

Cdc7 (h): 293T Lysate: sc-111960

BACKGROUND

The Dbf4/Cdc7 protein kinase is essential for the activation of replication origins during S phase. Cdc7/Dbf4 efficiently phosphorylates several proteins that are required for the initiation of DNA replication, including five of the six minichromosome maintenance (Mcm) proteins and the p180 subunit of DNA polymerase α -primase. This protein complex consists of the catalytic subunit Cdc7 associating with the regulatory and activating subunit Dbf4. The kinase activity of the complex is regulated throughout the cell cycle, mainly by fluctuating levels of Dbf4. Cdc7 is consistently expressed throughout the cell cycle, while the expression of Dbf4 is absent during G₁ phase and accumulates during S and G₂ phases. The anaphase-promoting complex rapidly degrades Dbf4 at the time of chromosome segregation, and the stability of Dbf4 remains low during pre-start G₁ phase. The coordinated degradation of Dbf4 and the time of chromosome separation is important to ensuring that prereplicative complexes, which assemble after chromosome segregation, do not immediately refire.

REFERENCES

1. Bousset, K. and Diffley, J.F. 1998. The Cdc7 protein kinase is required for origin firing during S phase. *Genes Dev.* 12: 480-490.
2. Lepke, M., Putter, V., Staib, C., Kneissl, M., Berger, C., Hoehn, K., Nanda, I., Schmid, M. and Grummt, F. 1999. Identification, characterization and chromosomal localization of the cognate human and murine DBF4 genes. *Mol. Gen. Genet.* 262: 220-229.
3. Masai, H., Sato, N., Takeda, T. and Arai, K. 1999. Cdc7 kinase complex as a molecular switch for DNA replication. *Front. Biosci.* 4: 834-840.
4. Weinreich, M. and Stillman, B. 1999. Cdc7p-Dbf4p kinase binds to chromatin during S phase and is regulated by both the APC and the RAD53 checkpoint pathway. *EMBO J.* 18: 5334-5346.
5. Jiang, W., McDonald, D., Hope, T.J. and Hunter, T. 1999. Mammalian Cdc7-Dbf4 protein kinase complex is essential for initiation of DNA replication. *EMBO J.* 18: 5703-5713.
6. Pasero, P., Duncker, B.P., Schwob, E. and Gasser, S.M. 1999. A role for the Cdc7 kinase regulatory subunit Dbf4p in the formation of initiation-competent origins of replication. *Genes Dev.* 13: 2159-2176.
7. Ferreira, M.F., Santocanale, C., Drury, L.S. and Diffley, J.F. 2000. Dbf4p, an essential S phase-promoting factor, is targeted for degradation by the anaphase-promoting complex. *Mol. Cell. Biol.* 20: 242-248.
8. Masai, H., Taniyama, C., Ogino, K., Matsui, E., Kakusho, N., Matsumoto, S., Kim, J.M., Ishii, A., Tanaka, T., Kobayashi, T., Tamai, K., Ohtani, K. and Arai, K. 2006. Phosphorylation of MCM4 by Cdc7 kinase facilitates its interaction with Cdc45 on the chromatin. *J. Biol. Chem.* 281: 39249-39261.
9. Tenca, P., Brotherton, D., Montagnoli, A., Rainoldi, S., Albanese, C. and Santocanale, C. 2007. Cdc7 is an active kinase in human cancer cells undergoing replication stress. *J. Biol. Chem.* 282: 208-215.

STORAGE

Store at -20° C. Repeated freezing and thawing should be minimized. Sample vial should be boiled once prior to use. Non-hazardous. No MSDS required.

CHROMOSOMAL LOCATION

Genetic locus: CDC7 (human) mapping to 1p22.2.

PRODUCT

Cdc7 (h): 293T Lysate represents a lysate of human Cdc7 transfected 293T cells and is provided as 100 μ g protein in 200 μ l SDS-PAGE buffer.

APPLICATIONS

Cdc7 (h): 293T Lysate is suitable as a Western Blotting positive control for human reactive Cdc7 antibodies. Recommended use: 10-20 μ l per lane.

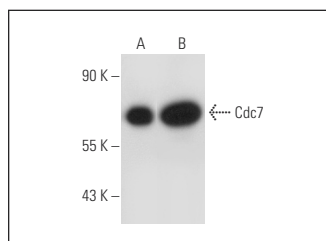
Control 293T Lysate: sc-117752 is available as a Western Blotting negative control lysate derived from non-transfected 293T cells.

Cdc7 (SPM171): sc-56275 is recommended as a positive control antibody for Western Blot analysis of enhanced human Cdc7 expression in Cdc7 transfected 293T cells (starting dilution 1:100, dilution range 1:100-1:1,000).

RECOMMENDED SUPPORT REAGENTS

To ensure optimal results, the following support reagents are recommended:
1) Western Blotting: use m-IgG κ BP-HRP: sc-516102 or m-IgG κ BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz Marker™
Molecular Weight Standards: sc-2035, UltraCruz® Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048.

DATA



Cdc7 (SPM171): sc-56275. Western blot analysis of Cdc7 expression in non-transfected: sc-117752 (A) and human Cdc7 transfected: sc-111960 (B) 293T whole cell lysates.

RESEARCH USE

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

PROTOCOLS

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