





This antibody was developed and validated by the Medical Research Council Protein Phosphorylation and **Ubiquitylation Unit (University of** Dundee, Dundee, UK).

# **Background**

Protein ubiquitylation and protein phosphorylation are the two major mechanisms that regulate the functions of proteins in eukaryotic cells. However, these different posttranslational modifications do not operate independently of one another, but are frequently interlinked to enable biological processes to be controlled in a more complex and sophisticated manner. Studying how protein phosphorylation events control the ubiquitin system and how ubiquitylation regulates protein phosphorylation has become a focal point of the study of cell regulation and human disease. The serumand glucocorticoid-inducible protein kinase (SGK) family is made up of three isoforms, SGK1, 2, and 3, that are phosphatidylinositide-3-kinase (PI3-K)-dependent, serine/ threonine kinases, with similar substrate specificity to protein kinase B (PKB). Consequently, the SGK family also regulates similar cell processes to the PKB kinases, including cell proliferation and survival (Bruhn et al., 2013). Cloning of the genefor SGK1 was first described by Webster et al. (1993). SGK1 is activated by insulin, growth factors and oxidative stress via PI3-K, 3-phosphoinositide-dependent kinase PDK1 and mTOR. Mechanisms employed by SGK1 in transport regulation include direct phosphorylation of target transport proteins, phosphorylation and thus activaSGK1 (human; full length), pAb

Alternate Names: Serine/threonine-protein kinase Sgk1, Serum/glucocorticoid-regulated kinase 1

Cat. No. 68-0033-100 Quantity: 100 µg Lot. No. -20°C 30272 Storage:

FOR RESEARCH USE ONLY NOT FOR USE IN HUMANS

**CERTIFICATE OF ANALYSIS** Page 1 of 2

## **Physical Characteristics**

Quantity: 100 µg

Concentration: to be provided on

shipping

Source: sheep polyclonal antibody

Immunogen: human SGK1 (residues

1-432) [GST-tagged]

Purification: affinity-purified using

immobilized immunogen

Formulation: phosphate-buffered

Specificity: detects SGK1 at ~49 kDa

Reactivity: human; other species not

tested

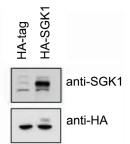
Stability/Storage: 12 months at -20°C; aliquot as required

# **Research Applications and Quality Assurance**

Western Immunoblotting:

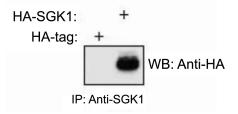
use 1 µg/ml

Immunoprecipitation: use 5 µg/mg of cell extract



### Western Blotting Analysis:

HEK293 cells were transfected with vectors expressing HA-SGK1 or HA-tag as a control. The cells were then lysed and the lysates denatured in SDS and subjected to SDS-PAGE on 8% gels. Western Blotting was carried out with 1 µg/ml anti-SGK1 antibody (Cat# 68-0033-100) or a commercially available anti-HA antibody.



#### Immunoprecipitation Assay:

Immunoprecipitation was performed from HEK293 cells over-expressing HA-tagged SGK1 (1 mg of cell extract) using 5 µg of anti-SGK1 antibody (Cat# 68-0033-100). SGK1 was subsequently detected by Western Blot using a commercially available anti-HA antibody.

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Lot-specific COA version tracker: v1.0.0







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## **Background**

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tion of other transport regulating kinases, stabilisation of membrane proteins by phosphorylation and thus inactivation of the ubiquitin E3 ligase NEDD4-2, as well as stimulation of transport protein expression by up-regulating transcription factors (e.g. nuclear factor kappa-B (NFkB)) and by fostering of protein translation. Moreover, excessive SGK1 activity has been shown to contribute to the pathophysiology of hypertension, obesity, diabetes, thrombosis, stroke, inflammation, autoimmune disease, fibrosis and tumour growth (Lang et al., 2014).

### **Antibody Production:**

Anti-SGK1 (human) polyclonal antibody was raised in sheep against SGK1 (residues 1-432 of human SGK1). The antibodies were purified by the Medical Research Council Protein Phosphorylation and Ubiquitylation Unit (MRC-PPU, University of Dundee, Dundee, U.K.) by affinity purification of the anti-SGK1 pAbs from the sheep serum using an antigenagarose column followed by depletion of any anti-GST antibodies using a GSTagarose column. Anti-SGK1 (human) pAb was sourced by Ubiquigent directly from the MRC-PPU.

#### General References:

Bruhn MA, Pearson RB, Hannan RD and Sheppard KE (2013) AKTindependent PI3-K signaling in cancer - emerging role for SGK3. Cancer Manag Res 5, 281-292.

Lang F, Stournaras C and Alesutan I (2014) Regulation of transport across cell membranes by the serum- and glucocorticoid-inducible kinase SGK1. Mol Membr Biol 31, 29-36.

Webster MK, Goya L, Ge Y, Maiyar AC and Firestone GL (1993) Characterization of sgk, a novel member of the serine/threonine protein kinase gene family which is transcriptionally induced by glucocorticoids and serum. Mol Cell Biol 13, 2031-2040.



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