Phospho-PKC (pan) (gamma Thr514) (PF4) rabbit mAb

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Applications	Detection	Clonality	Isotype
Flow Cytometry,WB	Anti-Rabbit IgG	Monoclonal	Rabbit IgGk

Format: Unconjugated

Cross Reactivity: Predicted to work with mouse, rat and other homologues.

Formulation: 1X PBS, 0.02% NaN3, 50% Glycerol, 0.1% BSA

Preparation: Protein A+G

Reactivity: Human,Mouse,Rat

Recommended Usage: 1µg/mL – 0.001µg/mL. It is recommended that the reagent be titrated for optimal performance for each application. See product image legends for additional information.

Immunogen: A synthetic phospho-peptide corresponding to residues surrounding Thr514 of human phospho PKCγ

Description: Protein Kinase (PKC) is a 12 member family of serine/threonine kinases termed conventional or classical (a b, g), novel (d,e,and q), atypical (z, l) and PKN and PKC-related (PKN1, PKN2 and PKN3) playing significant role in several signaling processes involved in physiological and pathological setting (1). PKC activation translates into gene expression modulation, cell division, migration, proliferation, differentiation, and cell survival and apoptosis (2). PKC members are classified based on their distinct cofactor requirements and the extent of homology between their regulatory elements (3,4). PKCalpha, beta I, beta II, and gamma constitute the conventional PKC isoforms, characterized by the presence of two cysteine-rich zinc finger domains, C1a and C1b (5,6) which bind to diacylglycerol (DAG) and phosphatidylserine (PS) (7). In addition, PKC contains a C2 domain, responsible for binding anionic phospholipids like phosphatidylinositol bisphosphate (PIP2) in a Ca^{2+}-dependent manner (8,9). The atypical PKC, PKCz andl share ATP-binding domain and the catalytic domain with PKC. They contain a single C1 domain that lacks residues necessary for binding DAG (10).

Association of PKCg to the membrane enables a conformation that permits phosphoinositide-dependent protein kinase1 (PDK-1) (11) to bind and phosphorylate a site in the activation loop, Thr514. (12,13). Thr514 phosphorylation leads to a conformation change enabling phosphorylation of at two carboxyl-terminal sites namely, the turn motif and hydrophobic motif, as a result of which the fully phosphorylated conventional PKC is released from the membrane, and positioned in the cytoplasm as an inactive form (14,15). Binding of Ca^{2+} induces low-affinity interaction with the membrane, whereas the membrane imbedded cofactor DAG to PKC results in high-affinity interaction of PKC with the membrane (16).

References:

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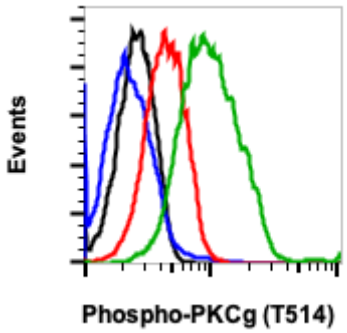
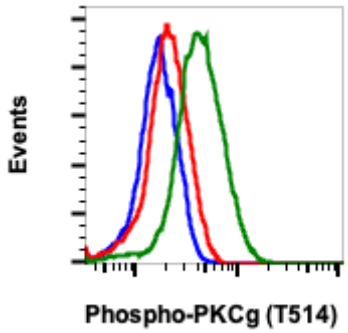
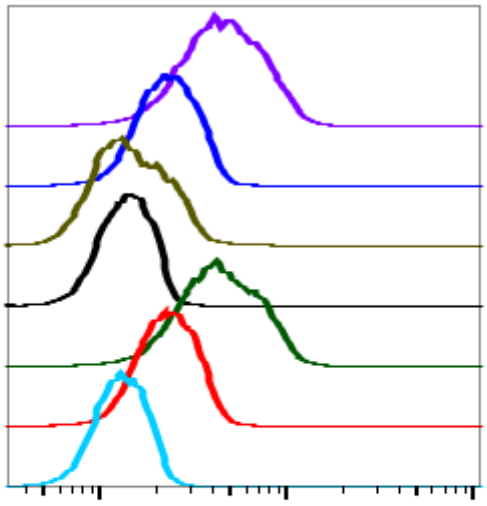


Flow cytometric analysis of HT1080 cells treated with staurosporine (red) or untreated (green) using Phospho-PKCg (Thr514) (PF4) Rabbit mAb. at 0.05 ug/mL, PKCgT514-PF4 #2451, or concentration-matched Rabbit (G9) mAb IgG Isotype Control #2141 for cells treated with staurosporine (black) or untreated (blue). Flow cytometric analysis of HT1080 cells, treated with staurosporine and stained with the secondary antibody only as negative control (blue) or treated with staurosporine (red) or untreated (green) using Phospho-PKC (pan) (gamma Thr514) antibody PKCgT514-PF4 at 0.05 µg/mL. Cat. #2451.

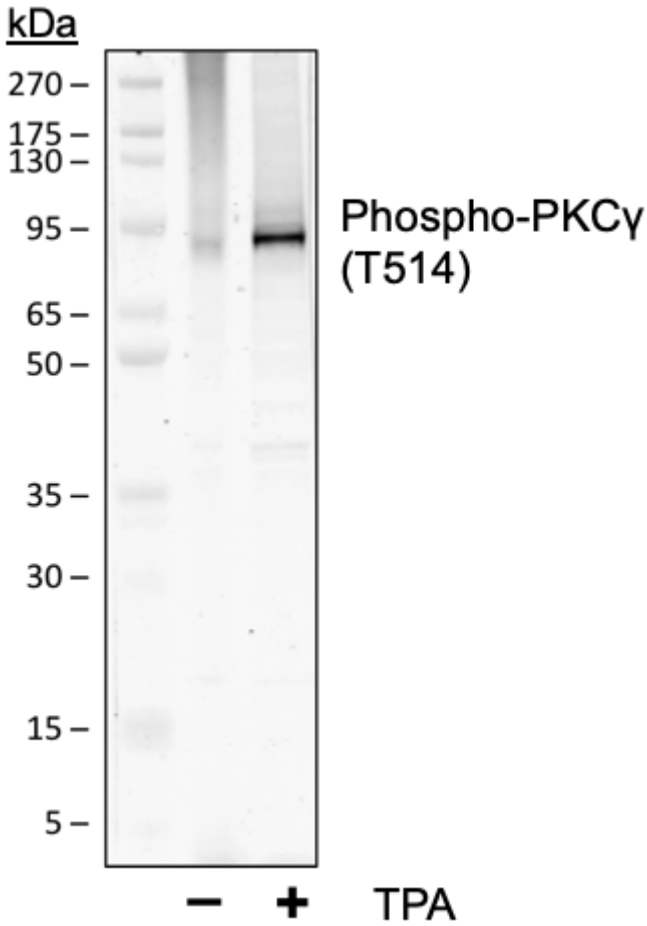


Flow cytometric analysis of Jurkat cells, treated with K252a and stained with the secondary antibody only negative control (blue) or treated with K252a (red) or untreated (green) using Phospho-PKC (pan) (gamma Thr514) antibody PKCgT514-PF4 at 0.1 µg/mL. Cat. #2451.

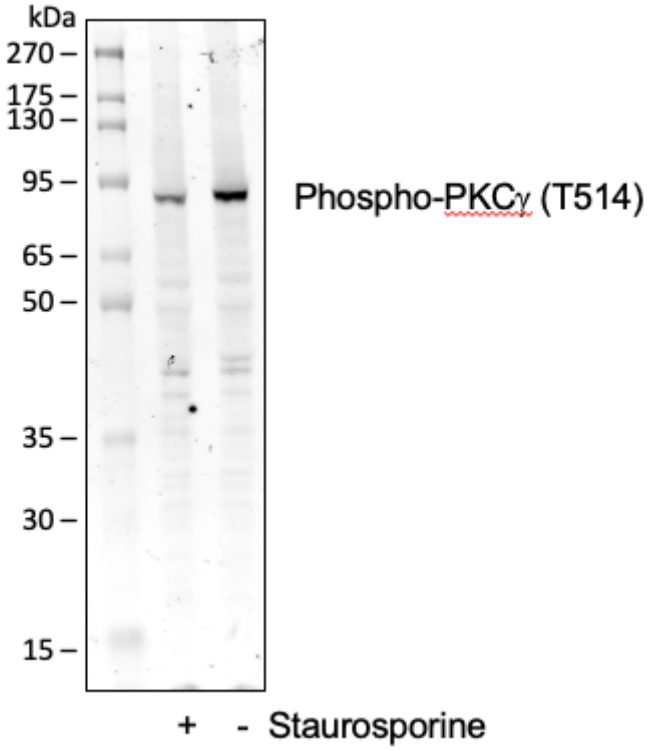


	$WELLID	Treatment	Median : BL1-A
☐	PF4 0.05+NP	Ctrl	4500
☐	PF4 0.05+NP	STAURO	2279
☐	PF4 0.05+PP	Ctrl	1409
☐	PF4 0.05+PP	STAURO	1378
☐	PF4 0.05	Ctrl	4278
☐	PF4 0.5	STAURO	2279
☐	2'AB	STAURO	1292

Peptide blocking flow cytometric analysis of HT1080 cells secondary antibody only negative control (light blue) or untreated (red) or treated with staurosporine (green) or untreated and blocked with phospho-peptide (black) or treated with staurosporine and blocked with phospho peptide (gold) or untreated and blocked with non-phospho peptide (dark blue) or treated with staurosporine and blocked with non-phospho peptide (purple) using Phospho-PKCg (Thr514) antibody PKCGT514-PF4 at 0.05 µg/mL. Cat. #2451.



Western blot analysis of NIH3T3 cell extract untreated or treated with TPA using Phospho-PKC (pan) (gamma Thr514) antibody PKCgT514-PF4 at 0.05 ug/mL. Cat. #2451.



Western blot analysis of C6 cell extract untreated or treated with staurosporine using Phospho-PKC (pan) gamma (Thr514) antibody PKCgT514-PF4 at 0.1 ug/mL. Cat. #2451.