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anti-CDC5L antibody (AA 109-303)

Images



Publications



Overview

Quantity:	50 μg
Target:	CDC5L
Binding Specificity:	AA 109-303
Reactivity:	Human, Mouse, Rat, Dog, Chicken
Host:	Mouse
Clonality:	Monoclonal
Conjugate:	This CDC5L antibody is un-conjugated
Application:	Western Blotting (WB), BioImaging (BI)

Product Details

Product Details	
Immunogen:	Rat CDC5L aa. 109-303
Clone:	21-CDC5L
Isotype:	lgG1
Cross-Reactivity:	Chicken, Dog (Canine), Human, Mouse (Murine)
Characteristics:	1. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
	2. This antibody has been developed and certified for the bioimaging application. However, a
	routine bioimaging test is not performed on every lot. Researchers are encouraged to titrate the
	reagent for optimal performance.
	3. Triton is a trademark of the Dow Chemical Company.
	4. Source of all serum proteins is from USDA inspected abattoirs located in the United States.
	5. Caution: Sodium azide yields highly toxic hydrazoic acid under acidic conditions. Dilute azide

Product Details

compounds in running water before discarding to avoid accumulation of potentially explosive deposits in plumbing.

6. Please refer to us for technical protocols.

Purification:

The monoclonal antibody was purified from tissue culture supernatant or ascites by affinity chromatography.

Target Details

Target: CDC5L

Alternative Name: CDC5L (CDC5L Products)

Background:

Splicing, the removal of introns from pre-mRNA, is mediated by spliceosomal complexes and occurs in two distinct catalytic steps. The first step involves cleavage of the 5' exon and the production of a lariat intermediate. In the second step, the 3'-splice site is cleaved and the exons are fused with concomitant release of the intron lariat. The spliceosome contains multiple snRNPs and a number of non-snRNP splicing factors. CDC5L is a non-snRNP component of the splicesome that is homologous to the yeast Cdc5 protein. The sequence for CDC5L contgains a helix-turn-helix DNA binding domain (DBD), four nuclear localization signals, and a hydrophilic, proline-rich central region that is similar to the transcriptional activating domain in Myb family transcription factors. CDC5L can interact with the nuclear PP1 inhibitor NIPP-1 and the WD40 domain protein PLRG1. Both of these interactions are critical for pre-mRNA splicing and may also be important for G 2/M phase transition. In addition, CDC5L translocates from the cytoplasm to the nucleus in the presence of serum. Thus, CDC5L localization and splicesome-regulating activity may be controlled by mitogen-activated signal transduction pathways.

Molecular Weight:

105 kDa

Pathways:

Activation of Innate immune Response, Chromatin Binding

Application Details

Application Notes:

Bioimaging

- 1. Seed the cells in appropriate culture medium at \sim 10,000 cells per well in an 96-well Imaging Plate and culture overnight.
- 2. Remove the culture medium from the wells, and fix the cells by adding 100 myl of Fixation Buffer to each well. Incubate for 10 minutes at room temperature (RT).
- 3. Remove the fixative from the wells, and permeabilize the cells using either 90% methanol, or

Triton™ X-100: a. Add 100 myl of -20°C 90% methanol to each well and incubate for 5 minutes at RT. OR b. Add 100 myl of 0.1% Triton™ X-100 to each well and incubate for 5 minutes at RT.

- 4. Remove the permeabilization buffer, and wash the wells twice with 100 myl of 1x PBS.
- 5. Remove the PBS, and block the cells by adding 100 myl of to each well. Incubate for 30 minutes at RT.
- 6. Remove the blocking buffer and add 50 myl of the optimally titrated primary antibody (diluted in Stain Buffer) to each well, and incubate for 1 hour at RT.
- 7. Remove the primary antibody, and wash the wells three times with 100 myl of 1x PBS.
- 8. Remove the PBS, and add the second step reagent at its optimally titrated concentration in 50 myl to each well, and incubate in the dark for 1 hour at RT.
- 9. Remove the second step reagent, and wash the wells three times with 100 myl of 1x PBS.
- 10. Remove the PBS, and counter-stain the nuclei by adding 200 myl per well of 2 myg/ml Hoechst 33342 in $1 \times$ PBS to each well at least 15 minutes before imaging.
- 11. View and analyze the cells on an appropriate imaging instrument.

Comment:	Related Products: ABIN968546, ABIN967389
Restrictions:	For Research Use only

Handling

Format:	Liquid
Concentration:	250 μg/mL
Buffer:	Aqueous buffered solution containing BSA, glycerol, and ≤0.09 % sodium azide.
Preservative:	Sodium azide
Precaution of Use:	This product contains Sodium azide: a POISONOUS AND HAZARDOUS SUBSTANCE which should be handled by trained staff only.
Storage:	-20 °C
Storage Comment:	Store undiluted at -20°C.

Publications

Product cited in:

Ajuh, Sleeman, Chusainow, Lamond: "A direct interaction between the carboxyl-terminal region of CDC5L and the WD40 domain of PLRG1 is essential for pre-mRNA splicing." in: **The Journal of biological chemistry**, Vol. 276, Issue 45, pp. 42370-81, (2001) (PubMed).

Ajuh, Kuster, Panov, Zomerdijk, Mann, Lamond: "Functional analysis of the human CDC5L complex and identification of its components by mass spectrometry." in: **The EMBO journal**, Vol. 19, Issue 23, pp. 6569-81, (2000) (PubMed).

Boudrez, Beullens, Groenen, Van Eynde, Vulsteke, Jagiello, Murray, Krainer, Stalmans, Bollen: "NIPP1-mediated interaction of protein phosphatase-1 with CDC5L, a regulator of pre-mRNA splicing and mitotic entry." in: **The Journal of biological chemistry**, Vol. 275, Issue 33, pp. 25411-7, (2000) (PubMed).

Bernstein, Coughlin: "Pombe Cdc5-related protein. A putative human transcription factor implicated in mitogen-activated signaling." in: **The Journal of biological chemistry**, Vol. 272, Issue 9, pp. 5833-7, (1997) (PubMed).

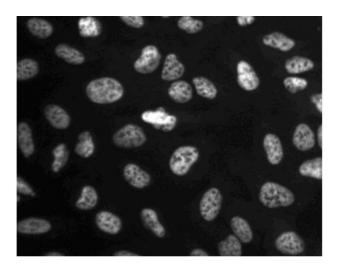
Images



Western Blotting

Image 1. Western blot analysis of CDC5L on a rat cerebrum lysate. Lane 1: 1:5000, lane 2: 1:10000, lane 3: 1:20000 dilution of the CDC5L antibody.





Immunofluorescence

Image 2. Immunofluorescent staining of U-2 OS (ATCC HTB-96) cells. Cells were seeded in a 96 well imaging plate at ~ 10 000 cells per well. After overnight incubation, cells were stained using the alcohol perm protocol and the anti-CDC5L antibody. The second step reagent was FITC goat anti mouse Ig. Images were taken on a BD Pathway™ 855 bioimaging system using a 20x objective. This antibody also stained HeLa (ATCC CCL-2) cells and worked with both the Triton™ X-100 and alcohol perm protocols. This antibody is



not recommended for staining A549 cells.