

**SPAK Antibody (Center)**  
**Purified Rabbit Polyclonal Antibody (Pab)**  
**Catalog # AP7968c****Specification**

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**SPAK Antibody (Center) - Product Information**

Application	<b>WB, IHC-P,E</b>
Primary Accession	<a href="#">O9UEW8</a>
Other Accession	<a href="#">O88506</a> , <a href="#">O9Z1W9</a>
Reactivity	<b>Human, Mouse</b>
Predicted	<b>Rat</b>
Host	<b>Rabbit</b>
Clonality	<b>Polyclonal</b>
Isotype	<b>Rabbit IgG</b>
Antigen Region	<b>346-376</b>

**SPAK Antibody (Center) - Additional Information****Gene ID** 27347**Other Names**

STE20/SPS1-related proline-alanine-rich protein kinase, Ste-20-related kinase, DCHT, Serine/threonine-protein kinase 39, STK39, SPAK

**Target/Specificity**

This SPAK antibody is generated from rabbits immunized with a KLH conjugated synthetic peptide between 346-376 amino acids from the Central region of human SPAK.

**Dilution**WB~~1:1000  
IHC-P~~1:50~100**Format**

Purified polyclonal antibody supplied in PBS with 0.09% (W/V) sodium azide. This antibody is prepared by Saturated Ammonium Sulfate (SAS) precipitation followed by dialysis against PBS.

**Storage**

Maintain refrigerated at 2-8°C for up to 2 weeks. For long term storage store at -20°C in small aliquots to prevent freeze-thaw cycles.

**Precautions**

SPAK Antibody (Center) is for research use only and not for use in diagnostic or therapeutic procedures.

**SPAK Antibody (Center) - Protein Information****Name** STK39

**Function** Effector serine/threonine-protein kinase component of the WNK-SPAK/OSR1 kinase cascade, which is involved in various processes, such as ion transport, response to hypertonic stress and blood pressure (PubMed:[16669787](#), PubMed:[18270262](#), PubMed:[21321328](#), PubMed:[34289367](#)). Specifically recognizes and binds proteins with a RFXV motif (PubMed:[16669787](#), PubMed:[21321328](#)). Acts downstream of WNK kinases (WNK1, WNK2, WNK3 or WNK4): following activation by WNK kinases, catalyzes phosphorylation of ion cotransporters, such as SLC12A1/NKCC2, SLC12A2/NKCC1, SLC12A3/NCC, SLC12A5/KCC2 or SLC12A6/KCC3, regulating their activity (PubMed:[21321328](#)). Mediates regulatory volume increase in response to hyperosmotic stress by catalyzing phosphorylation of ion cotransporters SLC12A1/NKCC2, SLC12A2/NKCC1 and SLC12A6/KCC3 downstream of WNK1 and WNK3 kinases (PubMed:[12740379](#), PubMed:[16669787](#), PubMed:[21321328](#)). Phosphorylation of Na-K-Cl cotransporters SLC12A2/NKCC1 and SLC12A2/NKCC1 promote their activation and ion influx; simultaneously, phosphorylation of K-Cl cotransporters SLC12A5/KCC2 and SLC12A6/KCC3 inhibit their activity, blocking ion efflux (PubMed:[16669787](#), PubMed:[19665974](#), PubMed:[21321328](#)). Acts as a regulator of NaCl reabsorption in the distal nephron by mediating phosphorylation and activation of the thiazide-sensitive Na-Cl cotransporter SLC12A3/NCC in distal convoluted tubule cells of kidney downstream of WNK4 (PubMed:[18270262](#)). Mediates the inhibition of SLC4A4, SLC26A6 as well as CFTR activities (By similarity). Phosphorylates RELT (By similarity).

#### **Cellular Location**

Cytoplasm. Nucleus. Note=Nucleus when caspase-cleaved.

#### **Tissue Location**

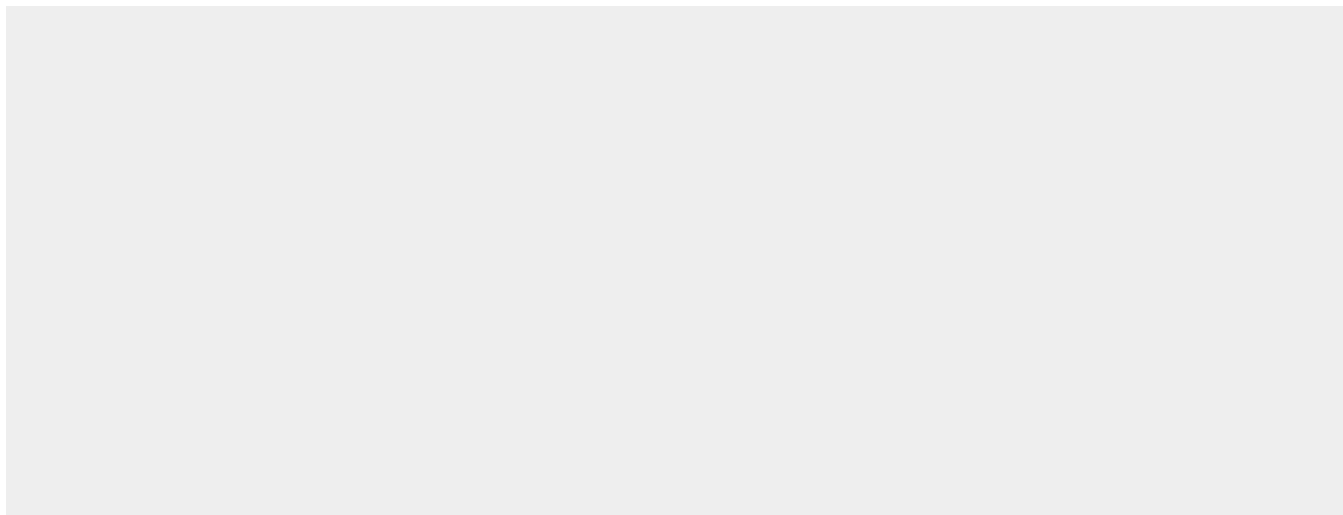
Predominantly expressed in brain and pancreas followed by heart, lung, kidney, skeletal muscle, liver, placenta and testis.

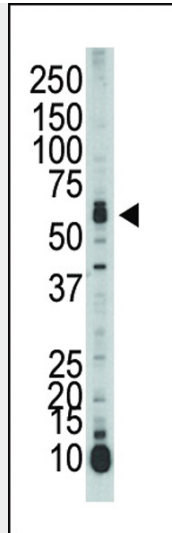
### **SPAK Antibody (Center) - Protocols**

Provided below are standard protocols that you may find useful for product applications.

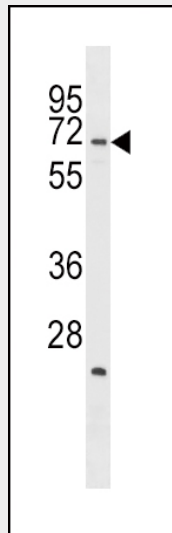
- [Western Blot](#)
- [Blocking Peptides](#)
- [Dot Blot](#)
- [Immunohistochemistry](#)
- [Immunofluorescence](#)
- [Immunoprecipitation](#)
- [Flow Cytometry](#)
- [Cell Culture](#)

### **SPAK Antibody (Center) - Images**

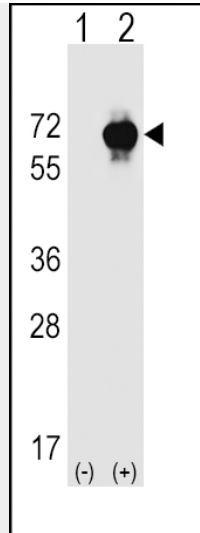




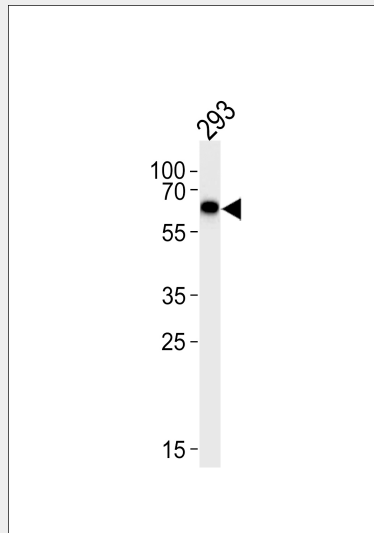
Western blot analysis of anti-SPAK Pab (Cat. #AP7968c) in mouse liver tissue lysate. SPAK (arrow) was detected using purified Pab. Secondary HRP-anti-rabbit was used for signal visualization with chemiluminescence.



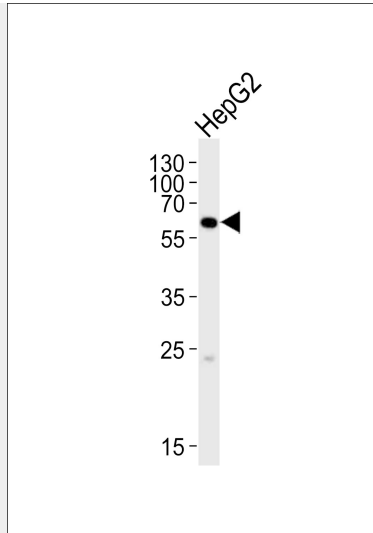
SPAK Antibody (A363) (Cat. #AP7968c) western blot analysis in U937 cell line lysates (35ug/lane). This demonstrates the SPAK antibody detected the SPAK protein (arrow).



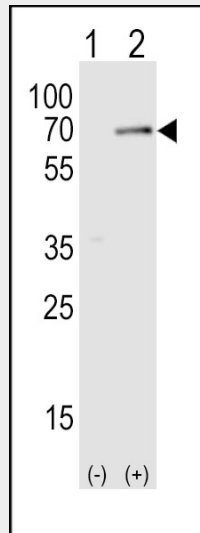
Western blot analysis of SPAK (arrow) using rabbit polyclonal SPAK Antibody (A363) (Cat. #AP7968c). 293 cell lysates (2 ug/lane) either nontransfected (Lane 1) or transiently transfected (Lane 2) with the SPAK gene.



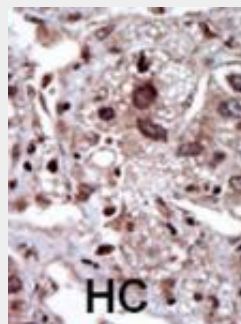
Western blot analysis of lysate from 293 cell line, using SPAK Antibody (A363)(Cat. #AP7968c). AP7968c was diluted at 1:1000 at each lane. A goat anti-rabbit IgG H&L(HRP) at 1:5000 dilution was used as the secondary antibody. Lysate at 35ug.



Western blot analysis of lysate from HepG2 cell line, using SPAK Antibody (A363)(Cat. #AP7968c). AP7968c was diluted at 1:1000. A goat anti-rabbit IgG H&L(HRP) at 1:10000 dilution was used as the secondary antibody. Lysate at 20ug.



Western blot analysis of SPAK (arrow) using rabbit polyclonal SPAK Antibody (A363) (Cat. #AP7968c). 293T cell lysates either nontransfected (Lane 1) or transiently transfected (Lane 2) with the SPAK gene.



Formalin-fixed and paraffin-embedded human cancer tissue reacted with the primary antibody, which was peroxidase-conjugated to the secondary antibody, followed by DAB staining. This data demonstrates the use of this antibody for immunohistochemistry; clinical relevance has not been evaluated. BC = breast carcinoma; HC = hepatocarcinoma.

## **SPAK Antibody (Center) - Background**

SPAK is a serine/threonine kinase containing an N-terminal series of proline and alanine repeats (PAPA box), followed by a serine/threonine kinase catalytic domain, a nuclear localization signal, a consensus caspase cleavage recognition motif, and a C-terminal region. Northern blot analysis detects ubiquitous expression, most abundantly in brain and pancreas.

SPAK can phosphorylate itself and an exogenous substrate *in vitro*. SPAK immunoprecipitates from transfected mammalian cells in a complex with another serine/threonine kinase that phosphorylates catalytically inactive SPAK. SPAK activates the p38 MAP kinase pathway in cotransfection assays. Full-length SPAK is expressed in the cytoplasm in transfected cells, while a mutant corresponding to caspase-cleaved STK39 localizes predominantly in the nucleus.

## **SPAK Antibody (Center) - References**

Dowd, B.F., et al., *J. Biol. Chem.* 278(30):27347-27353 (2003).

Johnston, A.M., et al., *Oncogene* 19(37):4290-4297 (2000).

### **SPAK Antibody (Center) - Citations**

- [STE20/SPS1-related proline/alanine-rich kinase is involved in plasticity of GABA signaling function in a mouse model of acquired epilepsy.](#)
- [From the Cover: Whole-genome association study identifies STK39 as a hypertension susceptibility gene.](#)
- [PKCdelta acts upstream of SPAK in the activation of NKCC1 by hyperosmotic stress in human airway epithelial cells.](#)
- [Role for protein phosphatase 2A in the regulation of Calu-3 epithelial Na<sup>+</sup>-K<sup>+</sup>-2Cl<sup>-</sup>, type 1 co-transport function.](#)