

RNA pol σ 70 (2G10): sc-56768

BACKGROUND

RNA polymerase transcribes DNA to synthesize RNA using the four ribonucleoside triphosphates as substrates. Prokaryotes contain a single RNA polymerase compared to three in eukaryotes. RNA polymerase requires one of a family of σ subunits, including σ 70 (RNA pol σ 70), for specific promoter recognition and initiation. RNA pol σ 70 represents the "housekeeping" σ factor, as it is necessary to transcribe most genes in growing cells. RNA pol σ 70 interacts with promoter DNA sequences via a mechanism mediated by an N-terminal inhibitory domain. This subunit normally functions through an allosteric interaction with the core subunits of RNA polymerase. RNA pol σ 70 also plays a critical regulatory role during transcription elongation at the bacteriophage λ late promoter where it mediates a pause in early elongation through contact with a DNA sequence element in the initially transcribed region that resembles a promoter -10 element.

REFERENCES

1. Dombroski, A.J., Walter, W.A., Record, M.T., Siegele, D.A. and Gross, C.A. 1992. Polypeptides containing highly conserved regions of transcription initiation factor σ 70 exhibit specificity of binding to promoter DNA. *Cell* 70: 501-512.
2. Dombroski, A.J., Walter, W.A. and Gross, C.A. 1994. The role of the σ subunit in promoter recognition by RNA polymerase. *Cell. Mol. Biol. Res.* 39: 311-317.
3. Bowers, C.W. and Dombroski, A.J. 1999. A mutation in region 1.1 of σ 70 affects promoter DNA binding by *Escherichia coli* RNA polymerase holoenzyme. *EMBO J.* 18: 709-716.
4. Panaghie, G., Aiyar, S.E., Bobb, K.L., Hayward, R.S. and de Haseth, P.L. 2000. Aromatic amino acids in region 2.3 of *Escherichia coli* σ 70 participate collectively in the formation of an RNA polymerase-promoter open complex. *J. Mol. Biol.* 299: 1217-1230.
5. Baldwin, N.E. and Dombroski, A.J. 2001. Isolation and characterization of mutation σ 70. *Mol. Microbiol.* 42: 427-437.
6. Vuthoori, S., Bowers, C.W., McCracken, A., Dombroski, A.J. and Hinton, D.M. 2001. Domain 1.1 of the σ (70) subunit of *Escherichia coli* RNA polymerase modulates the formation of stable polymerase/promoter complexes. *J. Mol. Biol.* 309: 561-572.
7. Gruber, T.M. and Gross, C.A. 2003. Multiple σ subunits and the partitioning of bacterial transcription space. *Annu. Rev. Microbiol.* 57: 441-466.
8. Geszvain, K., Gruber, T.M., Mooney, R.A., Gross, C.A. and Landick, R. 2004. A hydrophobic patch on the flap-tip helix of *E.coli* RNA polymerase mediates σ (70) region 4 function. *J. Mol. Biol.* 343: 569-587.
9. Nickels, B.E., Garrity, S.J., Mekler, V., Minakhin, L., Severinov, K., Ebright, R.H. and Hochschild, A. 2005. The interaction between σ 70 and the β -flap of *Escherichia coli* RNA polymerase inhibits extension of nascent RNA during early elongation. *Proc. Natl. Acad. Sci. USA* 102: 4488-4493.

PROTOCOLS

See our web site at www.scbt.com or our catalog for detailed protocols and support products.

SOURCE

RNA pol σ 70 (2G10) is a mouse monoclonal antibody specific for an epitope mapping between amino acids 470-486 of RNA polymerase σ factor 70 of *E. coli* origin.

PRODUCT

Each vial contains 100 μ g IgG_{2b} in 1.0 ml of PBS with < 0.1% sodium azide and 0.1% gelatin.

APPLICATIONS

RNA pol σ 70 (2G10) is recommended for detection of RNA pol σ 70 of *E. coli* origin by Western Blotting (starting dilution to be determined by researcher, dilution range 1:100-1:5000) and immunoprecipitation [1-2 μ g per 100-500 μ g of total protein (1 ml of cell lysate)]; may cross-react with other σ factors from a wide variety of bacteria, including *E. coli* σ factor F; non cross-reactive with *Borellia burgdorferi*.

RECOMMENDED SECONDARY REAGENTS

To ensure optimal results, the following support (secondary) reagents are recommended: 1) Western Blotting: use goat anti-mouse IgG-HRP: sc-2005 (dilution range: 1:2000-1:32,000) or Cruz Marker™ compatible goat anti-mouse IgG-HRP: sc-2031 (dilution range: 1:2000-1:5000), Cruz Marker™ Molecular Weight Standards: sc-2035, TBS Blotto A Blocking Reagent: sc-2333 and Western Blotting Luminol Reagent: sc-2048. 2) Immunoprecipitation: use Protein A/G PLUS-Agarose: sc-2003 (0.5 ml agarose/2.0 ml).

STORAGE

For immediate and continuous use, store at 4° C for up to one month. For sporadic use, freeze in working aliquots in order to avoid repeated freeze/thaw cycles. If turbidity is evident upon prolonged storage, clarify solution by centrifugation.

RESEARCH USE

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