bio-techne® RD SYSTEMS

Human/Mouse/Rat PKCα Antibody

Antigen Affinity-purified Polyclonal Goat IgG Catalog Number: AF5340

DESCRIPTION	
Species Reactivity	Human/Mouse/Rat
Specificity	Detects endogenous human, mouse, and rat PKCα in Western blots.
Source	Polyclonal Goat IgG
Purification	Antigen Affinity-purified
Immunogen	<i>E. coli</i> -derived recombinant human PKCα Lys604-Val672 Accession # P17252
Formulation	Lyophilized from a 0.2 µm filtered solution in PBS with Trehalose. See Certificate of Analysis for details. *Small pack size (-SP) is supplied either lyophilized or as a 0.2 µm filtered solution in PBS.

APPLICATIONS

Please Note: Optimal dilutions should be determined by each laboratory for each application. General Protocols are available in the Technical Information section on our website.

	Recommended Concentration	Sample
Western Blot	1 μg/mL	See Below
Immunohistochemistry	5-15 μg/mL	See Below
Simple Western	10 μg/mL	Saos-2 human osteosarcoma cell line

DATA



Detection of Human/Mouse/Rat PKCα by Western Blot. Western blot shows lysates of HeLa human cervical epithelial carcinoma cell line, Saos-2 human osteosarcoma cell line. Balb/3T3 mouse embryonic fibroblast cell line, and NRK rat normal kidney cell line. PVDF membrane was probed with 1 µg/mL of Human/Mouse/Rat PKCa Antigen Affinity-purified Polyclonal Antibody (Catalog # AF5340) followed by HRPconjugated Anti-Goat IgG Secondary Antibody (Catalog # Catalog # HAF109). A specific band was detected for PKCa at approximately 90 kDa (as indicated). This experiment was conducted under reducing conditions and using Immunoblot Buffer Group 1.

Immunohistochemistrv



PKCα in Human Breast Cancer Tissue. PKCa was detected in immersion fixed paraffinembedded sections of human breast cancer tissue using Goat Anti-Human/Mouse/Rat PKCa Antigen Affinity-purified Polyclonal Antibody (Catalog # AF5340) at 10 µg/mL overnight at 4 °C. Tissue was stained using the Anti-Goat HRP-DAB Cell & Tissue Staining Kit (brown; Catalog # Catalog # CTS008) and counterstained with hematoxylin (blue). Specific staining was localized to cancer cell cytoplasm. View our protocol for Chromogenic IHC Staining of Paraffin-embedded Tissue Sections.

Simple Western



Detection of Human PKCα by Simple Western ^M. Simple Western shows lysates of Saos-2 human osteosarcoma cell line, loaded at 0.5 mg/ml. A specific band was detected for PKCα at approximately 84 kDa (as indicated) using 10 µg/mL of Goat Anti-Human/Mouse/Rat PKCα Antigen Affinity-purified Polyclonal Antibody (Catalog # AF5340). This experiment was conducted under reducing conditions and using the 12-230kDa separation system.

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PREPARATION AND STORAGE		
Reconstitution	Reconstitute at 0.2 mg/mL in sterile PBS. For liquid material, refer to CoA for concentration.	
Shipping	Lyophilized product is shipped at ambient temperature. Liquid small pack size (-SP) is shipped with polar packs. Upon receipt, store immediately at the temperature recommended below.	
Stability & Storage	 Use a manual defrost freezer and avoid repeated freeze-thaw cycles. 12 months from date of receipt, -20 to -70 °C as supplied. 1 month, 2 to 8 °C under sterile conditions after reconstitution. 6 months, -20 to -70 °C under sterile conditions after reconstitution. 	

BACKGROUND

PKCα (Protein kinase C alpha) is an 80 kDa member of the PKC subfamily, AGC Ser/Thr protein kinase family of enzymes. It is widely expressed, and serves multiple cell-specific functions. PKCa is activated by increased diacylglycerol and intracellular calcium, and translocates to several sites such as the Golgi, nucleus and plasma membrane. Human PKCa is 672 amino acids (aa) in length. It contains two zinc finger regions (aa 36-151), a protein kinase domain (aa 339-557), and an AGC kinase region (aa 598-668). Phosphorylations on Thr497, Thr638 and Ser657 are necessary for kinase activity. Over aa 604-672, human PKCa shows 100% aa identity to mouse PKCa.