

Product Datasheet

RNF25 Rabbit Polyclonal Antibody (orb1939817)

Catalog Number	orb1939817
Category	Antibodies
Description	Anti-RNF25 Antibody. Tested in WB, IHC, IF, Flow Cytometry, ELISA applications. This antibody reacts with Human, Mouse, Rat.
Target	E3 ubiquitin-protein ligase RNF25
Clonality	Polyclonal
Species/Host	Rabbit
Isotype	IgG
Conjugation	Unconjugated
Reactivity	Human, Mouse, Rat
Form/Appearance	Lyophilized
Concentration	Adding 0.2 ml of distilled water will yield a concentration of 500 µg/ml.
Buffer/Preservatives	Each vial contains 4 mg Trehalose, 0.9 mg NaCl, 0.2 mg Na ₂ HPO ₄ .
Reconstitution	Adding 0.2 ml of distilled water will yield a concentration of 500 µg/ml.
Purification	Immunogen affinity purified.
Immunogen	E.coli-derived human RNF25 recombinant protein (Position: E44-R344). Human RNF25 shares 83.7% amino acid (aa) sequence identity with mouse RNF25.
UniProt ID	Q96BH1
MW	62 kDa

Biorbyt Ltd.

7 Signet Court, Swann Road
Cambridge
CB5 8LA
United Kingdom

Email: info@biorbyt.com, support@biorbyt.com

Phone: [+44 \(0\)1223 859353](tel:+44(0)1223859353) | Fax: [+1 \(415\) 651-8558](tel:+1(415)651-8558)

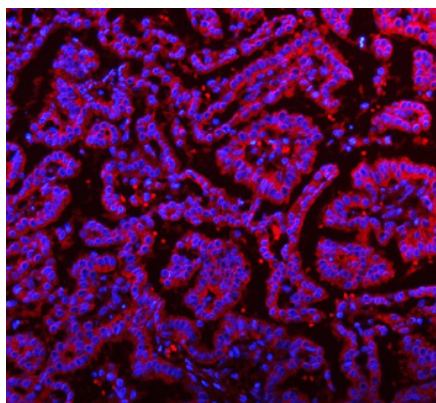
Biorbyt LLC

68 TW Alexander Drive
Research Triangle Park
Durham
NC 27713
United States

Email: info@biorbyt.com, support@biorbyt.com

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Tested applications	ELISA, FC, IF, IHC, WB
Dilution range	Western blot, 0.25-0.5 µg/ml, Human, Mouse, Rat Immunohistochemistry (Paraffin-embedded Section), 2-5 µg/ml, Human, Rat Immunofluorescence, 5 µg/ml, Human Flow Cytometry (Fixed), 1-3 µg/1x10 ⁶ cells, Human ELISA, 0.1-0.5 µg/ml, -
Cross Reactivity	No cross reactivity with other proteins.
Antibody Type	Primary Antibody
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
Expiration Date	12 months from date of receipt.



IF analysis of RNF25 using anti-RNF25 antibody. RNF25 was detected in a paraffin-embedded section of human lung 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 5 µg/mL rabbit anti-RNF25 Antibody overnight at 4°C. DyLight®594 Conjugated Goat Anti-Rabbit IgG was used as secondary antibody at 1:500 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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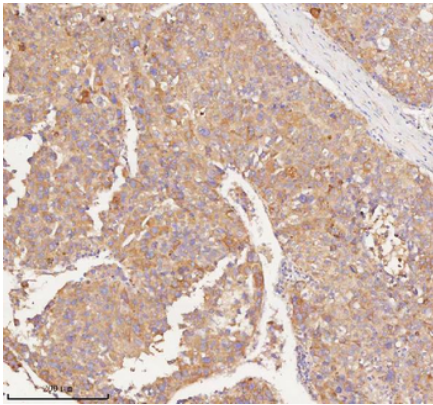
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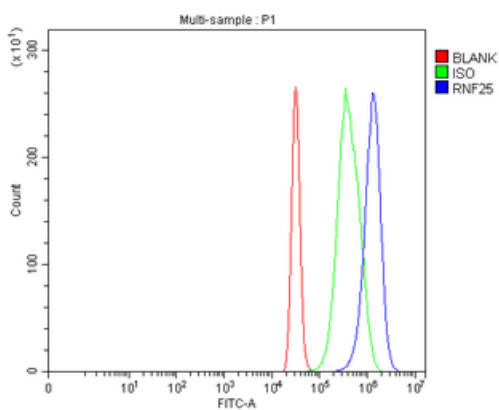
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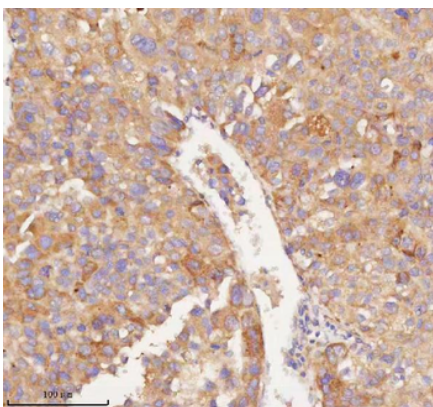
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IHC analysis of RNF25 using anti-RNF25 antibody. RNF25 was detected in a paraffin-embedded section of human liver cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 µg/ml rabbit anti-RNF25 Antibody overnight at 4°C. Peroxidase Conjugated Goat Anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Rabbit IgG Super Vision Assay Kit with DAB as the chromogen.



Flow Cytometry analysis of 293T cells using anti-RNF25 antibody. Overlay histogram showing 293T 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-RNF25 Antibody (1 µg/1x10⁶ cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-rabbit IgG (5-10 µg/1x10⁶ cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was rabbit IgG (1 µg/1x10⁶) used under the same conditions. Unlabelled sample (Red line) was also used as a control.



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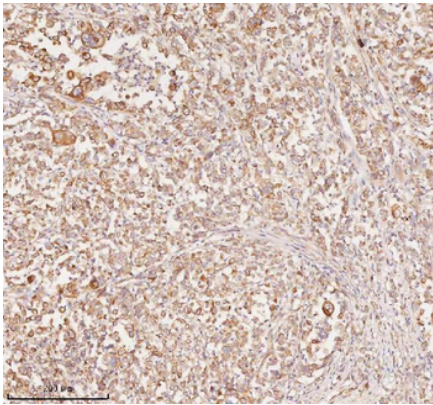
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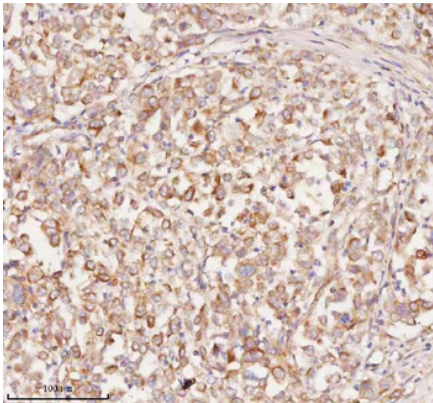
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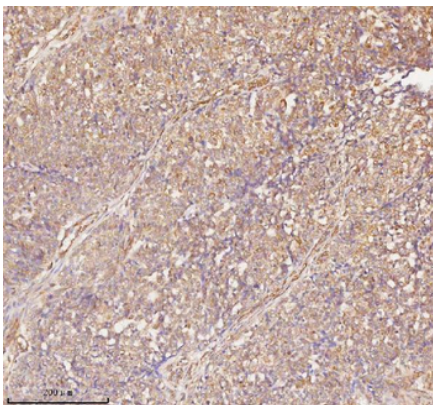
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IHC analysis of RNF25 using anti-RNF25 antibody. RNF25 was detected in a paraffin-embedded section of human lung adenocarcinoma tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 μg/ml rabbit anti-RNF25 Antibody overnight at 4°C. Peroxidase Conjugated Goat Anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Rabbit IgG Super Vision Assay Kit with DAB as the chromogen.



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IHC analysis of RNF25 using anti-RNF25 antibody. RNF25 was detected in a paraffin-embedded section of human ovarian 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 2 μg/ml rabbit anti-RNF25 Antibody overnight at 4°C. Peroxidase Conjugated Goat Anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Rabbit IgG Super Vision Assay Kit with DAB as the chromogen.

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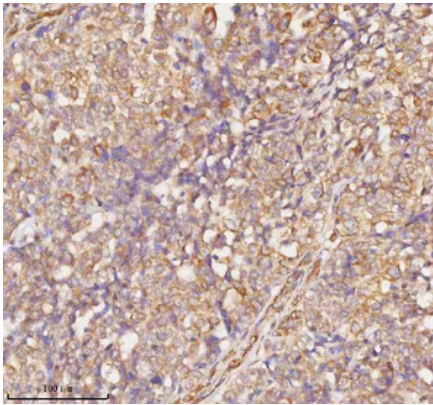
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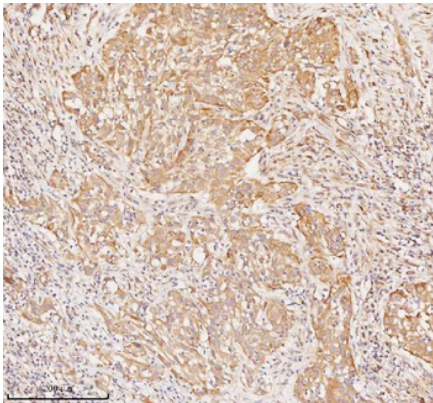
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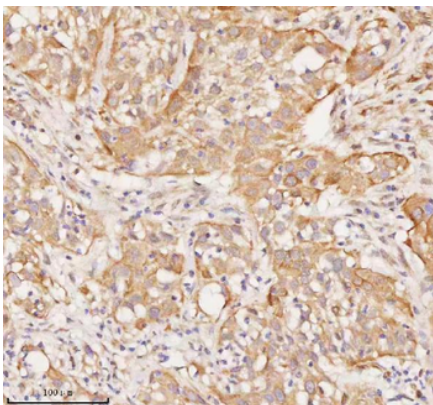
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IHC analysis of RNF25 using anti-RNF25 antibody. RNF25 was detected in a paraffin-embedded section of human urothelial carcinoma tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 μg/ml rabbit anti-RNF25 Antibody overnight at 4°C. Peroxidase Conjugated Goat Anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Rabbit IgG Super Vision Assay Kit with DAB as the chromogen.



IHC analysis of RNF25 using anti-RNF25 antibody. RNF25 was detected in a paraffin-embedded section of human urothelial 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 2 μg/ml rabbit anti-RNF25 Antibody overnight at 4°C. Peroxidase Conjugated Goat Anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Rabbit IgG Super Vision Assay Kit with DAB as the chromogen.

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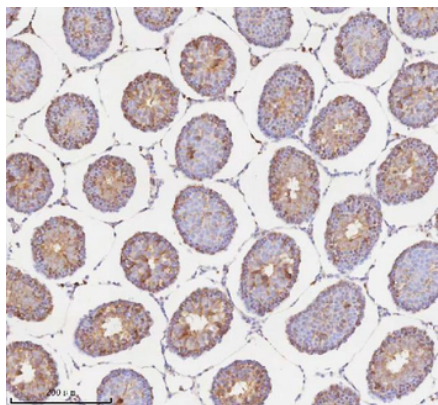
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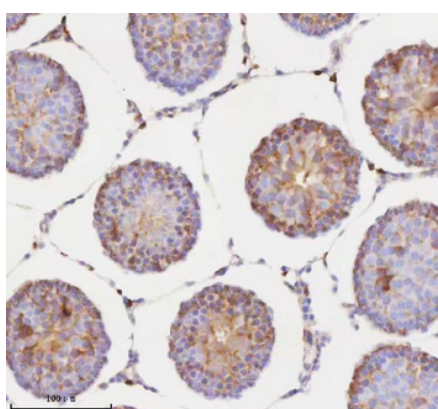
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IHC analysis of RNF25 using anti-RNF25 antibody. RNF25 was detected in a paraffin-embedded section of rat testis tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 $\mu\text{g}/\text{ml}$ rabbit anti-RNF25 Antibody overnight at 4°C. Peroxidase Conjugated Goat Anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Rabbit IgG Super Vision Assay Kit with DAB as the chromogen.



IHC analysis of RNF25 using anti-RNF25 antibody. RNF25 was detected in a 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 2 $\mu\text{g}/\text{ml}$ rabbit anti-RNF25 Antibody overnight at 4°C. Peroxidase Conjugated Goat Anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Rabbit IgG Super Vision Assay Kit with DAB as the chromogen.

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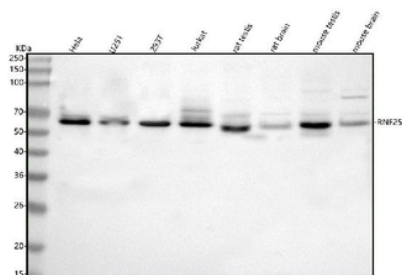
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Western blot analysis of RNF25 using anti-RNF25 antibody. Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 30 ug of sample under reducing conditions. Lane 1: human Hela whole cell lysates, Lane 2: human U251 whole cell lysates, Lane 3: human 293T whole cell lysates, Lane 4: human Jurkat whole cell lysates, Lane 5: rat testis tissue lysates, Lane 6: rat brain tissue lysates, Lane 7: mouse testis tissue lysates, Lane 8: mouse brain 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-RNF25 antigen affinity purified polyclonal antibody at 0.5 μ g/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 RNF25 at approximately 62 kDa. The expected band size for RNF25 is at 51 kDa.

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