

Background

Sphingolipids are metabolized into bioactive products that include ceramide, sphingosine, and sphingosine-1-phosphate (S1P). Sphingosine Kinase (SK) catalyzes the phosphorylation of the lipid sphingosine, creating S1P. S1P subsequently signals through cell surface G protein-coupled receptors, as well as intracellularly, to modulate cell proliferation, survival, motility and differentiation. Two isoforms of SK have been identified, SK1 and SK2. The mRNA for both of these isoforms is widely expressed with SK1 expression highest in brain, heart, kidney, thymus, spleen and lung, while SK2 is highest in kidney and liver. SKs can be activated through growth factor, G protein-coupled, and immunoglobulin receptor signalling. SK1 has been shown to mediate cell growth, prevention of apoptosis, and cellular transformation, and is upregulated in a variety of human tumors. Regulation of SK1 may occur through ERK mediated phosphorylation of Ser-225. This phosphorylation leads to increased activity and translocation to the plasma membrane.

Background References

Melendez, A.J. et al. (2000) *Gene* 251(1) :19.
 Pitson, S.M. et al. (2003) *EMBOJ.* 22(20) :5491.
 Pitson, S.M. et al. (2005) *J Exp. Med.* 201(1) :49.

Product Citations

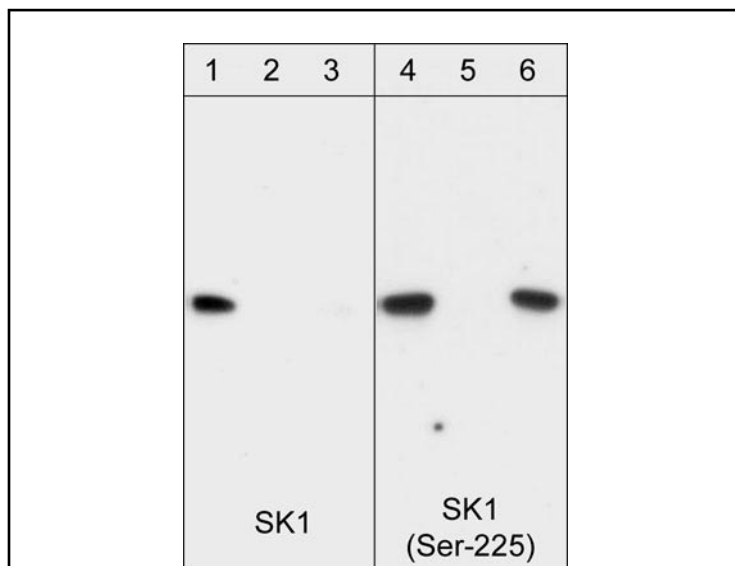
Bruno, G. et al. (2019) *Oncogene.* s41388-019-0993-1.
WB: human neuroblastoma

Bandara, G. et al. (2019) *Front Immunol.* 9:631.
WB: human mast cells

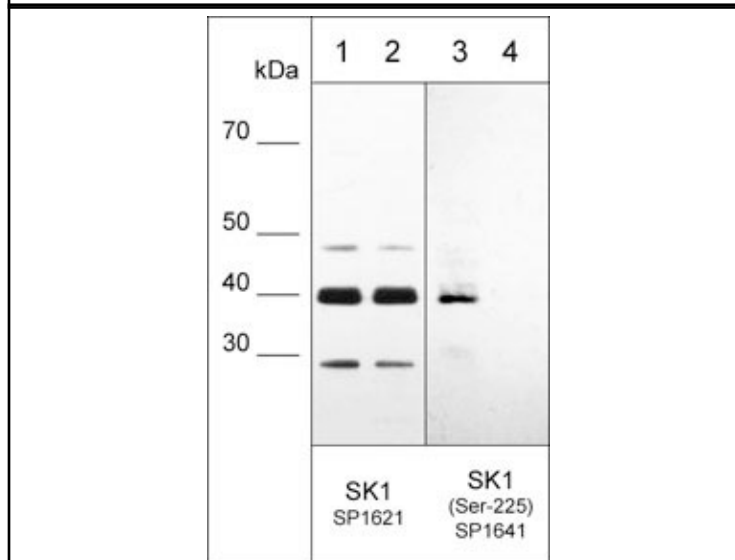
Hanyu, T. et al. (2018) *Surgery.* 163(6):1301.
IHC: human gastric cancer

Stayrook, KR et al. (2015) *Bonekey Rep.* 4:719.
IP: human MDA-MB-231 cells

Bruno, G et al. (2015) *Biochim Biophys Acta.* 1851(2):194.
WB: mouse C2C12 myoblasts



Western blot image of recombinant his-tagged human SK1 protein that was phosphorylated with ERK-2. Blots were probed with anti-SK1 (Central Region) (SP1621; lanes 1-3) and anti-SK1 (Ser-225) (SP1641; lanes 4-6). Both antibodies were used in the presence of no peptide (lanes 1 & 4), phospho-SK1 (Ser-225) peptide (SX1645; lanes 2 & 5), or unphosphorylated SK1 (Ser-225) peptide (SX1625; lanes 3 & 6).



Western blot of HeLa stimulated with calyculin A (lanes 1-4). The blots were untreated (lane 1 & 3) or treated with lambda phosphatase (lane 2 & 4), then probed with anti-SK1 (Central region) SP1621 (lanes 1 & 2) or anti-SK1 (Ser-225) SP1641 (lanes 3 & 4).

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Immunogen**Uniprot ID: Q9NYA1**

SK1 synthetic peptide (coupled to carrier protein) corresponding to an unphosphorylated sequence of amino acids surrounding serine 225 in the central region of human SK1. This sequence has 4 amino acid differences from mouse and 5 from rat SK1, and is not homologous to sequences in SK2.

Buffer and Storage

Rabbit polyclonal, affinity-purified antibody is supplied in 100µl phosphate-buffered saline, 50% glycerol, 1 mg/ml BSA, and 0.05% sodium azide. Store at -20°C. Stable for 1 year.

Applications

WB 1:1000
ELISA 1:2000

Species Reactivity

Hu

End user should determine optimal dilution for their particular applications and experiments.

Western blot membranes were incubated with diluted antibody in 5% non-fat milk, Tris buffer, 0.04% Tween20 for 1 hour at room temperature.

Abbreviations: E = ELISA, ICC = immunocytochemistry, IHC = immunohistochemistry, IP = immunoprecipitation, MS = mass spectrometry, WB = western blot
Hu = Human, Ms = Mouse, Rt = Rat, Ck = Chicken, F = Frog, B = Bovine

Specificity

This antibody was affinity purified using SK1 (Central Region) peptide. The purified antibody detects a 47 kDa* full-length recombinant human SK1 protein, as well as detects endogenous SK1 isoforms near 40 and 50 kDa in HeLa, MeWo, and PC3 cells. In mouse, the antibody detects SK1 in isolated cardiac myocytes, but not in SK1-null mice (Tao, R. et al. (2007) Cardiovas. Res. 74:56.).

*All molecular weights (MW) are confirmed by comparison to MW standards and to western blot mobilities of known proteins with similar MW.

"Native" western blot utilizes non-reducing sample buffer (no mercaptoethanol or SDS), normal SDS-PAGE gel electrophoresis, and no methanol in transfer buffers.

Related Products

SP1641 Sphingosine Kinase 1 (Ser-225), phospho-specific Rabbit Polyclonal

SX1645 phospho-SK1 (Ser-225) Blocking Peptide

SX1625 unphosphorylated SK1 (Ser-225) Blocking Peptide

SK6640 Sphingosine Kinase Activation Antibody Sampler Kit

SK6010 Sphingosine Kinase 1 Phospho-Regulation Antibody Sampler Kit

SK6590 Sphingosine Kinase 2 Phospho-Regulation Antibody Sampler Kit

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