Certificate of Analysis

REACTION BIOLOGY

ProQinase™ ATF2

activating transcription factor 2

Recombinant Protein Kinase Substrate

HGNC Symbol: ATF2

Synonyms: CRE-BP1, CREB2, HB16, TREB7

Product No.: 0594-0000-2

Lot: 014

Description: Human ATF2, N-terminal fragment, amino acids M_{19}-D_{110} (as in NCBI/Protein entry NP_001871.2), N-terminal GST-HIS_6 fusion protein with a 3C cleavage site, expressed in E.coli

Theoretical $MW_{Fusion\ Protein}$: 38,620 Da

Expression host: E.coli

Purification: GST-Affinity Chromatography

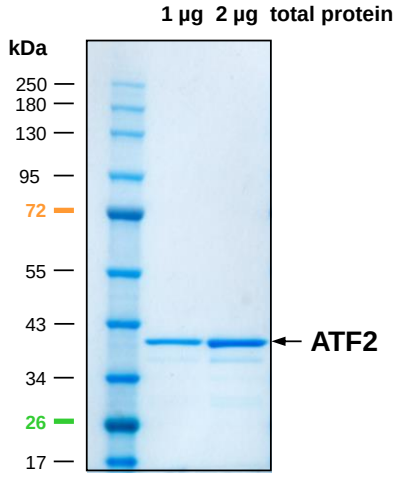
ATPase activity: In an ADP-Glo™ assay (Promega) with 10 µM ATP or 30 µM ATP, the ATP → ADP conversion within 30 min is <1% at a concentration of 100 µg/ml substrate.
Detailed ATPase assay conditions on request

Storage buffer: 50 mM HEPES pH 7.5, 100 mM NaCl, 5 mM DTT, 15 mM reduced glutathione, 10 % 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!

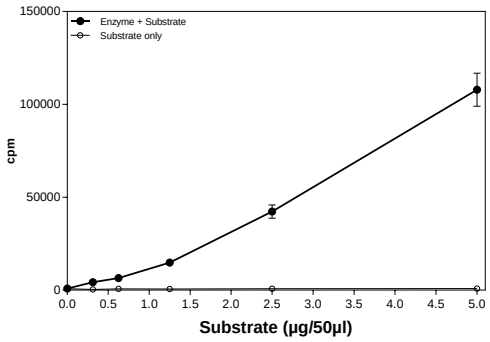
Protein concentration: 2.25 µg/µl
(Bradford method using BSA [Sigma, cat# A-7638, Lot 79H7641] as standard protein)

ATF2 Lot 014:
Coomassie stain



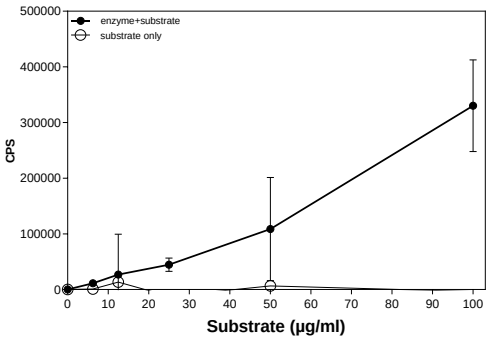
Phosphorylation of ATF2 Lot014 by p38beta

Radiometric filter binding assay



Assay conditions:
70 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: 1 µM
Substrate: variable concentration
Kinase: 1 µg/ml
MSFC filter plate

ADP-Glo™ assay (Promega)



Assay conditions:
70 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 PEG20.000
ATP: variable concentration
1 % (v/v) DMSO
Substrate: 100 µg/ml
Kinase: 1 µg/ml

Recombinant Proteins

ATF2

Product No.: 0594-0000-2

ATF2 Recombinant Fusion Protein Amino Acid Sequence

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  1  MSPILGYWKI KGLVQPTRLL LEYLEEKYEE HLYERDEGDK WRNKKFELGL EFPNLPYYID  60
 61  GDVKLTQSMA IIRYIADKHN MLGGCPKERA EISMLEGAVL DIRYGVSRIA YSKDFETLKV 120
121  DFLSKLPEML KMFEDRLCHK TYLNGDHVTH PDFMLYDALD VVLYMDPMCL DAFPKLVCFK 180
181  KRIEAIPQID KYLKSSKYIA WPLQGWQATF GGGDHPPKSD PMGHHHHHHG RDSLEVLFQG 240
241  PLAMSDDKPF LCTAPGCGQR FTNEDHLAVH KHKHEMTLKF GPARNDSVIV ADQTPTPTRF 300
301  LKNCEEVGLF NELASPFENE FKKASEDDIK KMPLD                            360
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1-218: GST **Red**: HIS6-tag **Green**: 3C cleavage site **blue**: ATF2 fragment

Recombinant Proteins