Datasheet for ABIN487481

anti-CDC7 antibody

3 Images

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Overview

Quantity:	0.1 mg
Target:	CDC7
Reactivity:	Human
Host:	Mouse
Clonality:	Monoclonal
Conjugate:	This CDC7 antibody is un-conjugated
Application:	Western Blotting (WB), Immunohistochemistry (Paraffin-embedded Sections) (IHC (p)), Immunoprecipitation (IP)

Product Details

Immunogen:	Full-length Human CDC7 fusion protein. Remarks: Hybridoma was established by fusion of Mouse myeloma cell NS-2 with Balb/cmouse splenocyte.
Clone:	DCS-342
Isotype:	IgG2b
Specificity:	This antibody reacts with Human CDC7
Cross-Reactivity (Details):	Species reactivity (tested):Human.
Characteristics:	Synonyms: CDC7L1, CDC7-related kinase, HsCdc7, huCdc7, Cell division cycle 7-related protein kinase
Purification:	Protein-A Sepharose Chromatography.

Target Details

Target:	CDC7
Alternative Name:	CDC7 (CDC7 Products)
Background:	Cell division cycle-7 (CDC7) was initially discovered as a temperature sensitive cell cycle mutant in Saccharomyces cerevisiae. CDC7 is required in the mitotic cell cycle for the initiation of DNA synthesis. CDC7 forms a complex with a Dbf4-related regulatory subunit to generate an active Ser/Thr kinase that functions in DNA replication, repair, and mitotic recombination. 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 alpha-primase. CDC7p-Dbf4p kinase binds to chromatin during S phase and is regulated by both the APC and the RAD53 checkpoint pathway. A human homolog of the yeast CDC7 is over-expressed in some tumors and transformed cell lines.Synonyms: CDC7-related kinase, CDC7L1, Cell division cycle 7-related protein kinase, HsCdc7, huCdc7
Gene ID:	8317
UniProt:	O00311
Pathways:	Mitotic G1-G1/S Phases, DNA Replication

Application Details

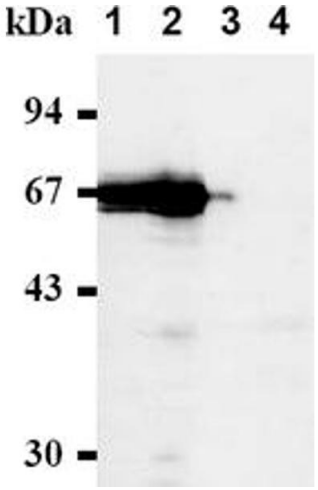
Application Notes:	Westzern Blotting: 0.2 µg/mLImmunoprecipitation: 3 µg/200-300 µL of cell extract. Positive Controls: HeLa, Raji cells. Immunohistochemistry: 1-5 µg/mLHeat treatment is necessary for Paraffin Embedded Sections. Microwave oven: 2 times for 10 minutes each in 10 mM citrate buffer (pH 6.5)Positive Control: Tonsil Tissue. Detailed procedure is provided in Protocols. Other applications not tested. Optimal dilutions are dependent on conditions and should be determined by the user.
Protocol:	SDS-PAGE & Western Blotting1) Wash the cells 3 times with PBS and suspend with 10 volume of cold Lysis buffer (50 mMTris-HCl, pH 7. 2, 250 mM NaCl, 0. 1% NP-40, 2 mM EDTA, 10% glycerol) containingappropriate protease inhibitors. Incubate it at 4°C with rotating for 30 minutes, thensonicate briefly (up to 10 seconds). 2) Centrifuge the tube at 12,000 x g for 10 minutes at 4°C and transfer the supernatant toanother tube. Measure the protein concentration of the supernatant and add the Lysisbuffer to make 8 mg/mL solution. 3) Mix the sample with equal volume of Laemmli's sample buffer. 4) Boil the samples for 2 minutes and centrifuge. Load 10 µL of the sample per lane in a 1mm thick SDS-polyacrylamide gel for electrophoresis. 5) Blot the protein to a polyvinylidene difluoride (PVDF) membrane at 1 mA/cm2 for 1 hourin a semi-dry transfer system. (Transfer Buffer: 25 mM Tris, 190 mM glycine, 20% MeOH). See the

Application Details

	manufacture's manual for the transfer procedure. 6) To reduce nonspecific binding, soak the membrane in 10% skimmed milk (in PBS, pH7. 2) for 1 hour at room temperature, or overnight at 4°C. 7) Incubate the membrane with the anti-CDC7 monoclonal antibody (0. 2 µg/mL) dilutedwith 1% skimmed milk (in PBS, pH 7. 2) for 1 hour at room temperature. 8) Wash the membrane with PBS (5 minutes x 6 times). 9) Incubate the membrane with the 1: 10000 HRP-conjugated anti-mouse IgG diluted with1% skimmed milk (in PBS, pH 7. 2) for 1 hour at room temperature. 10) Wash the membrane with PBS (5 minutes x 6 times). 11) Wipe excess buffer from the membrane, then incubate it with appropriatechemiluminescence reagents for 1 minute. Remove extra reagent from the membrane bydabbing with a paper towel, and seal it in plastic wrap. 12) Expose to an X-ray film in a dark room for 5 minutes. Develop the film as usual. Theconditions for exposure and development may vary. Positive Controls for Western blotting: HeLa, Raji. Immunoprecipitation1) Wash the cells 3 times with PBS and suspend with 10 volume of cold Lysis buffer (50 mMTris-HCl, pH 7. 2, 250 mM NaCl, 0. 1% NP-40, 2 mM EDTA, 10% glycerol) containingappropriate protease inhibitors. Incubate it at 4°C with rotating for 30 minutes, thensonicate briefly (up to 10 seconds). 2) Centrifuge the tube at 12,000 x g for 10 minutes at 4°C and transfer the supernatant toanother tube. 3) Add 3 µg of the anti-CDC7 monoclonal antibody into 250 µL of the supernatant. Mix welland incubate with gentle agitation for 30-120 minutes at 4°C. Add 20 µL of 50% ProteinA-agarose beads resuspended in the Lysis buffer. Mix well and incubate with gentleagitation for 60 minutes at 4°C. 4) Wash the beads 3-5 times with ice-cold Lysis buffer (centrifuge the tube at 2,500 x g for
Restrictions:	For Research Use only

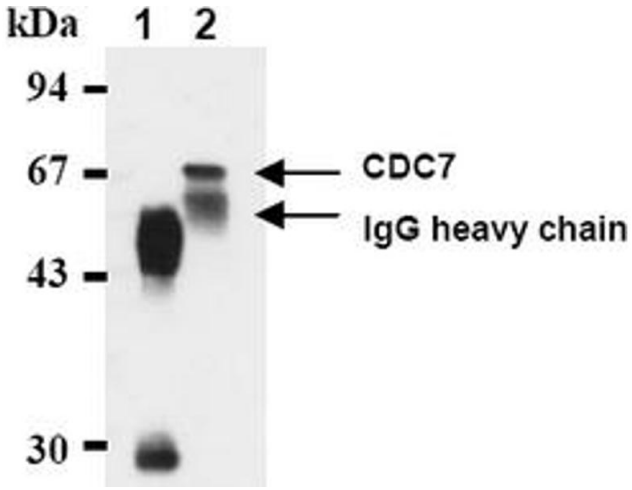
Handling

Concentration:	1.0 mg/mL
Buffer:	PBS, pH 7.2 containing 50 % Glycerol without preservatives.
Preservative:	Without preservative
Storage:	-20 °C
Storage Comment:	Store the antibody undiluted at -20 °C. Shelf life: one year from despatch.
Expiry Date:	12 months



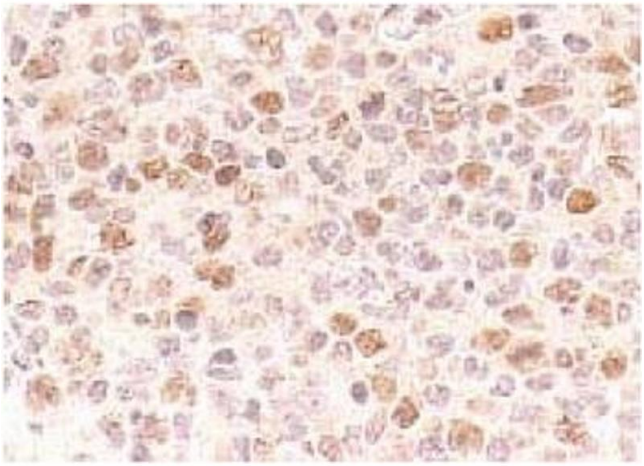
Western Blotting

Image 1.



Western Blotting

Image 2.



Immunohistochemistry

Image 3.