Recombinant Human Active JNK2α1

PRODUCT ANALYSIS SHEET

Catalog Number:	PHO3034	PHO3035	PHO3031
Quantity:	10 µg	25 µg	100 µg

Lot Number: See product label

Concentration: See product label

Molecular Weight: ~45 kDa

Purity: >95% as determined by SDS-PAGE analysis.

Amino Acid Sequence MSDSKCDSQF YSVQVADSTF TVLKRYQQLK PIGSGAQGIV CAAFDTVLGI NVAVKKLSRP FQNQTHAKRA YRELVLLKCV NHKNIISLLN VFTPQKTLEE FQDVYLVMEL MDANLCQVIH MELDHERMSY LLYQMLCGIK HLHSAGIIHR DLKPSNIVVK SDCTLKILDF GLARTACTNF MMTPYVVTRY YRAPEVILGM GYKENVDIWS VGCIMGELVK GCVIFQGTDH IDQWNKVIEQ LGTPSAEFMK KLQPTVRNYV ENRPKYPGIK FEELFPDWIF PSESERDKIK TSQARDLLSK MLVIDPDKRI SVDEALRHPY ITVWYDPAEA EAPPPQIYDA QLEEREHAIE EWKELIYKEV MDWEERSKNG VVKDQPSAQM QQ

Biological Activity: ~125 units/mg. One unit of JNK2α1 activity is equal to 1 nanomole of phosphate transferred to recombinant ATF-2 fusion protein (Catalog # PHF0041) per minute at 30°C with a final ATP concentration of 100 µM. Recombinant active JNK2α1 is capable of autophosphorylation and also phosphorylates c-Jun at serine 63 and serine 73. Kinase activity may vary depending on the substrate and reaction conditions. The optimal concentration should be determined for each specific application. JNK2α1 is shorter than JNK2α2, and contains a five nucleotide insertion in the C terminal region which causes a shift in the reading frame and subsequent premature termination.

Formulation: 50 mM Tris, pH 7.5, 0.15 M NaCl, 0.27 M sucrose, 10 mM β-mercaptoethanol, 1 mM EGTA, 0.1% Triton X-100, carrier–free.

Sterility: Filtered through a 0.22 micron sterile filter.

Production: Recombinant human active JNK2α1 (amino acids 1-382) is produced in *E. coli* and purified via sequential chromatography.

Handling Recommendation: We recommend that this vial be briefly centrifuged prior to opening to bring the contents to the bottom.

Storage: Liquid recombinant human active JNK2α1 should be kept as a solution in order to maintain full activity. This stock solution should be apportioned into working aliquots and stored at ≤−80°C. Keep freeze-thaw cycles to a minimum.

Expiration Date: Expires one year from date of receipt when stored as instructed.

References:

Sluss, H.K, T. Barrett, B. Derijard, and R.J. Davis (1994) Signal transduction by tumor necrosis factor mediated by JNK protein kinases. Mol. Cell. Biol. 14:8376-8384.

This product is for research use only. Not for use in diagnostic procedures.

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PI Hu JNK2α1 (Rev 2.0) DCC-08-1232

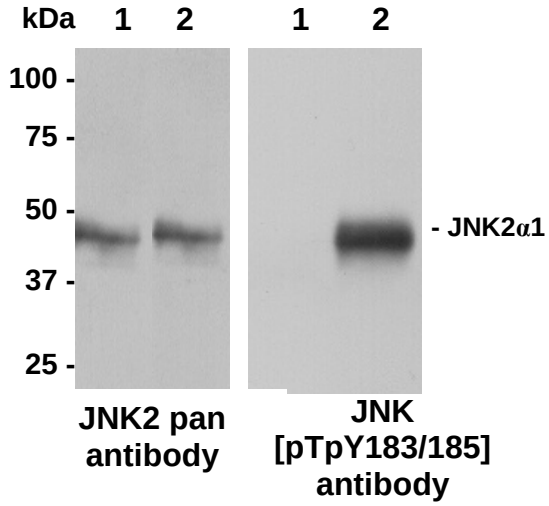
Kallunki, T., B. Su, I. Tsigelny, H.K. Sluss, B. Derijard, G. Moore, R. Davis, and M. Karin (1994) JNK2 contains a specificity-determining region responsible for efficient c-Jun binding and phosphorylation. Genes Dev. 8:2996-3007.

References: (Continued)

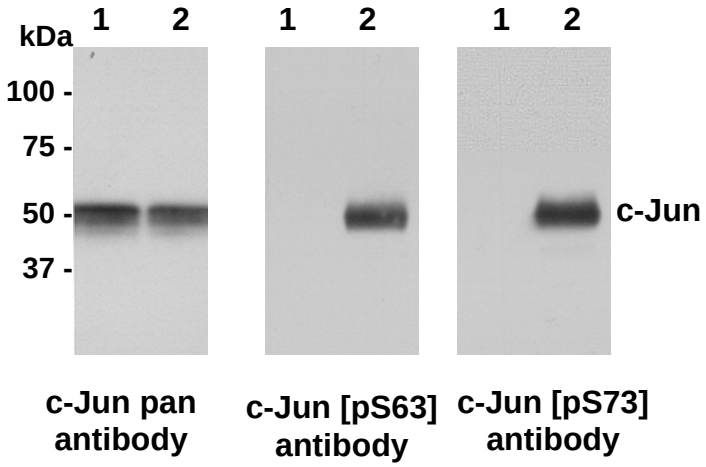
Gupta, S., T. Barrett, A.J. Whitmarsh, J. Cavanagh, H.K. Sluss, B. Derijard, and R.J. Davis (1996) Selective interaction of JNK protein kinase isoforms with transcription factors. EMBO J. 15:2760-2770.

Davis, R.J. (2000) Signal transduction by the JNK group of MAP kinases. Cell 103:239-252.

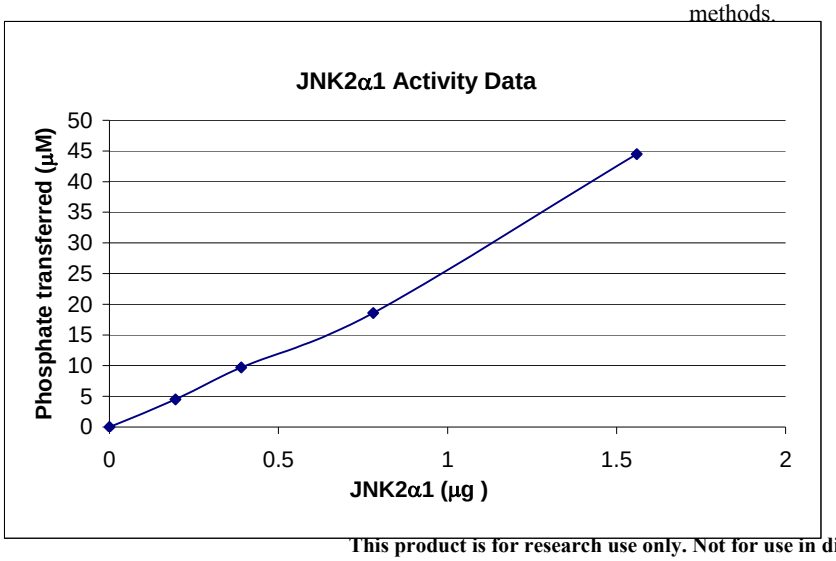
Lin, A. (2003) Activation of the JNK signaling pathway: Breaking the brake on apoptosis. Bioessays 25:17-24.



Recombinant active JNK2α1 is phosphorylated at threonine 183/tyrosine 185. Recombinant human inactive (lane 1) and active (lane 2) JNK2α1 were subjected to Western blot analysis using an anti-JNK pan antibody or anti-JNK [pTpY183/185] antibody (Catalog #44-682) in conjunction with chemiluminescence detection methods.



Recombinant active JNK2α1 phosphorylates c-Jun fusion protein. Recombinant c-Jun fusion protein (2 µg; Catalog #PHF0051) was incubated without (lane 1) and with (lane 2) 0.5 µg of recombinant human active JNK2α1 in an *in vitro* kinase reaction. Kinase reactions were analyzed by Western blot analysis using an anti-c-Jun pan antibody, anti-c-Jun [pS63] antibody, or anti-c-Jun [pS73] antibody (Catalog #44-292) in conjunction with chemiluminescence detection methods.



JNK2α1 activity. Varying amounts of recombinant JNK2α1 were incubated with 100 µM ATP and 100 µg GST-ATF2 (Catalog #PHF0041) in an *in vitro* kinase reaction. Phosphate transferred from ATP to the GST-ATF2 was measured using an ATP consumption assay and activity calculated based on the amount of phosphate specifically transferred to the ATF2 per minute at 30°C. Kinase conditions were chosen so that autophosphorylation is not detected.