## For Research Use Only

# AKT Substrate Essentials Antibody Kit

Catalog Number: PK30021



www.ptglab.com

#### Description

The AKT Substrate Essentials Antibody Kit provides a cost-effective tool for studying key proteins that are phosphorylated by AKT. Perfect for signal transduction researchers starting a new project, screening multiple prospective targets, or those who simply require less volume.

#### **Product Information**

The AKT Substrate Essentials Kit contains 5 antibodies against downstream phosphorylation targets of AKT.

Antigen	Catalog No.	Host, clonality	Tested Reactivity	Applications	Volume
Phospho-GSK3B (Ser9)	<u>67558-1-lg</u>	Mouse monoclonal	Н	WB, IHC, IF	20 uL
Phospho-mTOR (Ser2448)	80596-1-RR	Rabbit Monoclonal	H, R	WB, IF	20 uL
Phospho-TSC2 (Thr1462)	80698-1-RR	Rabbit Monoclonal	Н	WB	20 uL
Phospho-TSC2 (Ser939)	81654-1-RR	Rabbit Monoclonal	Н	WB	20 uL
Phospho-FOXO1 (Ser319)	28757-1-AP	Rabbit Polyclonal	Н	WB	20 uL

Also see our 'AKT Substrate Expanded Antibody Kit' on the following page

https://www.ptglab.com/products/AKT-Substrate-Expanded-Antibody-Kit-PK30022.htm

**Package** 

5×20 uL

Storage

Store at -20°C. Stable for one year from the date of receipt.

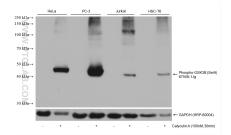
**Background Information** 

Upon its activation, AKT phosphorylates a number of different substrates, resulting in the activation or inactivation of downstream cellular functions. The phosphorylation of GSK3B at Ser9, inhibits its activity and subsequent glycogen synthesis. Meanwhile, AKT-mediated phosphorylation of mTOR at Ser2448 results in its activation and formation of the mTORC1 complex. TSC2, and inhibitor of mTORC1 activation, is blocked through phosphorylation at Thr1462 and Ser939 residues. Finally, FOXO1, a transcription factor invovled in cell cycle arrest and apoptosis, is inhibited by AKT through phosphorylation at Ser319, which leads to its nuclear export.

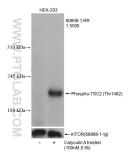
**Standard Protocols** 

Click <a href="here">here</a> to view our standard protocols for various applications including WB, IP, IHC, IF, FC, and ELISA.

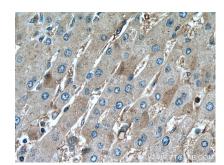
### Validation Data



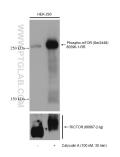
Non-treated and Calyculin A treated cell lysates were subjected to SDS PAGE followed by western blot with 67558-1-lg (Phospho-GSK3B (Ser9) antibody) at dilution of 1:5000 incubated at room temperature for 1.5 hours. The membrane was stripped and reblotted with HRP-conjugated GAPDH Monoclonal antibody (HRP-60004) as loading... control.



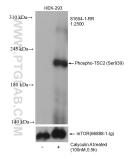
Non-treated and Calyculin A treated HEK-293 cells were subjected to SDS PAGE followed by western blot with 80698-1-RR (Phospho-TSC2 (Thr1462) antibody) at dilution of 1:5000 incubated at room temperature for 1.5 hours. The membrane was stripped and re-blotted with mTOR antibody (66888-1-lg) as the loading control.



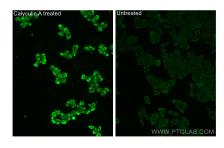
Immunohistochemical analysis of paraffinembedded human liver cancer tissue slide using 67558-1-lg (GSK3B-phospho-59 antibody) at dilution of 1:300 (under 40x lens). Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0).



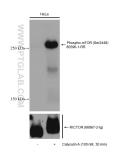
Non-treated and Calyculin A treated HEK-293 cells were subjected to SDS PAGE followed by western blot with 80596-1-RR (Phospho-mTOR (Ser2448) antibody) at dilution of 1:10000 incubated at room temperature for 1.5 hours. The membrane was stripped and re-blotted with RICTOR antibody (66867-2-lg) subsequently.



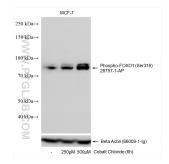
Non-treated and Calyculin A treated HEK-293 cells were subjected to SDS PAGE followed by western blot with 81654-1-RR (Phospho-TSC2 (Ser939) antibody) at dilution of 1:2500 incubated at room temperature for 1.5 hours. The membrane was stripped and re-blotted with mTOR antibody (66888-1-Ig) as the loading control.



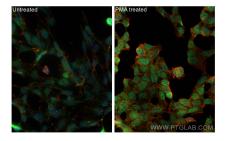
Immunofluorescent analysis of (-20°C Ethanol) fixed Calyculin A treated PC-3 cells and nontreated PC-3 cells using Phospho-GSK3B (Ser9) antibody (67558-1-lg, Clone: 1C9E2) at dilution of 1:200 and CoraLite@488-Conjugated AffiniPure Goat Anti-Mouse IgG(H+L).



Non-treated and Calyculin A treated HeLa cells were subjected to SDS PAGE followed by western blot with 80596-1-RR (Phospho-mTOR (Ser2448) antibody) at dilution of 1:10000 incubated at room temperature for 1.5 hours. The membrane was stripped and re-blotted with RICTOR antibody (66867-2-lg) subsequently.



Non-treated MCF-7 cells and Cobalt Chloride treated MCF-7 cells were subjected to SDS PAGE followed by western blot with 28757-1-AP (Phospho-FOXO1 (Ser319) antibody) at dilution of 1:500 incubated at room temperature for 1.5 hours. The membrane was stripped and re-blotted with Beta Actin antibody as loading control.



Immunofluorescent analysis of (4% PFA) fixed PMA treated HEK-293 cells using Phospho-mTOR (Ser2448) antibody (80596-1-RR, Clone: 3L18) at dilution of 1:1000 and Coralite® 488-Conjugated AffiniPure Goat Anti-Rabbit IgG(H+L), CL594-phalloidin (red).