

BRSK2 [6His-tagged]

Kinase

Alternate Names: BR Serine/Threonine Kinase 2; SAD1; PEN11B

Cat. No. 66-0001-050

Lot. No. 2138

Quantity: 50 µg

Storage: -70°C

FOR RESEARCH USE ONLY

NOT FOR USE IN HUMANS



CERTIFICATE OF ANALYSIS Page 1 of 2

Background

by Sir Phillip Cohen

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. Cloning of human Brain Specific Kinase 2 (BRSK2) was first described by Miura *et al.* (1998). BRSK2 is a member of the subfamily of protein kinases that include the AMP-activated protein kinase (AMPK) and, like AMPK itself, is activated by the tumour suppressor kinase LKB1 (Lizcano *et al.*, 2004). As implied by its name BRSK2 is expressed in the brain where it plays an essential role in controlling neuronal cell polarisation. BRSK2 contains a ubiquitin-like domain adjacent to the kinase catalytic domain (Al-Hakim *et al.*, 2008).

References:

Al-Hakim AK, Zagorska A, Chapman L, Deak M, Peggie M, Alessi DR (2008) Control of AMPK-related kinases by USP9X and atypical Lys(29)/Lys(33)-linked polyubiquitin chains. *Biochem J* 411, 249-60.

Hastie CJ, McLauchlan HJ, Cohen P (2006) Assay of protein kinases using radiolabeled ATP: a protocol. *Nat Protoc* 1, 968-71.

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Physical Characteristics

Species: human

Source: Sf21 insect cell-baculovirus expression

Quantity: 50 µg

Concentration: 1 mg/ml

Formulation: 50 mM Tris/HCl pH 7.5, 0.1 mM EGTA, 150 mM NaCl, 270 mM sucrose, 0.03% Brij, 0.1% β-Mercaptoethanol, 1 mM Benzamidine, 0.2 mM PMSF

Molecular Weight: ~78.3 kDa

Purity: >95% by InstantBlue™ SDS-PAGE

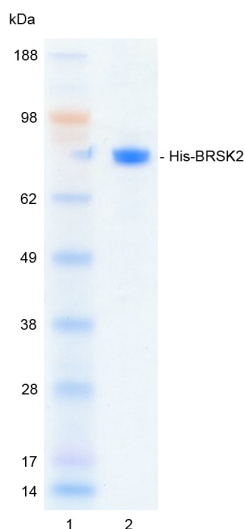
Stability/Storage: 12 months at -70°C; aliquot as required

Protein Sequence: Please see page 2

Quality Assurance

Purity:

4-12% gradient SDS-PAGE
InstantBlue™ staining
Lane 1: MW markers
Lane 2: 1 µg His-BRSK2



Protein Identification:

Confirmed by mass spectrometry.

Activity Assay:

The specific activity of His-BRSK2 was determined using the method described by Hastie *et al.* (2006) with the enzyme being assayed at several concentrations. His-BRSK2 was incubated for 10 minutes at 30°C in kinase reaction buffer in the presence of CHKtide substrate (300µM) and [γ -³²P]ATP (100µM). Duplicate reactions were stopped by spotting the assay mixture onto Whatman P81 paper – capturing the phosphorylated substrate. The radioactivity incorporated was measured on a scintillation counter and the enzyme's mean specific activity was calculated.

His-BRSK2 specific activity:

333.7 Units/mg (333.7 Units/ml)

1 Unit = 1 nmole of phosphate incorporated into the substrate in 1 minute

Substrate: CHKtide (KKKVSRSGLYRSPSPENLNRRP)



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Lot-specific COA version tracker: v1.0.0

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Background by Sir Phillip Cohen

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Miura K, Masuzaki H, Ishimaru T, Niikawa N, Jinno Y (1998) A HhaI/BstUI polymorphism in a novel gene at human chromosome 11p15.5. *J Hum Genet* **90**, 283-4.

Lizcano JM, Goransson O, et al. (2004) LKB1 is a master kinase that activates 13 kinases of the AMPK subfamily, including MARK/PAR-1. *EMBO J* **23**, 833-43.

Background kindly written by:

Sir Phillip Cohen FRS, FRSE
University of Dundee

Director of the Medical Research Council Protein Phosphorylation Unit (1990-2012)

Director of the Scottish Institute for Cell Signalling incorporating the Protein Ubiquitylation Unit (2008-2012)

Co-Director of the Division of Signal Transduction Therapy (1998-2012)

Deputy Director of the Division of Signal Transduction Therapy (from July 2012)

Professor Cohen's research group is studying the interplay between protein phosphorylation and protein ubiquitylation in the regulation of innate immunity.

Physical Characteristics

Continued from page 1

Protein Sequence:

MSY YHHHHHDYDIPTTENLYFQGAMGS T
STGKDGGGAQHAQYVGPYRLEKTLGKGQT
GLV KLG VHC VTC QKVAIKIVNREKLS
ESVLMKVEREIAILKLI EHPHVLKLDH
VYENKKYLYLVLEHVS GGELFDYLVK
KGR LTPKEARKFFRQIIISALDFCHSH
SICHRDLKPENLLLD EKNNIRIADFG
MASLQVGD SLETS CGSPHYACPEVIR
GEKYDGRKADVWSCGVILFALLVGALPFD
DDNLRQLLEKVKRGV FHM PFI PPDC
QSLLRGMIEVDAARRLTLEHIQKHIWYIG
GKNEPEPEQPIPRKVQIRSLPSLEDIDP
DVLDSMHSLGCFRDRNKLLQDLLSEEN
QEKMIYFLLLDRKERYPSQEDEDLPPR
NEIDPPRKRVDSPMLNRHGKRRPERKS
MEVLSVTDGGSPVPARRAIEMAQH
GQRSRSISGASSGLSTSP LSSPRVT
PHSPRGSPLPTPKGTPVHTPKESPAGT
PNPTPPSSPSVGGVWRARLNSIKNS
FLGSPRFHRRKLQVPTPEEMSNLTPSS
PELAKKSWFGNFI SLEKEEQIFVVIKDK
PLSSIKADIVHAFSLI PSLSHSVISQTS
FRAEYKATGGPAVFQKPKVKFQVDI
TYTEGGEAQKENGIIYSVTFLLSGPSR
RFKR VVETIQ AQLLSTHDPPAAQHLSEPP
PPAPGLSWGAGLKGQKVATSYESSL

Tag (**bold text**): N-terminal His

Protease cleavage site: TEV (**ENLYF**▼**QG**)

BRSK2 (regular text): Start **bold italics** (amino acid residues 2-674)

Accession number: AAP97725.1



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