

Anti-Phospho-PKC alpha (Thr638) Antibody (5U868)

Product Details

Ig Type:	IgG
Reactivity:	Human,Mouse,Rat
Molecular Weight:	Theoretical: 77 kDa. Actual: 80 kDa.
Clone:	5U868
Purification:	Protein A purified

Applications

1. Blank control: MCF7.

Primary Antibody (green line): Rabbit Anti-Phospho-PKC alpha (Thr638) antibody (TMAB-11147)

Dilution: 2 µg /10⁶ cells;

Isotype Control Antibody (orange line): Rabbit IgG.

Secondary Antibody: Goat anti-rabbit IgG-AF488

Dilution: 1 µg /test.

Protocol

The cells were fixed with 4% PFA (10 min at room temperature) and then permeabilized with 90% ice-cold methanol for 20 min at -20°C. The cells were then incubated in 5% BSA to block non-specific protein-protein interactions for 30 min at room temperature. Cells stained with Primary Antibody for 30 min at room temperature. The secondary antibody used for 40 min at room temperature. Acquisition of 20,000 events was performed.

2. Paraformaldehyde-fixed, paraffin embedded Mouse Liver; Antigen retrieval by boiling in sodium citrate buffer (pH6.0) for 15 min; The section was incubated with Phospho-PKC alpha (Thr638) Monoclonal Antibody, Unconjugated (TMAB-11147) at 1: 200 overnight at 4°C, followed by conjugation to the Goat Anti-Rabbit IgG H&L Secondary Antibody-HRP and DAB staining.

3. Paraformaldehyde-fixed, paraffin embedded Rat Liver; Antigen retrieval by boiling in sodium citrate buffer (pH6.0) for 15 min; The section was incubated with Phospho-PKC alpha (Thr638) Monoclonal Antibody, Unconjugated (TMAB-11147) at 1: 200 overnight at 4°C, followed by conjugation to the Goat Anti-Rabbit IgG H&L Secondary Antibody-HRP and DAB staining.

4. Paraformaldehyde-fixed, paraffin embedded Mouse Cerebrum; Antigen retrieval by boiling in sodium citrate buffer (pH6.0) for 15 min; The section was incubated with Phospho-PKC alpha (Thr638) Monoclonal Antibody, Unconjugated (TMAB-11147) at 1: 200 overnight at 4°C, followed by conjugation to the Goat Anti-Rabbit IgG H&L Secondary Antibody-HRP and DAB staining.

5. Paraformaldehyde-fixed, paraffin embedded Rat Cerebrum; Antigen retrieval by boiling in sodium citrate buffer (pH6.0) for 15 min; The section was incubated with Phospho-PKC alpha (Thr638) Monoclonal Antibody, Unconjugated (TMAB-11147) at 1: 200 overnight at 4°C, followed by conjugation to the Goat Anti-Rabbit IgG H&L Secondary Antibody-HRP and DAB staining.

6. Paraformaldehyde-fixed, paraffin embedded Human Cerebrum; Antigen retrieval by boiling in sodium citrate buffer (pH6.0) for 15 min; The section was incubated with Phospho-PKC alpha (Thr638) Monoclonal Antibody, Unconjugated (TMAB-11147) at 1: 200 overnight at 4°C, followed by conjugation to the Goat Anti-Rabbit IgG H&L Secondary Antibody-HRP and DAB staining.

Verified Activity:

7. Paraformaldehyde-fixed, paraffin embedded Mouse Spleen; Antigen retrieval by boiling in sodium citrate buffer (pH6.0) for 15 min; The section was incubated with Phospho-PKC alpha (Thr638) Monoclonal Antibody, Unconjugated (TMAB-11147) at 1: 200 overnight at 4°C, followed by conjugation to the Goat Anti-Rabbit IgG H&L Secondary Antibody-HRP and DAB staining.
8. Paraformaldehyde-fixed, paraffin embedded Rat Spleen; Antigen retrieval by boiling in sodium citrate buffer (pH6.0) for 15 min; The section was incubated with Phospho-PKC alpha (Thr638) Monoclonal Antibody, Unconjugated (TMAB-11147) at 1: 200 overnight at 4°C, followed by conjugation to the Goat Anti-Rabbit IgG H&L Secondary Antibody-HRP and DAB staining.
9. Paraformaldehyde-fixed, paraffin embedded Human Spleen; Antigen retrieval by boiling in sodium citrate buffer (pH6.0) for 15 min; The section was incubated with Phospho-PKC alpha (Thr638) Monoclonal Antibody, Unconjugated (TMAB-11147) at 1: 200 overnight at 4°C, followed by conjugation to the Goat Anti-Rabbit IgG H&L Secondary Antibody-HRP and DAB staining.
10. Paraformaldehyde-fixed, paraffin embedded Human Glioma; Antigen retrieval by boiling in sodium citrate buffer (pH6.0) for 15 min; The section was incubated with Phospho-PKC alpha (Thr638) Monoclonal Antibody, Unconjugated (TMAB-11147) at 1: 200 overnight at 4°C, followed by conjugation to the Goat Anti-Rabbit IgG H&L Secondary Antibody-HRP and DAB staining.
11. Paraformaldehyde-fixed, paraffin embedded Human Breast Cancer; Antigen retrieval by boiling in sodium citrate buffer (pH6.0) for 15 min; The section was incubated with Phospho-PKC alpha (Thr638) Monoclonal Antibody, Unconjugated (TMAB-11147) at 1: 200 overnight at 4°C, followed by conjugation to the Goat Anti-Rabbit IgG H&L Secondary Antibody-HRP and DAB staining.
12. 25 µg total protein per lane of various lysates (see on figure) probed with p-PKC alpha (T638) monoclonal antibody, unconjugated (TMAB-11147) at 1:1000 dilution and 4°C overnight incubation. Followed by conjugated secondary antibody incubation at r. T. for 60 min.

Application: WB,IHC-P,IHC-Fr,ICC/IF,IF,FCM

Recommended WB: 1:500-2000; IHC-P: 1:50-200; IHC-Fr: 1:50-200; ICC/IF: 1:50-200; IF: 1:50-200; FCM: 1:50-100

Properties

Stability & Storage: Store at 2°C-8°C for 1 month. Store at -20°C or -80°C for 12 months. Avoid repeated freeze-thaw cycles.

Shipping: Shipping with blue ice.

Antigen Details

Immunogen: KLH conjugated synthesised phosphopeptide: human PKC alpha around the phosphorylation site of Thr638

Antigen Species: Human

Gene ID: 5578

Uniprot ID: P17252

Research Background

Protein Kinase c alpha (PKC alpha) is an 77 kDa member of the conventional group (cPKCs: sensitive to calcium, 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 and transcriptional regulation. PKC alpha is an ubiquitously expressed PKC isozyme that has been implicated in the regulation of a broad range of cellular functions including proliferation, differentiation, development, migration, cell cell adhesion,

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cell extracellular matrix adhesion, and solute transport. The activation loop threonine (threonine 497 in PKC alpha) of conventional PKCs is phosphorylated by phosphoinositide dependent kinase 1 (PDK1). This phosphorylation is necessary for the autophosphorylation of threonine 638 in the carboxy terminus of PKC alpha, a step that is critical for regulating the rate of PKC alpha dephosphorylation and inactivation.

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Tel:781-999-4286 E_mail:info@targetmol.com Address:34 Washington Street,Wellesley Hills,MA 02481