SANTA CRUZ BIOTECHNOLOGY, INC.

Stat1 oligonucleotide

BACKGROUND

Membrane receptor signaling by various ligands, including interferons and growth hormones such as EGF, induces activation of JAK kinases which then leads to tyrosine phosphorylation of the various Stat transcription factors. Stat1 and Stat2 are induced by IFN-α and form a heterodimer which is part of the ISGF3 transcription factor complex. Although early reports indicate Stat3 activation by EGF and IL-6, it has been shown that Stat3β appears to be activated by both while Stat3α is activated by EGF, but not by IL-6. Highest expression of Stat4 is seen in testis and myeloid cells. IL-12 has been identified as an activator of Stat4. Stat5 has been shown to be activated by prolactin and by IL-3. Stat6 is involved in IL-4 activated signaling pathways.

REFERENCES

1. Zhong, Z., et al. 1994. Stat3: a Stat family member activated by tyrosine phosphorylation in response to epidermal growth factor and interleukin-6. Science 264: 95-98.
2. Darnell, J.E., et al. 1994. JAK-Stat pathways and transcriptional activation in response to IFNs and other extracellular signaling proteins. Science 264: 1415-1421.

GEL SHIFT ASSAYS

For gel shift analysis, prepare nuclear extracts following the method of Dignam, et al (1).

- **NOTE**: Spin oligonucleotide vial before opening. Product may be lodged in vial cap.
- Label oligonucleotide probe (TransCruz™ Gel Shift Oligonucleotides) with [γ^{32} P]-ATP to 50,000 cpm/ng by using polynucleotide kinase.
- Prepare gel shift reaction buffer as follows: 10 mM Tris (Tris: sc-3715), pH 7.5, 50 mM NaCl (NaCl: sc-29108, 1 mM dithiothreitol (DTT: sc-29089), 1 mM EDTA (EDTA: sc-29092), 5% glycerol (glycerol: sc-29095).
- Prepare 20 µl reaction mixture containing 3-10 µg nuclear extract and 1 µg poly dI-dC in gel shift reaction buffer. Add 0.5 ng labeled oligonucleotide probe and incubate for 20 minutes at room temperature. This constitutes the control sample for detection of DNA-protein complexes (2).
- To detect an antibody supershift or block of the DNA-protein complex, prepare reaction mixture as described above, also adding 1-2 µl of the appropriate TransCruz™ Gel Supershift antibody per 20 µl of reaction volume. Antibody is normally added subsequent to addition of labeled oligonucleotide probe, but result may be improved by adding antibody prior to probe. Incubate at 4° C for 1 hour to overnight, or at room temperature for 15-45 minutes.
- Resolve DNA-protein complexes by electrophoresis (25-35 ma) through a 4% polyacrylamide gel containing 50 mM Tris, pH 7.5, 0.38 M glycine (glycine: sc-29096) and 2 mM EDTA. Dry the gel and visualize by autoradiography.

RESEARCH USE

For research use only, not for use in diagnostic procedures.

PRODUCT

Stat1 p84/p91 CONSENSUS OLIGONUCLEOTIDE: sc-2573

- binding site for Stat1 p84/p91 (3)

5' - CAT GTT ATG CAT ATT CCT GTA AGT G - 3'
3' - GTA CAA TAC GTA TAA GGA CAT TCA C - 5'

Stat1 p84/p91 MUTANT OLIGONUCLEOTIDE: sc-2574

- identical to sc-2573 with the exception of a "CCT"→"GGA" substitution in the Stat1 p84/p91 binding motif (3)

5' - CAT GTT ATG CAT ATT **GGA** GTA AGT G - 3'
3' - GTA CAA TAC GTA TAA CCT CAT TCA C - 5'

SELECT PRODUCT CITATIONS

1. Chaturvedi, P., et al. 1997. Abrogation of interleukin-3 dependence of myeloid cells by the v-Src oncogene requires SH2 and SH3 domains which specify activation of STATs. Mol. Cell. Biol. 17: 3295-3304.
2. Campbell, J.S., et al. 2001. Expression of suppressors of cytokine signaling during liver regeneration. J. Clin. Invest. 107: 1285-1292.
3. Foley, H.A., et al. 2002. Stat3 β inhibits γ-globin gene expression in erythroid cells. J. Biol. Chem. 277: 16211-16219.
4. Zimmers, T.A., et al. 2003. Massive liver growth in mice induced by systemic interleukin 6 administration. Hepatology 38: 326-334.
5. Wang, T.L., et al. 2004. Angiotensin II signals mechanical stretch-induced cardiac matrix metalloproteinase expression via JAK-STAT pathway. J. Mol. Cell. Cardiol. 37: 785-794.
6. Feriotto, G. 2005. Myocyte enhancer factor 2 activates promoter sequences of the human AβH-J-J locus, encoding aspartyl-β-hydroxylase, junctin, and junctate. Mol. Cell. Biol. 25: 3261-3275.
7. Muthian, G., et al. 2006. 1,25 Dihydroxyvitamin-D_3 modulates JAK-STAT pathway in IL-12/IFNγ axis leading to Th1 response in experimental allergic encephalomyelitis. J. Neurosci. Res. 83: 1299-1309.
8. Norkina, O., et al. 2008. Acute alcohol intake induces SOCS1 and SOCS3 and inhibits cytokine-induced Stat1 and Stat3 signaling in human monocytes. Alcohol. Clin. Exp. Res. 32: 1565-1573.
9. Kim, H.J., et al. 2011. Loss of the promyelocytic leukemia protein in gastric cancer: implications for IP-10 expression and tumor-infiltrating lymphocytes. PLoS ONE 6: e26264.
10. Lee, S.H., et al. 2014. Benzylideneacetophenone derivatives attenuate IFN-γ-induced IP-10/CXCL10 production in orbital fibroblasts of patients with thyroid-associated ophthalmopathy through STAT-1 inhibition. Exp. Mol. Med. 46: e100.

STORAGE

Store at -20° C; stable for one year from the date of shipment.

NOTE: Spin oligonucleotide vial before opening. Product may be lodged in vial cap.