

# Cdt1 (N-20): sc-18558

## BACKGROUND

Human Cdt1 is a nuclear localizing replication initiation factor that is expressed only during the G<sub>1</sub> and S phases of the cell cycle. In conjunction with Cdc18, Cdt1 is required to load the MCM protein Cdc21 onto chromatin at the end of mitosis which is necessary to initiate DNA replication. After S phase onset, Cdt1 protein levels decrease and are barely detectable in cells in early S phase or G<sub>2</sub>. However, Cdt1 mRNA is expressed in S phase-arrested cells, and its levels do not change dramatically during the cell cycle, suggesting that proteolytic degradation rather than transcriptional controls ensure proper accumulation of Cdt1. Cdt1 can associate with the DNA replication inhibitor geminin, which is present in the S and G<sub>2</sub> phases of the cell cycle. Inhibition of DNA replication by Geminin in cell-free DNA replication extracts can be reversed by the addition of excess Cdt1. Geminin may be responsible for preventing inappropriate origin firing by targeting Cdt1.

## REFERENCES

- Hofmann, J.F. and Beach, D. 1994. Cdt1 is an essential target of the Cdc10/Sct1 transcription factor: requirement for DNA replication and inhibition of mitosis. *EMBO J.* 13: 425-434.
- Wohlschlegel, J.A., et al. 2000. Inhibition of eukaryotic DNA replication by Geminin binding to Cdt1. *Science* 290: 2309-2312.
- Maiorano, D., et al. 2000. XCdt1 is required for the assembly of pre-replicative complexes in *Xenopus laevis*. *Nature* 404: 622-625.
- Nishitani, H., et al. 2000. The Cdt1 protein is required to license DNA for replication in fission yeast. *Nature* 404: 625-628.
- Nishitani, H., et al. 2001. The human licensing factor for DNA replication Cdt1 accumulates in G1 and is destabilized after initiation of S-phase. *J. Biol. Chem.* 276: 44905-44911.

## CHROMOSOMAL LOCATION

Genetic locus: CDT1 (human) mapping to 16q24.3.

## SOURCE

Cdt1 (N-20) is an affinity purified goat polyclonal antibody raised against a peptide mapping near the N-terminus of Cdt1 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.

Blocking peptide available for competition studies, sc-18558 P, (100 µg peptide in 0.5 ml PBS containing < 0.1% sodium azide and 0.2% BSA).

## STORAGE

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

## RESEARCH USE

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

## APPLICATIONS

Cdt1 (N-20) is recommended for detection of Cdt1 of 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).

Suitable for use as control antibody for Cdt1 siRNA (h): sc-37544, Cdt1 shRNA Plasmid (h): sc-37544-SH and Cdt1 shRNA (h) Lentiviral Particles: sc-37544-V.

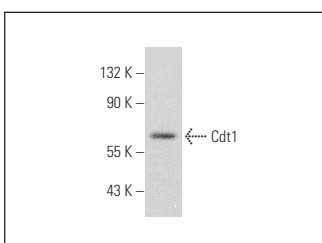
Molecular Weight of Cdt1: 65 kDa.

Positive Controls: Jurkat nuclear extract: sc-2132.

## RECOMMENDED SECONDARY REAGENTS

To ensure optimal results, the following support (secondary) reagents are recommended: 1) Western Blotting: use donkey anti-goat IgG-HRP: sc-2020 (dilution range: 1:2000-1:100,000) or Cruz Marker™ compatible donkey anti-goat IgG-HRP: sc-2033 (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 donkey anti-goat IgG-FITC: sc-2024 (dilution range: 1:100-1:400) or donkey anti-goat IgG-TR: sc-2783 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941.

## DATA



Cdt1 (N-20): sc-18558. Western blot analysis of Cdt1 expression in Jurkat nuclear extract.

## SELECT PRODUCT CITATIONS

- Sugimoto, N. 2004. Cdt1 Phosphorylation by cyclin A-dependent kinases negatively regulates its function without affecting Geminin binding. *J. Biol. Chem.* 279: 19691-19697.

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