MAPK12 (SAPK3) [GST-tagged]

Kinase

Alternate Names: Extracellular Signal-Regulated Kinase 6, ERK6, p38-Gamma

Cat. No.	**66-0032-050**	Quantity:	50 µg
Lot. No.	**30311**	Storage:	-70˚C

FOR RESEARCH USE ONLY

NOT FOR USE IN HUMANS

CERTIFICATE OF ANALYSIS Page 1 of 2

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. MAP kinases are serine, threonine, and tyrosine specific protein kinases that regulate proliferation, gene expression, differentiation, mitosis, cell survival, and apoptosis in response to stimuli, such as mitogens, osmotic stress, heat shock and pro-inflammatory cytokines. Cloning of human Mitogen Activated Protein kinase 12 (MAPK12 or SAPK3) was first described by Li *et al.* (1996). An example of such interplay between phosphorylation and ubiquitylation has been highlighted in a recent study showing that MAPK12 regulates RhoC (Ras homolog gene family, member C) expression by preventing RhoC ubiquitylation and subsequent lysosomal degradation. The findings from this study identify a novel mechanism for regulating RhoC expression and suggest that MAPK12 may be a candidate therapeutic target (Rosenthal *et al.*, 2011).

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Physical Characteristics

Species: human

Source: *E. coli*

Quantity: 50 µg

Concentration: 4.78 mg/ml

Formulation: 50 mM Tris/HCl pH7.5, 0.1 mM EGTA, 150 mM NaCl, 0.1% ß-Mercaptoethanol, 270 mM sucrose, 0.03% Brij-35, 1 mM Benzamidine, 0.2 mM PMSF

Molecular Weight: ~68.5 kDa

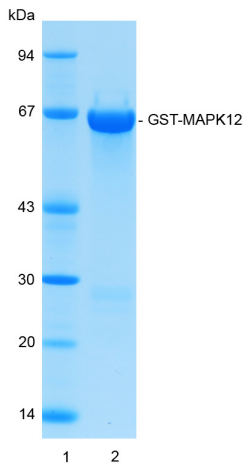
Purity: >95% by InstantBlue™ SDS-PAGE

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: 2.5 µg GST-MAPK12



Protein Identification:
Confirmed by mass spectrometry.

Activity Assay:
The specific activity of GST-MAPK12 was determined using the method described by Hastie *et al.* (2006) with the enzyme being assayed at several concentrations. GST-MAPK12 was incubated for 10 minutes at 30˚C in kinase reaction buffer in the presence of MBP substrate (0.333 mg/ml) and [γ-32P]ATP (100 µM). 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.

GST-MAPK12 specific activity:
266.5 Units/mg (1274 Units/ml)

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

Substrate: Myelin Basic Protein (MBP)

www.ubiquigent.com
Dundee, Scotland, UK

ORDERS / SALES SUPPORT
International: +1-617-245-0003
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.

Lot-specific COA version tracker: v1.0.0

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Background

Continued from page 1

References:

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

Li Z, Jiang Y, Ulevitch RJ and Han J (1996) The primary structure of p38 gamma: a new member of p38 group of MAP kinases. *Biochem Biophys Res Commun* **228**, 334-340.

Rosenthal DT, Iyer H, Escudero S, Bao L, Wu Z, Ventura AC *et al.* (2011) p38gamma promotes breast cancer cell motility and metastasis through regulation of RhoC GTPase, cytoskeletal architecture, and a novel leading edge behavior. *Cancer Res* **71**, 6338-6349.

Physical Characteristics

Continued from page 1

Protein Sequence:

MSPILGYWKIKGLVQPTRLLLEYLEEKYEEH
LYERDEGDKWRNKKFELGLEFPNLPYYIDGD
VKLTQSMAIIRYIADKHNMLGGCPKERAEISM
LEGAVLDIRYGVSRIAYSKDFETLKVDFL
SKLPEMLKMFEDRLCHKTYLNGDHVTHPD
FMLYDALDVVLYMDPMCLDAFPKLVCFK
KRIEAIPQIDKYLKSSKYIAWPLQGWQAT
FGGGDHPPKSDLVPRGSPEF***M***SSPPPARS**
GFYRQEVTKTAWEVRAVYRDLQPVGSGAY
GAVCSAVDGRTGAKVAIKKLYRPFQSEL
FAKRAYRELRLLKHMRHENVIGLLDVFTPDE
TLDDFTDFYLVMPFMGTDLGKLMKHEKLGE
DRIQFLVYQMLKGLRYIHAAGIIHRDLKPGN
LAVNEDCELKILDFGLARQADSEMTGYVVTRW
YRAPEVILNWMRYTQTVDIWSVGCIMAEMIT
GKTLFKGSDHLDQLKEIMKVTGTPPAEFVQR
LQSDEAKNYMKGLPELEKKDFASILTNAS
PLAVNLLEKMLVLDAEQRVTAGEALAHPY
FESLHDTEDEPQVQKYDDSFDDVDRTL
DEWKRVTYKEVLSFKPPRQLGARVSKETPL

Tag (**bold text**): N-terminal GST
Protease cleavage site: Thrombin (LVPR▼GS)
MAPK12 (regular text): Start ***bold italics*** (amino acid residues 1-367).
Accession number: CAA71511.1

www.ubiquigent.com
Dundee, Scotland, UK

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International: +1-617-245-0003
US Toll-Free: 1-888-4E1E2E3 (1-888-431-3233)
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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.

Lot-specific COA version tracker: v1.0.0