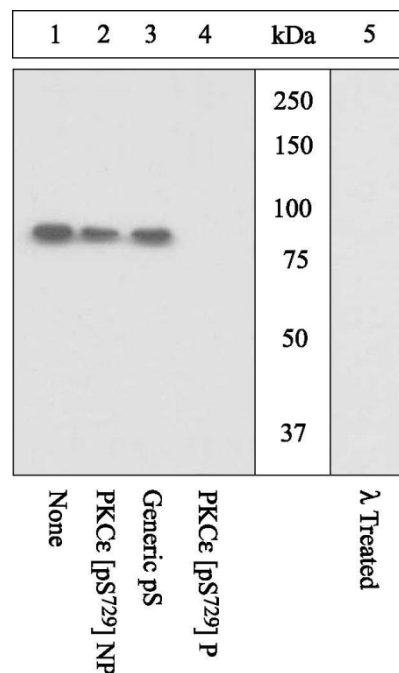
**Lot:** See product label

### Product Applications

Application	Species	Test Material	Concentration
Western Blotting	Mouse	NIH3T3 cells treated with PDGF	1:1000
	Human	K562 cells treated with PMA; Recombinant PKC Epsilon Protein	

### Storage and Handling

Store at -20°C. We recommend a brief centrifugation before opening to settle vial contents. Then, apportion into working aliquots and store at -20°C. For short term storage (up to one week), 2 to 8°C is sufficient.



**Figure 1 Peptide Competition and Phosphatase Treatment**

Lysates prepared from K562 cells stimulated with PMA were resolved by SDS-PAGE on a 10% polyacrylamide gel and transferred to PVDF. Membranes were either left untreated (lanes 1-4) or treated with lambda (λ) phosphatase (lane 5), blocked with a 3% BSA-TBST buffer for one hour at room temperature, and incubated with PKCε [pS<sup>729</sup>] antibody for two hours at room temperature in a 3% BSA-TBST buffer, following prior incubation with: no peptide (lanes 1, 5), the non-phosphopeptide corresponding to the immunogen (lane 2), a generic phosphoserine-containing peptide (lane 3), or the phosphopeptide immunogen (lane 4). After washing, membranes were incubated with goat F(ab')<sub>2</sub> anti-rabbit IgG HRP conjugate and bands were detected using the Pierce SuperSignal<sup>®</sup> method.

The data show that only the peptide corresponding to PKCε [pS<sup>729</sup>] blocks the antibody signal. The data also show that phosphatase stripping eliminates the signal, verifying that the antibody is phospho-specific.

**For research use only. Not for use in diagnostic procedures.**

Manufacturing Site • 7335 Executive Way • Frederick • MD 21704 • E-mail: [techsupport@lifetech.com](mailto:techsupport@lifetech.com)

## Stability

When stored as instructed, expires one year from date of receipt unless otherwise indicated on product label.

## Storage Buffer

Dulbecco's phosphate buffered saline (without  $Mg^{2+}$  and  $Ca^{2+}$ ), pH 7.3 ( $\pm 0.1$ ), 50% glycerol with 1.0 mg/mL BSA (IgG, protease free) as a carrier. Contains 0.05% sodium azide.

**Caution:** Sodium azide is an extremely toxic and dangerous compound particularly when combined with acids or metals. Properly dispose of solutions containing sodium azide.








## Safety Data Sheets (SDS)

Safety Data Sheets (SDSs) are available at [www.lifetechnologies.com/support](http://www.lifetechnologies.com/support).

## Certificate of Analysis

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### Explanation of symbols

Symbol	Description	Symbol	Description	Symbol	Description
	Manufacturer		Catalog number		Batch code
	Use by		Temperature limitation		
	Consult instructions for use		Caution, consult accompanying documents		

### Limited Use Label License: Research Use Only

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