

This antibody was developed and validated by the Medical Research Council Protein Phosphorylation and **Ubiguitylation 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 gene for 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 activa-

Continued on page 2

www.ubiquigent.com

SGK1 (human; residues 412-431), pAb

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

Cat.	No.	68
Lot.	No.	30

8-0034-100 0273

Quantity: Storage:

100 µg -20°C

NOT FOR USE IN HUMANS

CERTIFICATE OF ANALYSIS

FOR RESEARCH USE ONLY

Page 1 of 2

Physical Characteristics

Quantity: 100 µg

Concentration: to be provided on shipping

Source: sheep polyclonal antibody

Immunogen: human SGK1 (residues 412-431) [KEAAEAFLGFSYAPPTDSFL]

Purification: affinity-purified using immobilized immunogen

Formulation: phosphate-buffered saline

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

HA-SGK1:

Immunoprecipitation: use 2 µg/mg of cell extract



Western Blotting Analysis:

HEK293 cells were transfected with vectors expressing HA-SGK1, HA-SGK2, HA-SGK3 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# 68-0034-100) or a commercially available anti-HA antibody.

Immunoprecipitation Assay:

Immunoprecipitation was performed on HEK293 cells over-expressing HA-SGK1, HA-SGK2, HA-SGK3 or HA-tag (0.5 mg of cell extract) using 1 µg of anti-SGK1 antibody (Cat# 68-0034-100) or a commercially available anti-HA antibody. Western Blotting was subsequently performed using a commercially available anti-HA antibody. SGK1 antibody (Cat# 68-0034-100) immunoprecipitates HA-SGK1, HA-SGK2 and HA-SGK3 as well as the anti-HA antibody does.

© Ubiquigent 2014. Unless otherwise noted, Ubiquigent, Ubiquigent logo and all other trademarks are the property of Ubiquigent, Ltd. **ORDERS / SALES SUPPORT UK HQ and TECHNICAL SUPPORT** International: +44 (0) 1382 381147 (9AM-5PM UTC) International: +1-617-245-0020 Limited Terms of Use: For research use only. Not for use in humans or for diagnostics. Not for distribution or resale in any form, modification or derivative OR for use in providing services to a third party (e.g. screening or profiling) without the written permission of Ubiquigent, Ltd. US Toll-Free: 1-888-4E1E2E3 (1-888-431-3233) US/Canada: +1-617-245-0020 (9AM-5PM UTC) Email: sales.support@ubiquigent.com Email: tech.support@ubiquigent.com Email services@ubiquigent.com for enquiries regarding compound profiling and/or custom assay development services. Lot-specific COA version tracker: v1.0.0 Dundee, Scotland, UK



SGK1 (human; residues 412-431), pAb

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

Cat.	No.	68-0
Lot.	No.	302

FOR RESEARCH USE ONLY

68-0034-100 30273 Quantity: Storage:

100 μg -20°C

NOT FOR USE IN HUMANS

CERTIFICATE OF ANALYSIS

Page 2 of 2

Background

Continued from page 1

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 (NF κ B)) 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 412-431 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 a GST-tagged antigen-agarose column. 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.



ORDERS / SALES SUPPORT

 International:
 +1-617-245-0020

 US Toll-Free:
 1-888-4E1E2E3 (1-888-431-3233)

 Email:
 sales.support@ubiquigent.com

UK HQ and TECHNICAL SUPPORT

 International:
 +44 (0) 1382 381147
 (9AM-5PM UTC)

 US/Canada:
 +1-617-245-0020
 (9AM-5PM UTC)

 Email:
 tech.support@ubiquigent.com

Email services@ubiquigent.com for enquiries regarding compound profiling and/or custom assay development services. © Ubiquigent 2014. Unless otherwise noted, Ubiquigent, Ubiquigent logo and all other trademarks are the property of Ubiquigent, Ltd.

Limited Terms of Use: For research use only. Not for use in humans or for diagnostics. Not for distribution or resale in any form, modification or derivative OR for use in providing services to a third party (e.g. screening or profiling) without the written permission of Ubiquigent, Ltd.

Lot-specific COA version tracker: v1.0.0