Certificate of Analysis

REACTION BIOLOGY

Recombinant Proteins

ProQinase™ p38-beta

Mitogen-activated protein kinase 11

Recombinant Human Active Protein Kinase

Synonyms: MAPK11; PRKM11; SAPK2 ;SAPK2B

Product No.: 0455-0000-3

Lot: 004

Description: Human p38-beta, full length, amino acids M_1-Q_{364} (as in NCBI/Protein entry NP_002742.3), activated, untagged, expressed in E. coli

Product identity: p38-beta, Lot 004, was confirmed as p38-beta by mass spectroscopy LC-ESI-MS/MS (Protagen AG, Germany)

Theoretical $MW_{Fusion\ Protein}$: 41,485 Da

Expression: E.coli

Activation: With MKK6 SDTD

Purification: Immobilized Metal Affinity Chromatography

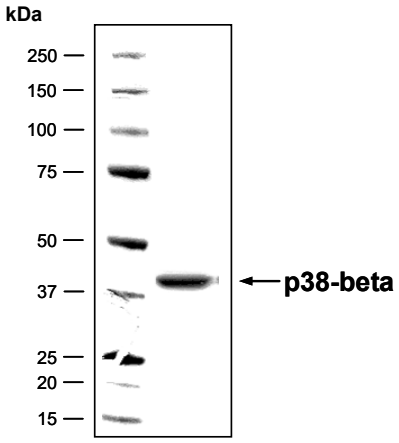
Storage buffer: 50 mM HEPES pH 7.5, 100 mM NaCl, 3 mM βME, 20% glycerol

Storage temperature: -80°C
For complete recovery, mix well and spin before use. Product must not be stored in diluted solutions, aliquots below 10µl are not advisable. Avoid repeated freeze-thaw cycles!

Protein concentration: 0.810 µg/µl
(Bradford method using BSA [Sigma, cat# A-7638, Lot 79H7641] as standard protein)

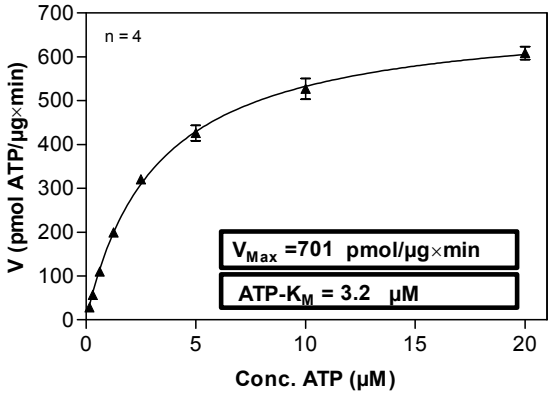
Specific activity: 701 pmol/µg×min

Coomassie stain:



2.0 µg p38-beta

Determination of K_m value for ATP:



Determination of K_m value & Specific activity:

- Assay conditions:
  60 mM HEPES-NaOH, pH 7.5
  3 mM $MgCl_2$
  3 mM $MnCl_2$
  3 µM Na-orthovanadate
  1.2 mM DTT
  50 µg / ml $PEG_{20.000}$
  ATP (variable)
  Substrate: rec. ATF2, 200 µg / ml
  p38-beta: 1.0 µg / ml
- Filter binding assay
  MSFC membrane (Millipore)

Additional assay technology: p38-beta Lot 004 was also successfully tested by Reaction Biology for the use with the ADP-Glo™ Kinase assay from ADP-Glo assay conditions may vary from radiometric assay conditions, please inquire for assay details

Promega

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ProQinase™ p38-beta

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p38-beta Recombinant Fusion Protein Amino Acid Sequence

1	GAMSGPRAGF YRQELNKTVW EVPQRLQGLR PVGSGAYGSV CSAYDARLRQ KVAVKKLSRP	60
61	FQSLIHARRT YRELRLLKHL KHENVIGLLD VFTPATSIED FSEVYLVTTL MGADLNNIVK	120
121	CQALSDEHVQ FLVYQLLRGL KYIHSAGIIH RDLKPSNVAV NEDCELRILD FGLARQADEE	180
181	MTGYVATRWY RAPEIMLNWM HYNQTVDIWS VGCIMAELLQ GKALFPGSDY IDQLKRIMEV	240
241	VGTPSPEVLA KISSEHARTY IQSLPPMPQK DLSSIFRGAN PLAIDLLGRM LVLDSDQRVS	300
301	AAEALAHAYF SQYHDPEDEP EAEPYDESVE AKERTLEEWK ELTYQEVLSF KPPEPPKPPG	360
361	SLEIEQ	420

1-2: leagcy from TEV cleavage **blue**: **MAPK11**

p38-beta wt[1] Amino Acid Sequence

1	MSGPRAGFYR QELNKTVWEV PQRLQGLRPV GSGAYGSVCS AYDARLRQKV AVKKLSRPFQ	60
61	SLIHARRTYR ELRLLKHLKH ENVIGLLDVF TPATSIEDFS EVYLVTTLMG ADLNNIVKCQ	120
121	ALSDEHVQFL VYQLLRGLKY IHSAGIIHRD LKPSNVAVNE DCELRILDFG LARQADEEMT	180
181	GYVATRWYRA PEIMLNWMHY NQTVDIWSVG CIMAELLQGK ALFPGSDYID QLKRIMEVVG	240
241	TPSPEVLAKI SSEHARTYIQ SLPPMPQKDL SSIFRGANPL AIDLLGRMLV LDSDQRVSAA	300
301	EALAHAYFSQ YHDPEDEPEA EPYDESVEAK ERTLEEWKEL TYQEVLSFKP PEPPKPPGSL	360
361	EIEQ	420

blue: p38-beta sequence expressed in fusionprotein

[1]NCBI/Protein accession number NP_002742.3

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