



Product Datasheet

Anti-Cyclin B1/CCNB1 Antibody (orb654315)

Description	Anti-Cyclin B1/CCNB1 Antibody. Tested in ELISA, Flow Cytometry, IF, IHC, ICC, WB applications. This antibody reacts with Human, Mouse, Rat.
Species/Host	Rabbit
Reactivity	Human, Mouse, Rat
Conjugation	Unconjugated
Tested Applications	ELISA, FC, ICC, IF, IHC, WB
Immunogen	E.coli-derived human Cyclin B1/CCNB1 recombinant protein (Position: M1-L383).
Form/Appearance	Lyophilized
Concentration	Adding 0.2 ml of distilled water will yield a concentration of 500 μ g/ml.
Storage	Maintain refrigerated at 2-8°C for up to 2 weeks. For long term storage store at - 20°C in small aliquots to prevent freeze-thaw cycles.
Note	For research use only
Application notes	Western blot, 0.25-0.5µg/ml, Human, Mouse, Rat Immunohistochemistry (Paraffin-embedded Section), 0.5-1µg/ml, Human, Rat Immunocytochemistry/Immunofluorescence, 2µg/ml, Human Flow Cytometry (Fixed), 1-3µg/1x106 cells, Human ELISA, 0.1-0.5µg/ml, Add 0.2ml of distilled water will yield a concentration of 500ug/ml
lsotype	Rabbit IgG
Clonality	Polyclonal
Antibody Type	Primary Antibody
MW	55 kDa
Uniprot ID	P14635

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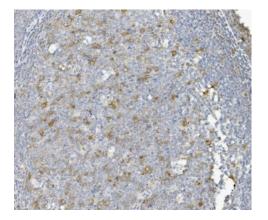
7 Signet Court, Swann's Road, Cambridge, CB5 8LA, United Kingdom Email: <u>info@biorbyt.com</u>, <u>support@biorbyt.com</u> Phone: <u>+44 (0) 1223 859-353</u> | Fax: <u>+1 (415) 651-8558</u>

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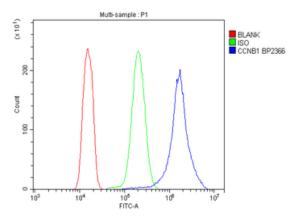


Expiration Date

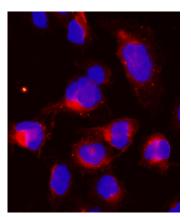
12 months from date of receipt.



IHC analysis of CCNB1 using anti-CCNB1 antibody. CCNB1 was detected in paraffin-embedded section of human tonsil tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1 μ g/ml rabbit anti-CCNB1 Antibody overnight at 4°C. Biotinylated goat anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Strepavidin-Biotin-Complex (SABC) with DAB as the chromogen.



Flow Cytometry analysis of A431 cells using anti-CCNB1 antibody. Overlay histogram showing A431 cells (Blue line). To facilitate intracellular staining, cells were fixed with 4% paraformaldehyde and permeabilized with permeabilization buffer. The cells were blocked with 10% normal goat serum. And then incubated with rabbit anti-CCNB1 Antibody (1 μ g/1x10^6 cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-rabbit IgG (5-10 μ g/1x10^6 cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was rabbit IgG (1 μ g/1x10^6) used under the same conditions. Unlabelled sample without incubation with primary antibody and secondary antibody (Red line) was used as a blank control.



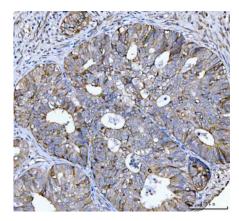
IF analysis of CCNB1 using anti-CCNB1 antibody. CCNB1 was detected in immunocytochemical section of U20S cells. Enzyme antigen retrieval was performed using IHC enzyme antigen retrieval reagent for 15 mins. The cells were blocked with 10% goat serum. And then incubated with 2 μ g/mL rabbit anti-CCNB1 Antibody overnight at 4°C. Cy3 Conjugated Goat Anti-Rabbit IgG was used as secondary antibody at 1:100 dilution and incubated for 30 minutes at 37°C. The section was counterstained with DAPI. Visualize using a fluorescence microscope and filter sets appropriate for the label used.

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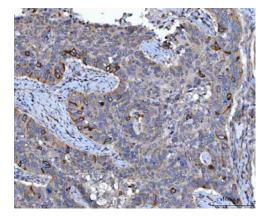
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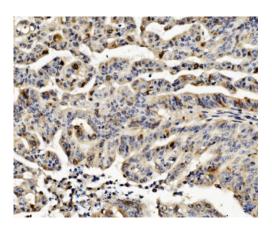




IHC analysis of CCNB1 using anti-CCNB1 antibody. CCNB1 was detected in paraffin-embedded section of human endometrial adenocarcinoma tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1 μ g/ml rabbit anti-CCNB1 Antibody overnight at 4°C. Biotinylated goat anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Strepavidin-Biotin-Complex (SABC) with DAB as the chromogen.



IHC analysis of CCNB1 using anti-CCNB1 antibody. CCNB1 was detected in paraffin-embedded section of human laryngeal squamous cell carcinoma tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1 μ g/ml rabbit anti-CCNB1 Antibody overnight at 4°C. Biotinylated goat anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Strepavidin-Biotin-Complex (SABC) with DAB as the chromogen.



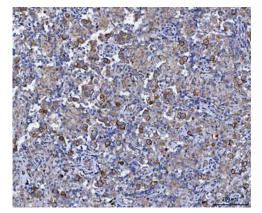
IHC analysis of CCNB1 using anti-CCNB1 antibody. CCNB1 was detected in paraffin-embedded section of human rectal cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1 µg/ml rabbit anti-CCNB1 Antibody overnight at 4°C. Biotinylated goat anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Strepavidin-Biotin-Complex (SABC) with DAB as the chromogen.

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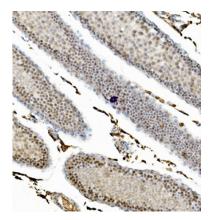
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IHC analysis of CCNB1 using anti-CCNB1 antibody. CCNB1 was detected in paraffin-embedded section of human seminoma testis tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1 μ g/ml rabbit anti-CCNB1 Antibody overnight at 4°C. Biotinylated goat anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Strepavidin-Biotin-Complex (SABC) with DAB as the chromogen.



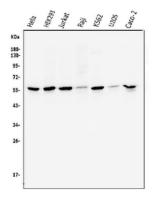
IHC analysis of CCNB1 using anti-CCNB1 antibody. CCNB1 was detected in paraffin-embedded section of rat testis tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1 μ g/ml rabbit anti-CCNB1 Antibody overnight at 4°C. Biotinylated goat anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Strepavidin-Biotin-Complex (SABC) with DAB as the chromogen.

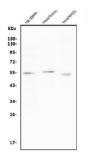
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Western blot analysis of CCNB1 using anti-CCNB1 antibody. Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 50 ug of sample under reducing conditions. Lane 1: human Hela whole cell lysates, Lane 2: human HEK293 whole cell lysates, Lane 3: human Jurkat whole cell lysates, Lane 4: human Raji whole cell lysates, Lane 5: human K562 whole cell lysates, Lane 6: human U2OS whole cell lysates, Lane 7: human CACO-2 whole cell lysates. After Electrophoresis, proteins were transferred to a Nitrocellulose membrane at 150 mA for 50-90 minutes. Blocked the membrane with 5% Non-fat Milk/ TBS for 1.5 hour at RT. The membrane was incubated with rabbit anti-CCNB1 antigen affinity purified polyclonal antibody at 0.5 µg/mL overnight at 4°C, then washed with TBS-0.1% Tween 3 times with 5 minutes each and probed with a goat anti-rabbit IgG-HRP secondary antibody at a dilution of 1:5000 for 1.5 hour at RT. The signal is developed using an Enhanced Chemiluminescent detection (ECL) kit with Tanon 5200 system. A specific band was detected for CCNB1 at approximately 55 KD. The expected band size for CCNB1 is at 55 KD.

Western blot analysis of CCNB1 using anti-CCNB1 antibody. Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 50 ug of sample under reducing conditions. Lane 1: rat spleen tissue lysates, Lane 2: mouse thymus tissue lysates, Lane 3: mouse NIH-3T3 whole cell lysates. After Electrophoresis, proteins were transferred to a Nitrocellulose membrane at 150 mA for 50-90 minutes. Blocked the membrane with 5% Non-fat Milk/ TBS for 1.5 hour at RT. The membrane was incubated with rabbit anti-CCNB1 antigen affinity purified polyclonal antibody at 0.5 µg/mL overnight at 4°C, then washed with TBS-0.1% Tween 3 times with 5 minutes each and probed with a goat anti-rabbit IgG-HRP secondary antibody at a dilution of 1:5000 for 1.5 hour at RT. The signal is developed using an Enhanced Chemiluminescent detection (ECL) kit with Tanon 5200 system. A specific band was detected for CCNB1 at approximately 55 KD. The expected band size for CCNB1 is at 55 KD.

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