BOSTER BIOLOGICAL TECHNOLOGY
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Anti-U2AF65/U2AF2 Antibody Picoband®

Catalog Number: A03639-2

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 Antibody Picoband®
Reactive Species	Human, Mouse, Rat
Description	Boster Bio Anti-U2AF65/U2AF2 Antibody Picoband® catalog # A03639-2. Tested in ELISA, Flow Cytometry, IP, 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	ELISA, Flow Cytometry, IP, IF, IHC, ICC, WB
Clonality	Polyclonal
Formulation	Each vial contains 4mg Trehalose, 0.9mg NaCl, 0.2mg Na2HPO4, 0.05mg NaN3.
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	Rabbit
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-Rabbit IgG (EK1002) for Western

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	blot, and HRP Conjugated anti-Rabbit IgG Super Vision Assay Kit (SV0002-1) for IHC(P) and ICC.
Cross Reactivity	No cross-reactivity with other proteins.
Isotype	Rabbit IgG
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, 0.25-0.5ug/ml, Human, Mouse, Rat Immunohistochemistry (Paraffin-embedded Section), 0.5-1ug/ml, Human, Mouse, Rat Immunocytochemistry/Immunofluorescence, 2ug/ml, Human Immunoprecipitation, 0.5-2 ug/ml, Human Flow Cytometry (Fixed), 1-3ug/1x10^6 cells, Human, Mouse, Rat ELISA, 0.1-0.5ug/ml, - 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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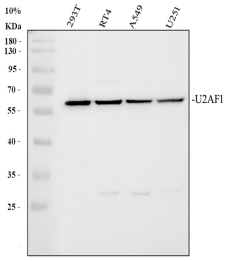
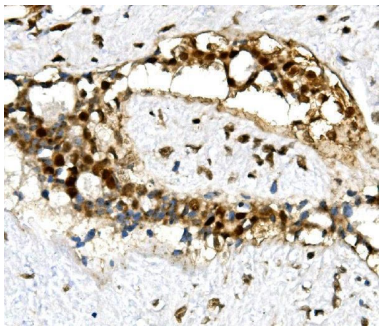
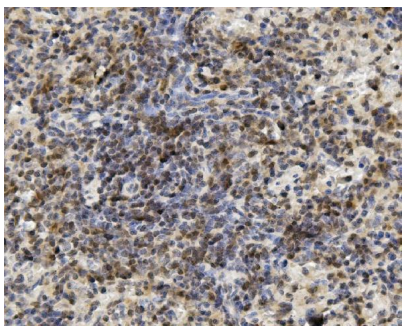
Anti-U2AF65/U2AF2 Antibody Picoband® (A03639-2) Images



Western blot analysis of U2AF2 using anti-U2AF2 antibody (A03639-2). Electrophoresis was performed on a 10% SDS-PAGE gel at 80V (Stacking gel) / 120V (Resolving gel) for 2 hours. The sample well of each lane was loaded with 30 ug of sample under reducing conditions. Lane 1: human 293T whole cell lysates, Lane 2: human RT4 whole cell lysates, Lane 3: human A549 whole cell lysates, Lane 4: human U251 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-U2AF2 antigen affinity purified polyclonal antibody (A03639-2) at 0.5 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-rabbit IgG-HRP secondary antibody at a dilution of 1:5000 for 1.5 hour at RT. The signal is developed using an ECL Plus Western Blotting Substrate (Catalog # AR1196-200) with Tanon 5200 system. A specific band was detected for U2AF2 at approximately 65 kDa. The expected band size for U2AF2 is at 65 kDa.



IHC analysis of U2AF2 using anti-U2AF2 antibody (A03639-2). 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 1ug/ml rabbit anti-U2AF2 Antibody (A03639-2) 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) (Catalog # SA1022) with DAB as the chromogen.



IHC analysis of U2AF2 using anti-U2AF2 antibody (A03639-2). U2AF2 was detected in paraffin-embedded section of mouse spleen 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 1ug/ml rabbit anti-U2AF2 Antibody (A03639-2) 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) (Catalog # SA1022) with DAB as the chromogen.

IHC analysis of U2AF2 using anti-U2AF2 antibody (A03639-2). U2AF2 was detected in paraffin-embedded section of rat spleen tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat

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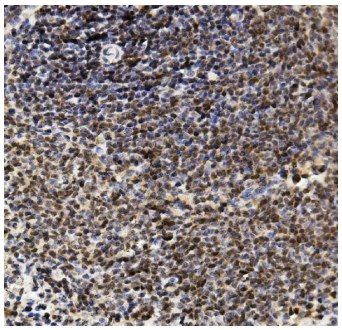
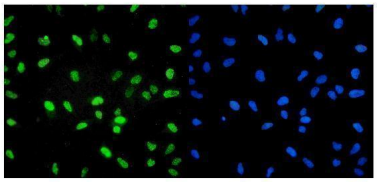
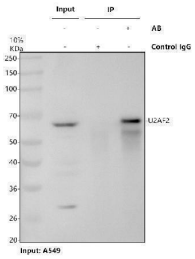


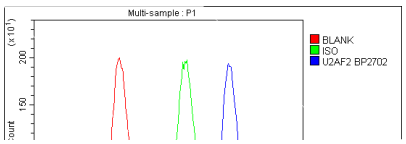
serum. The tissue section was then incubated with 1ug/ml rabbit anti-U2AF2 Antibody (A03639-2) overnight at 4°C. Biotinylated goat anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Strepavidin-Biotin-Complex (SABC) (Catalog # SA1022) with DAB as the chromogen.



IF analysis of U2AF2 using anti-U2AF2 antibody (A03639-2). U2AF2 was detected in immunocytochemical section of Hela 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 2ug/mL rabbit anti-U2AF2 Antibody (A03639-2) overnight at 4°C. DyLight®488 Conjugated Goat Anti-Rabbit IgG (BA1127) 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.



Immunoprecipitating U2AF2 in A549 whole cell lysate. Western blot analysis of U2AF2 using anti-U2AF2 antibody (A03639-2). Lane 1: A549 whole cell lysates (30ug), Lane 2: Rabbit control IgG instead of anti-U2AF2 antibody in A549 whole cell lysate, Lane 3: anti-U2AF2 antibody (2ug) + A549 whole cell lysate (500ug). After electrophoresis, proteins were transferred to a membrane. Then the membrane was incubated with rabbit anti-U2AF2 antigen affinity purified polyclonal antibody (A03639-2) at a dilution of 1:50 and probed with a mouse anti-rabbit IgG-HRP secondary antibody (Light Chain). The signal is developed using ECL Plus Western Blotting Substrate (Catalog # AR1197). A specific band was detected for U2AF2 at approximately 65 kDa. The expected band size for U2AF2 is at 53 kDa.



Flow Cytometry analysis of PC-3 cells using anti-U2AF2 antibody (A03639-2). Overlay histogram showing PC-3 cells stained with A03639-2 (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-U2AF2 Antibody (A03639-2, 1ug/1x106 cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-rabbit IgG (BA1127, 5-10ug/1x106 cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was rabbit 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.

Flow Cytometry analysis of ANA-1 cells using anti-U2AF2

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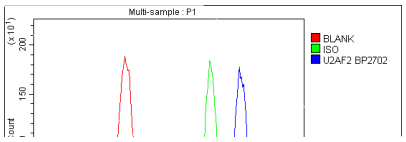
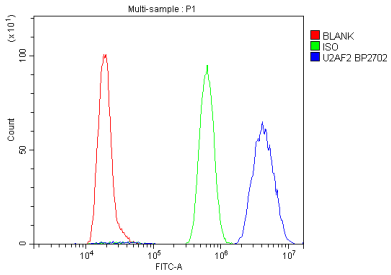


antibody (A03639-2). Overlay histogram showing ANA-1 cells stained with A03639-2 (Blue line). To facilitate intracellular staining, cells were fixed with 4% paraformaldehyde and permeabilized with permeabilization buffer. The cells were blocked with 10% normal goat serum. And then incubated with rabbit anti-U2AF2 Antibody (A03639-2, 1ug/1x106 cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-rabbit IgG (BA1127, 5-10ug/1x106 cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was rabbit 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.



Flow Cytometry analysis of NRK cells using anti-U2AF2 antibody (A03639-2). Overlay histogram showing NRK cells stained with A03639-2 (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-U2AF2 Antibody (A03639-2, 1ug/1x106 cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-rabbit IgG (BA1127, 5-10ug/1x106 cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was rabbit 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.

Anti-U2AF65/U2AF2 Antibody

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