SANTA CRUZ BIOTECHNOLOGY, INC.

CstF-64 (H-300): sc-28201

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The Power to Question

BACKGROUND

Polyadenylation of mRNA precursors is a two-step reaction that requires multiple protein factors. The first step, endonucleolytic cleavage of polyadenylation substrates, requires CstF (cleavage stimulation factor), a heterotrimer that is composed of three distinct subunits. CstF-64 contains an RNA binding domain and is responsible for the RNA binding activity of CstF. CstF-64 is expressed in all somatic cells and in pre- and postmeiotic, but not meiotic, germ cells. However, a large variant of CstF-64, called τ CstF-64, is abundantly expressed in meiotic and postmeiotic cells in the testis and to a lesser extent in the brain, and promotes the germ cell pattern of polyadenylation. The gene encoding CstF-64 (also designated CSTF2) maps to the X chromosome, whereas τ CstF-64 is encoded by an autosomal gene. The increase in CstF-64 concentration during B cell activation switches IgM heavy chain mRNA expression from membrane-bound to secreted forms, suggesting that CstF-64 plays a key role in regulating IgM heavy chain expression during B cell differentiation.

REFERENCES

1. Takagaki, Y., et al. 1990. A multisubunit factor, CstF, is required for polyadenylation of mammalian pre-mRNAs. Genes Dev. 4: 2112-2120.
2. Takagaki, Y., et al. 1996. The polyadenylation factor CstF-64 regulates alternative processing of IgM heavy chain pre-mRNA during B cell differentiation. Cell 87: 941-952.
3. Takagaki, Y., et al. 1998. Levels of polyadenylation factor CstF-64 control IgM heavy chain mRNA accumulation and other events associated with B cell differentiation. Mol. Cell 2: 761-771.
4. Kleiman, F.E., et al. 1999. Functional interaction of BRCA1-associated BARD1 with polyadenylation factor CstF-50. Science 285: 1576-1579.
5. Wallace, A.M., et al. 1999. Two distinct forms of the 64,000 Mr protein of the cleavage stimulation factor are expressed in mouse male germ cells. Proc. Natl. Acad. Sci. USA 96: 6763-6768.
6. Takagaki, Y., et al. 2000. Complex protein interactions within the human polyadenylation machinery identify a novel component. Mol. Cell. Biol. 20: 1515-1525.

CHROMOSOMAL LOCATION

Genetic locus: CSTF2 (human) mapping to Xq22.1, CSTF2T (human) mapping to 10q21.1; Cstf2 (mouse) mapping to X E3, Cstf2 (mouse) mapping to 19 C1.

SOURCE

CstF-64 (H-300) is a rabbit polyclonal antibody raised against amino acids 278-577 mapping at the C-terminus of CstF-64 of human origin.

PRODUCT

Each vial contains 200 µg IgG in 1.0 ml of PBS with < 0.1% sodium azide and 0.1% gelatin.

STORAGE

Store at 4° C, **DO NOT FREEZE**. Stable for one year from the date of shipment. Non-hazardous. No MSDS required.

APPLICATIONS

CstF-64 (H-300) is recommended for detection of CstF-64 and CstF-64T of mouse, rat and human origin by Western Blotting (starting dilution 1:200, dilution range 1:100-1:1000), immunoprecipitation [1-2 µg per 100-500 µg of total protein (1 ml of cell lysate)], immunofluorescence (starting dilution 1:50, dilution range 1:50-1:500) and solid phase ELISA (starting dilution 1:30, dilution range 1:30-1:3000).

CstF-64 (H-300) is also recommended for detection of CstF-64 and CstF-64T in additional species, including equine, canine, bovine and porcine.

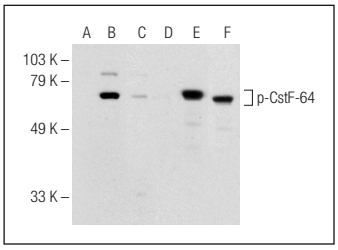
Molecular Weight of CstF-64: 64 kDa.

Positive Controls: CstF-64T (h): 293T Lysate: sc-114748, HeLa + PMA nuclear extract: sc-2121 or BJAB nuclear extract: sc-2145.

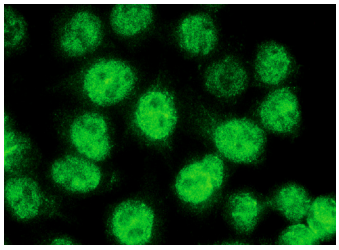
RECOMMENDED SECONDARY REAGENTS

To ensure optimal results, the following support (secondary) reagents are recommended: 1) Western Blotting: use goat anti-rabbit IgG-HRP: sc-2004 (dilution range: 1:2000-1:100,000) or Cruz Marker™ compatible goat anti-rabbit IgG-HRP: sc-2030 (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). 3) Immunofluorescence: use goat anti-rabbit IgG-FITC: sc-2012 (dilution range: 1:100-1:400) or goat anti-rabbit IgG-TR: sc-2780 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941.

DATA



Western blot analysis of CstF-64 phosphorylation in non-transfected: sc-117752 (**A**,**D**), untreated human CstF-64T transfected: sc-114748 (**B**,**E**) and lambda protein phosphatase (sc-200312A) treated human CstF-64T transfected: sc-114748 (**C**,**F**) 293T whole cell lysates. Antibodies tested include p-CstF-64 (Ser 83)-R: sc-16480-R (**A**,**B**,**C**) and CstF-64 (H-300): sc-28201 (**D**,**E**,**F**).



CstF-64 (H-300): sc-28201. Immunofluorescence staining of methanol-fixed HeLa cells showing nuclear localization.

RESEARCH USE

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