



## Product Datasheet Anti-CD44 Antibody (orb402179)

**Description** Anti-CD44 Antibody. Tested in ELISA, Flow Cytometry, IF, IHC, IHC-F, 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, IHC-Fr, WB

**Immunogen** E. coli-derived human CD44 recombinant protein (Position: Q21-H259).

Form/Appearance Lyophilized

**Concentration** Adding 0.2 ml of distilled water will yield a concentration of 500 μg/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, 0.1-0.5μg/ml Immunohistochemistry (Paraffin-embedded Section),

0.5-1µg/ml Immunohistochemistry (Frozen Section), 0.5-1µg/ml

Immunocytochemistry,  $0.5-1\mu g/ml$  Immunofluorescence,  $2\mu g/ml$  Flow Cytometry (Fixed),  $1-3\mu g/1x106$  cells ELISA,  $0.1-0.5\mu g/ml$ . Add 0.2ml of distilled water will

yield a concentration of 500ug/ml

**Isotype** Rabbit IgG

**Clonality** Polyclonal

**Antibody Type** Primary Antibody

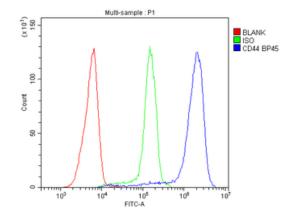
MW 82 kDa

Uniprot ID P16070

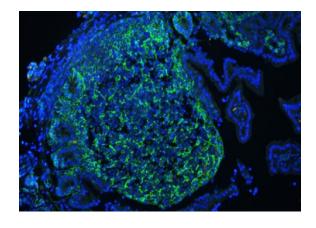


## **Expiration Date**

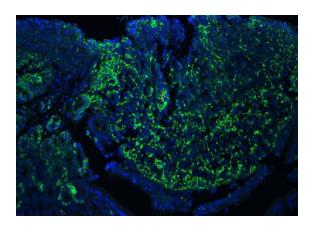
12 months from date of receipt.



Flow Cytometry analysis of Jurkat cells using anti-CD44 antibody. Overlay histogram showing Jurkat 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-CD44 Antibody (1  $\mu g/1x10^6$  cells) for 30 min at 20°C. DyLight® 488 conjugated goat anti-rabbit IgG (5-10  $\mu g/1x10^6$  cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was rabbit IgG (1  $\mu g/1x10^6$ ) 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 CD44 using anti-CD44 antibody CD44 was detected in paraffin-embedded section of mouse lymphaden tissues. Heat mediated antigen retrieval was performed in citrate buffer (pH6, epitope retrieval solution) for 20 mins. The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1 µg/mL rabbit anti-CD44 Antibody overnight at 4°C. DyLight® 488 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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## **Biorbyt Ltd.**

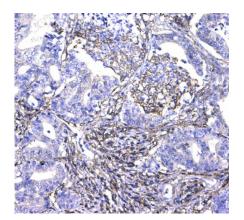
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## **Biorbyt LLC.**

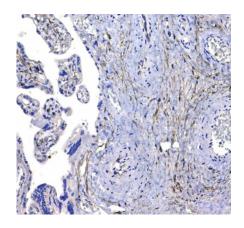
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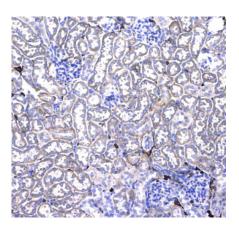




IHC analysis of CD44 using anti-CD44 antibody. CD44 was detected in paraffin-embedded section of human colon cancer tissue. Heat mediated antigen retrieval was performed in citrate buffer (pH6, epitope retrieval solution) for 20 mins. The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1  $\mu$ g/ml rabbit anti-CD44 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 CD44 using anti-CD44 antibody. CD44 was detected in paraffin-embedded section of human placenta tissue. Heat mediated antigen retrieval was performed in citrate buffer (pH6, epitope retrieval solution) for 20 mins. The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1  $\mu$ g/ml rabbit anti-CD44 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.

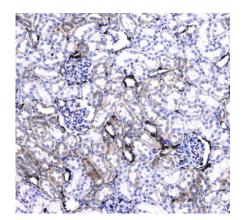


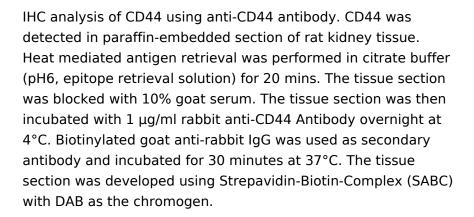
IHC analysis of CD44 using anti-CD44 antibody. CD44 was detected in paraffin-embedded section of mouse kidney tissue. Heat mediated antigen retrieval was performed in citrate buffer (pH6, epitope retrieval solution) for 20 mins. The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1  $\mu$ g/ml rabbit anti-CD44 Antibody overnight at 4°C. Biotinylated goat anti-rabbit lgG 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.

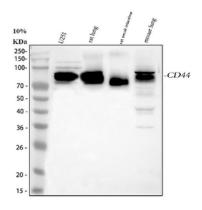
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Western blot analysis of CD44 using anti-CD44 antibody. Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 30 ug of sample under reducing conditions. Lane 1: human U251 whole cell lysates, Lane 2: rat lung tissue lysates, Lane 3: rat small intestine tissue lysates, Lane 4: mouse lung tissue tissue 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-CD44 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 CD44 at approximately 82 kDa. The expected band size for CD44 is at 82 kDa.