Datasheet for ABIN968445

anti-Cyclin A antibody (AA 26-144)

3 Images | 5 Publications

Go to Product page

Overview

Quantity:	150 µg
Target:	Cyclin A (CCNA2)
Binding Specificity:	AA 26-144
Reactivity:	Human
Host:	Mouse
Clonality:	Monoclonal
Conjugate:	This Cyclin A antibody is un-conjugated
Application:	Western Blotting (WB), BioImaging (BI)

Product Details

Immunogen:	Human Cyclin A aa. 26-144
Clone:	25-Cyclin A
Isotype:	IgG1
Characteristics:	1. Since applications vary, each investigator should titrate the reagent to obtain optimal results. 2. Please refer to us for technical protocols. 3. 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. 4. Caution: Sodium azide yields highly toxic hydrazoic acid under acidic conditions. Dilute azide compounds in running water before discarding to avoid accumulation of potentially explosive deposits in plumbing.

Product Details

	5. Source of all serum proteins is from USDA inspected abattoirs located in the United States. 6. Triton is a trademark of the Dow Chemical Company.
Purification:	The monoclonal antibody was purified from tissue culture supernatant or ascites by affinity chromatography.

Target Details

Target:	Cyclin A (CCNA2)
Alternative Name:	Cyclin A (CCNA2 Products)
Background:	Progression of the mammalian cell cycle is regulated by phosphorylation of many key proteins. Several classes of cyclins (A-E) act as regulatory subunits for cyclin-dependent kinases (cdks). These cyclin-cdk holoenzymes are essential for proper control of cell cycle progression. They phosphorylate and regulate a variety of substrates whose activity is required for cell cycle transitions. The temporal expression of cyclins is tightly regulated throughout the cell cycle by synthesis and degradation. Such regulation plays a critical role in controlling the enzymatic activity of the cdks. Cyclin A, one of the mitotic cyclins, activates Cdk2 near the start of S phase and is necessary for the initiation of DNA replication. In mammalian somatic cells, Cyclin A is required during S phase and passage through G2. The D and E type cyclins regulate passage through G1, while Cyclin B is a critical regulator of mitosis. It has been shown in a number of species that mutation or disruption of normal Cyclin A expression causes cells to arrest at G2. Cyclin A binds both the cdc2 (Cdk1) and Cdk2 kinases and may also have a role in mitotic dependence on S phase completion.
Molecular Weight:	60 kDa
Pathways:	PI3K-Akt Signaling, Cell Division Cycle, AMPK Signaling, Mitotic G1-G1/S Phases, DNA Replication, M Phase, Synthesis of DNA

Application Details

Application Notes:	Bioimaging 1. Seed the cells in appropriate culture medium at ~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

Application Details

	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 1× 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 1× 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 1× PBS. 10. Remove the PBS, and counter-stain the nuclei by adding 200 myl per well of 2 myg/ml Hoechst 33342 in 1× PBS to each well at least 15 minutes before imaging. 11. View and analyze the cells on an appropriate imaging instrument.
Comment:	Related Products: ABIN967389, ABIN968533
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:	Saitoh, Pizzi, Wang: "Perturbation of SUMOlation enzyme Ubc9 by distinct domain within nucleoporin RanBP2/Nup358." in: **The Journal of biological chemistry**, Vol. 277, Issue 7, pp. 4755-63, (2002) (PubMed). Henglein, Chenivesse, Wang, Eick, Bréchot: "Structure and cell cycle-regulated transcription of

Publications

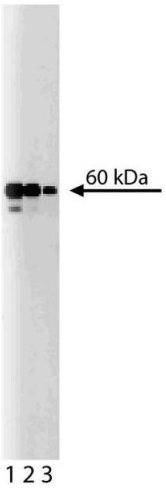
the human cyclin A gene." in: **Proceedings of the National Academy of Sciences of the United States of America**, Vol. 91, Issue 12, pp. 5490-4, (1994) (PubMed).

Pines, Hunter: "The differential localization of human cyclins A and B is due to a cytoplasmic retention signal in cyclin B." in: **The EMBO journal**, Vol. 13, Issue 16, pp. 3772-81, (1994) (PubMed).

Pines: "Cyclins and cyclin-dependent kinases: take your partners." in: **Trends in biochemical sciences**, Vol. 18, Issue 6, pp. 195-7, (1993) (PubMed).

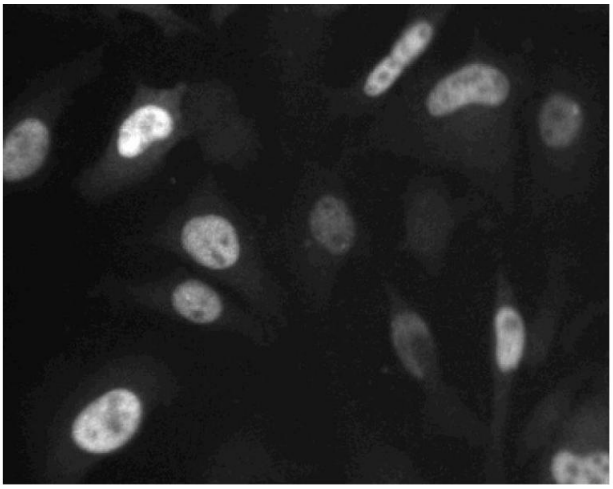
Giordano, Whyte, Harlow, Franza, Beach, Draetta: "A 60 kd cdc2-associated polypeptide complexes with the E1A proteins in adenovirus-infected cells." in: **Cell**, Vol. 58, Issue 5, pp. 981-90, (1989) (PubMed).

Images



Western Blotting

Image 1. Western blot analysis of Cyclin A on a A431 lysate. Lane 1: 1:250, lane 2: 1:500, lane 3: 1:1000 dilution of the Cyclin A antibody.



Immunofluorescence

Image 2. Immunofluorescent staining of HeLa (ATCC CCL-2) 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-Cyclin A antibody. The second step reagent was FITC goat anti mouse Ig. The image was taken on a BD Pathway™ 855 Bioimager system using a 20x objective. This antibody also stained A549 (ATCC CCL-185) and U-2 OS (ATCC HTB-96)

cells using both the Triton™ X-100 and alcohol perm protocols.

Image 3.