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Anti-U2AF65/U2AF2 Picoband® Antibody (monoclonal, 3G9)

Catalog Number: M03639-1

About U2AF2

Splicing factor U2AF 65 kDa subunit is a protein that in humans is encoded by the U2AF2 gene. It is mapped to 19q13.42. U2 auxiliary factor (U2AF), comprised of a large and a small subunit, is a non-snRNP protein required for the binding of U2 snRNP to the pre-mRNA branch site. This gene encodes the U2AF large subunit which contains a sequence-specific RNA-binding region with 3 RNA recognition motifs and an Arg/Ser-rich domain necessary for splicing. The large subunit binds to the polypyrimidine tract of introns early during spliceosome assembly. Multiple transcript variants have been detected for this gene, but the full-length natures of only two have been determined to date.

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Overview

Product Name	Anti-U2AF65/U2AF2 Picoband® Antibody (monoclonal, 3G9)
Reactive Species	Human, Mouse, Rat
Description	Boster Bio Anti-U2AF65/U2AF2 Picoband® Antibody (monoclonal, 3G9) catalog # M03639-1. Tested in Flow Cytometry, IF, IHC, ICC, WB applications. This antibody reacts with Human, Mouse, Rat. The brand Picoband indicates this is a premium antibody that guarantees superior quality, high affinity, and strong signals with minimal background in Western blot applications. Only our best-performing antibodies are designated as Picoband, ensuring unmatched performance.
Application	Flow Cytometry, IF, IHC, ICC, WB
Clonality	Monoclonal 3G9
Formulation	Each vial contains 4mg Trehalose, 0.9mg NaCl and 0.2mg Na2HPO4.
Storage Instructions	Store at -20˚C for one year from date of receipt. After reconstitution, at 4˚C for one month. It can also be aliquotted and stored frozen at -20˚C for six months. Avoid repeated freeze-thaw cycles.
Host	Mouse
Uniprot ID	P26368

Technical Details

Immunogen	E.coli-derived human U2AF65/U2AF2 recombinant protein (Position: M238-H470).
Recommended Detection Systems	Boster recommends Enhanced Chemiluminescent Kit with anti-Mouse IgG (EK1001) for Western

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	blot, and HRP Conjugated anti-Mouse IgG Super Vision Assay Kit (SV0001-1) for IHC(P) and ICC.
Cross Reactivity	No cross-reactivity with other proteins.
Isotype	Mouse IgG2a
Form	Lyophilized
Concentration	Adding 0.2 ml of distilled water will yield a concentration of 500 ug/ml.
Purification	Immunogen affinity purified.
Suggested Dilutions	Western blot, 0.1-0.25ug/ml, Human, Mouse, Rat Immunohistochemistry (Paraffin-embedded Section), 2-5ug/ml, Human, Mouse, Rat Immunocytochemistry/Immunofluorescence, 5ug/ml, Human Flow Cytometry (Fixed), 1-3ug/1x10^6 cells, Human Note: For flow cytometry (FCM), suitable for fixed cells only. Not suitable for FACS or surface flow cytometry using live cells.

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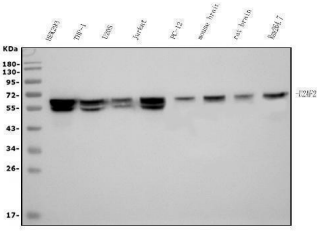
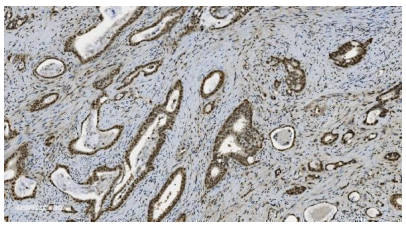
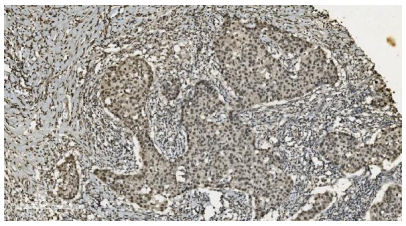
Anti-U2AF65/U2AF2 Picoband® Antibody (monoclonal, 3G9) (M03639-1) Images



Western blot analysis of U2AF65/U2AF2 using anti-U2AF65/U2AF2 antibody (M03639-1). 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 50ug of sample under reducing conditions. Lane 1: human HEK293 whole cell lysates, Lane 2: human THP-1 whole cell lysates, Lane 3: human U20S whole cell lysates, Lane 4: human Jurkat whole cell lysates, Lane 5: rat PC-12 whole cell lysates, Lane 6: mouse brain tissue lysates, Lane 7: rat brain tissue lysates, Lane 8: mouse RAW264.7 whole cell lysates. After Electrophoresis, proteins were transferred to a Nitrocellulose membrane at 150mA for 50-90 minutes. Blocked the membrane with 5% Non-fat Milk/ TBS for 1.5 hour at RT. The membrane was incubated with mouse anti-U2AF65/U2AF2 antigen affinity purified monoclonal antibody (Catalog # M03639-1) at 0.25 ug/mL overnight at 4°C, then washed with TBS-0.1%Tween 3 times with 5 minutes each and probed with a goat anti-mouse IgG-HRP secondary antibody at a dilution of 1:10000 for 1.5 hour at RT. The signal is developed using an Enhanced Chemiluminescent detection (ECL) kit (Catalog # EK1001) with Tanon 5200 system. A specific band was detected for U2AF65/U2AF2 at approximately 65KD. The expected band size for U2AF65/U2AF2 is at 65KD.



IHC analysis of U2AF65/U2AF2 using anti U2AF65/U2AF2 antibody (M03639-1). U2AF65/U2AF2 was detected in paraffin-embedded section of human gallbladder 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 2ug/ml mouse anti-U2AF65/U2AF2 Antibody (M03639-1) overnight at 4°C. Biotinylated goat anti-mouse IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Strepavidin-Biotin-Complex (SABC) (Catalog # SA1021) with DAB as the chromogen.



IHC analysis of U2AF65/U2AF2 using anti U2AF65/U2AF2 antibody (M03639-1). U2AF65/U2AF2 was detected in paraffin-embedded section of human mammary 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 2ug/ml mouse anti-U2AF65/U2AF2 Antibody (M03639-1) overnight at 4°C. Biotinylated goat anti-mouse IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Strepavidin-Biotin-Complex (SABC) (Catalog # SA1021) with DAB as the chromogen.

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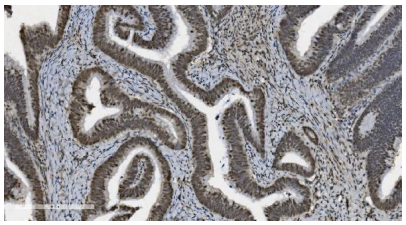
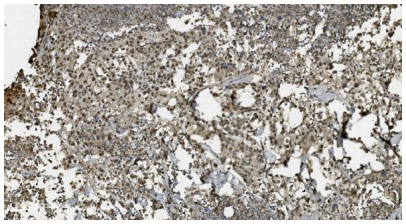
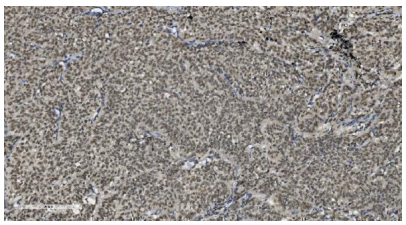


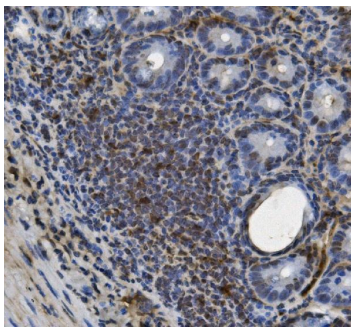
IHC analysis of U2AF65/U2AF2 using anti U2AF65/U2AF2 antibody (M03639-1). U2AF65/U2AF2 was detected in paraffin-embedded section of human colonic adenocarcinoma tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2ug/ml mouse anti-U2AF65/U2AF2 Antibody (M03639-1) overnight at 4°C. Biotinylated goat anti-mouse IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Strepavidin-Biotin-Complex (SABC) (Catalog # SA1021) with DAB as the chromogen.



IHC analysis of U2AF65/U2AF2 using anti U2AF65/U2AF2 antibody (M03639-1). U2AF65/U2AF2 was detected in paraffin-embedded section of human pancreatic 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 2ug/ml mouse anti-U2AF65/U2AF2 Antibody (M03639-1) overnight at 4°C. Biotinylated goat anti-mouse IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Strepavidin-Biotin-Complex (SABC) (Catalog # SA1021) with DAB as the chromogen.



IHC analysis of U2AF65/U2AF2 using anti U2AF65/U2AF2 antibody (M03639-1). U2AF65/U2AF2 was detected in 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 2ug/ml mouse anti-U2AF65/U2AF2 Antibody (M03639-1) overnight at 4°C. Biotinylated goat anti-mouse IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Strepavidin-Biotin-Complex (SABC) (Catalog # SA1021) with DAB as the chromogen.



IHC analysis of U2AF65/U2AF2 using anti U2AF65/U2AF2 antibody (M03639-1). U2AF65/U2AF2 was detected in paraffin-embedded section of mouse intestine 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 2ug/ml mouse anti-U2AF65/U2AF2 Antibody (M03639-1) overnight at 4°C. Biotinylated goat anti-mouse IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Strepavidin-Biotin-Complex (SABC) (Catalog # SA1021) with DAB as the chromogen.

IHC analysis of U2AF65/U2AF2 using anti U2AF65/U2AF2 antibody (M03639-1). U2AF65/U2AF2 was detected in paraffin-embedded section of rat intestine tissue. Heat

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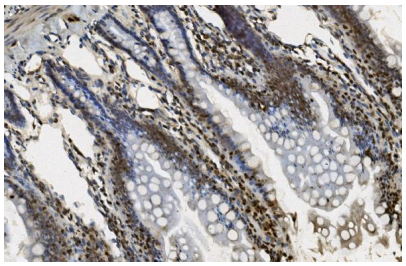
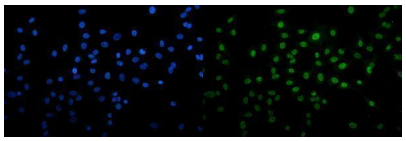
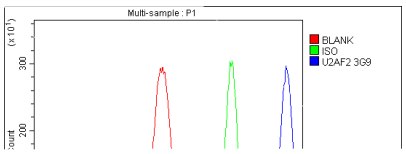


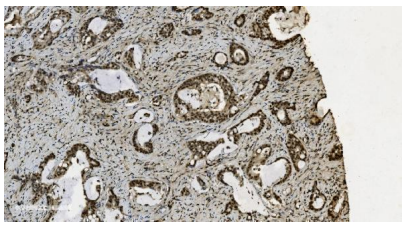
mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2ug/ml mouse anti-U2AF65/U2AF2 Antibody (M03639-1) overnight at 4°C. Biotinylated goat anti-mouse IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Strepavidin-Biotin-Complex (SABC) (Catalog # SA1021) with DAB as the chromogen.



IF analysis of U2AF65/U2AF2 using anti-U2AF65/U2AF2 antibody (M03639-1). U2AF65/U2AF2 was detected in immunocytochemical section of A549 cells. Enzyme antigen retrieval was performed using IHC enzyme antigen retrieval reagent (AR0022) for 15 mins. The cells were blocked with 10% goat serum. And then incubated with 5ug/mL mouse anti-U2AF65/U2AF2 Antibody (M03639-1) overnight at 4°C. DyLight®488 Conjugated Goat Anti-Mouse IgG (BA1126) 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.



Flow Cytometry analysis of A549 cells using anti-U2AF65/U2AF2 antibody (M03639-1). Overlay histogram showing A549 cells stained with M03639-1 (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 mouse anti-U2AF65/U2AF2 Antibody (M03639-1, 1ug/1x106 cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-mouse IgG (BA1126, 5-10ug/1x106 cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was mouse IgG (1ug/1x106) used under the same conditions. Unlabelled sample without incubation with primary antibody and secondary antibody (Red line) was used as a blank control.



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