abcam

Product datasheet

c-Jun Transcription Factor Assay Kit (Colorimetric) ab207195

画像数 1

医薬用外劇物

製品の概要

製品名	c-Jun Transcription Factor Assay Kit (Colorimetric)
検出方法	Colorimetric
サンプルの種類	Nuclear Extracts
アッセイタイプ	Semi-quantitative
検出感度	< 1250 ng/well
全工程の試験時間	3h 30m
種交差性	**交差種:** Mouse, Human
製品の概要	c-Jun Transcription Factor Assay Kit (Colorimetric) (ab207195) is a high throughput assay to quantify AP-1 c-Jun activation in nuclear extracts. This assay combines a quick ELISA format with a sensitive and specific non-radioactive assay for transcription factor activation. A specific double stranded DNA sequence containing the TPA-responsive element (TRE) (5´–TGAGTCA– 3´) has been immobilized onto a 96-well plate. AP1 present in the nuclear extract specifically binds to the oligonucleotide. AP1 family member c-Jun is detected by a primary antibody that recognizes an epitope of c-Jun accessible upon DNA binding. An HRP-conjugated secondary antibody provides sensitive colorimetric readout at OD 450 nm. This product detects only human and mouse c-Jun. Key performance and benefits: Assay time: 3.5 hours (cell extracts preparation not included). Detection limit: < 1.25 µg nuclear extract/well. Detection range: 0.1 – 20 µg nuclear extract/well.
特記事項	The activator protein-1 (AP1) transcription factors belong to a large family of structurally related transcription factors that includes ATF1-4, c-Fos, c-Jun, c-Myc and C/EBP. The members of this family, named bZIP, share a dimerization domain with a leucine zipper motif and a DNA binding domain rich in basic residues (lysines and arginines). AP1 is composed of a mixture of heterodimeric complexes of proteins derived from the Fos and Jun families including c-Fos, FosB,

Fra-1, Fra-2, c-Jun, JunB and JunD. Only Jun proteins can form transcriptionally active homodimers within AP1 members, or heterodimers with CREB/ATF members, to bind the CRE element (5´ - TGACGTCA - 3´). Primarily, AP1 dimers bind to DNA on a TPA-response element (TRE) with the 5´ - TGA(C/G)TCA - 3´ sequence. Jun-Fos heterodimers form more stable complexes with TREs. These complexes display stronger transactivating activity than Jun-Jun homodimers.

Phosphorylation of AP1 family members by kinases is required for transactivation activity. In the case of c-Jun, the activation domain is regulated to a large extent by the JNK family of MAP kinases. JNK kinases phosphorylate c-Jun at Ser-63, resulting in the binding of c-Jun to the CBP/p300 family of transcriptional co-activators.

AP1 expression is induced by multiple stimuli such as serum, growth factors, phorbol esters and oncogenes. These include peptide growth factors, cytokines of the TGF beta, TNF, and interferon families, neuronal depolarization and cellular stress. Upon serum starvation of human fibroblast cells, Fos and Jun protein production can be induced for up to 4 hours by adding serum. Interestingly, serum starvation lowers basal expression of FosB and c-Fos but has no significant effect on c-Jun.

AP1 proteins play a role in the expression of many genes involved in proliferation and cell cycle progression including neuronal apoptosis, learning process, drug-induced behavorial responses, bone growth and differentiation, and embryo development. For instance, cell transformation by oncogenes that function in the growth factor signal transduction pathway, such as *ras*, *ras*F and *mek*, results in a high increase in AP1 component protein expression. Therefore, AP1-regulated genes support the invasive process observed during malignancy and metastasis.

試験プラットフォーム Microplate reader

製品の特性

保存方法 Please refer to protocols.

内容	1 x 96 tests	5 x 96 tests
10X Antibody Binding Buffer	1 x 2.2ml	1 x 11ml
10X Wash Buffer	1 x 22ml	1 x 110ml
96-well assay plate	1 unit	5 units
Anti-rabbit HRP-conjugated IgG	1 x 11µl	1 x 55µl
AP-1 Mutated oligonucleotide (10 pmol/µL)	1 x 100µl	1 x 500µl
AP-1 Wild-type oligonucleotide (10 pmol/µL)	1 x 100µl	1 x 500µl
Binding Buffer	1 x 10ml	1 x 50ml
Developing Solution	1 x 11ml	1 x 55ml
Dithiothreitol (DTT) (1 M)	1 x 100µl	1 x 500µl
K-562(TPA) nuclear extract (2.5µg/µL)	1 x 40µl	1 x 200µl
Lysis Buffer	1 x 10ml	1 x 50ml

内容	1 x 96 tests	5 x 96 tests
Phospho-c-Jun antibody	1 x 22µl	1 x 110µl
Plate sealer	1 unit	5 units
Poly [d(I-c)] (17 µg/µL)	1 x 100µl	1 x 500µl
Protease Inhibitor Cocktail	1 x 100µl	1 x 500µl
Stop Solution	1 x 11ml	1 x 55ml

機能

Transcription factor that recognizes and binds to the enhancer heptamer motif 5'-TGA[CG]TCA-3'. Promotes activity of NR5A1 when phosphorylated by HIPK3 leading to increased steroidogenic gene expression upon cAMP signaling pathway stimulation. Involved in activated KRAS-mediated transcriptional activation of USP28 in colorectal cancer (CRC) cells (PubMed:24623306). Binds to the USP28 promoter in colorectal cancer (CRC) cells (PubMed:24623306).

配列類似性

Belongs to the bZIP family. Jun subfamily.
Contains 1 bZIP (basic-leucine zipper) domain.

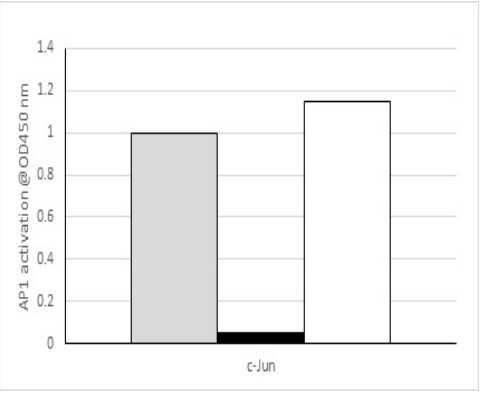
翻訳後修飾

Ubiquitinated by the SCF(FBXW7), leading to its degradation. Ubiquitination takes place following phosphorylation, that promotes interaction with FBXW7.
Phosphorylated by CaMK4 and PRKDC; phosphorylation enhances the transcriptional activity. Phosphorylated by HIPK3. Phosphorylated by DYRK2 at Ser-243; this primes the protein for subsequent phosphorylation by GSK3B at Thr-239. Phosphorylated at Thr-239, Ser-243 and Ser-249 by GSK3B; phosphorylation reduces its ability to bind DNA. Phosphorylated by PAK2 at Thr-2, Thr-8, Thr-89, Thr-93 and Thr-286 thereby promoting JUN-mediated cell proliferation and transformation. Phosphorylated by PLK3 following hypoxia or UV irradiation, leading to increase DNA-binding activity.
Acetylated at Lys-271 by EP300.

細胞内局在

Nucleus.

画像



Nuclear extracts from K-562 cells stimulated with TPA were assayed for activity of AP1 family member c-Jun.

Nuclear extracts from K-562 cells stimulated with TPA (Gray) were assayed for activity of AP1 family member c-Jun with 5 µg/well of nuclear extract in the absence or presence of wild-type (Black) or mutated (White) consensus binding oligonucleotides. These results are provided for demonstration purposes only.

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