



Product Information

c-Jun Kinase Substrate

Rat, Recombinant
Expressed in *E. coli*

Product Number **C 3233**
Storage Temperature $-70\text{ }^{\circ}\text{C}$

Synonyms: JNK substrate, c-Jun (1-79)-GST

Product Description

c-Jun is a component of the transcription factor AP-1 that binds and activates transcription at TRE/AP-1 elements. The transcriptional activity of c-Jun is regulated by phosphorylation at Ser⁶³ and Ser⁷³ by c-Jun-N-terminal kinases (JNKs).^{1,2} Extracellular signals including growth factors, transforming oncoproteins, hydrogen peroxide, and UV irradiation stimulate phosphorylation of c-Jun at Ser^{63/73} and activate c-Jun-dependent transcription.^{3,4} Mutation of Ser^{63/73} renders c-Jun nonresponsive to mitogenic and stress induced signaling pathways. Phosphorylated c-Jun homodimerizes or forms a heterodimeric complex with c-Fos creating Activator Protein (AP)-1 transcription factor.³

c-Jun Kinase Substrate is the rat c-Jun (1-79) activation domain that has been tagged with glutathione-S-transferase (GST) at the amino terminus. The fusion protein is expressed in *E. coli*. It is a highly specific substrate for JNK/SAPK. This protein is not phosphorylated by MAP kinase or by p38 kinase. The molecular weight is 37 kDa.

Reagent

c-Jun Kinase Substrate is supplied as a 1 mg/mL solution in 25 mM Tris buffer, pH 7.4, containing 50 mM NaCl, 0.5 mM EDTA, and 5 mM 2-mercaptoethanol.

Precautions and Disclaimer

Please consult the Material Safety Data Sheet for handling recommendations before working with this material.

Product Profile

Purity is >90% by SDS PAGE.

The standard reaction contains 0.1 μg c-Jun N-terminal kinase (JNK; C 3108) and 2 μg c-Jun Kinase Substrate in 25 mM HEPES, pH 7.4, 10 mM magnesium acetate, and 50 μM ATP (2 μCi [³²P]-ATP) in a final volume of 40 μl . Allow the reaction to proceed for 30 min at 30 $^{\circ}\text{C}$.

Note: Optimal assay conditions must be determined by the user for different enzymes and systems.

Storage/Stability

The product should be stored at $-70\text{ }^{\circ}\text{C}$. After initial thawing, store the solution in working aliquots at $-70\text{ }^{\circ}\text{C}$. Avoid repeated freeze/thaw cycles.

References

1. Binetruy, B., et al., Ha-Ras augments c-Jun activity and stimulates phosphorylation of its activation domain. *Nature*, **351**, 122-127 (1991).
2. Smeal, T., et al., Oncogenic and transcriptional cooperation with Ha-Ras requires phosphorylation of c-Jun on serines 63 and 73. *Nature*, **354**, 494-496 (1991).
3. Nose, K. et al., Transcriptional activation of early-response genes by hydrogen peroxide in a mouse osteoblastic cell line. *Eur J Biochem*, **201**, 99-106 (1991)
4. Derijard, B., et al., JNK1: a protein kinase stimulated by UV light and Ha-Ras that binds and phosphorylates the c-Jun activation domain. *Cell*, **76**, 1025-1037 (1994).

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