

Anti-KPNB1 Antibody Picoband™ (monoclonal, 3l11F2)

Catalog # ABO16576

Anti-KPNB1 Antibody Picoband[™] (monoclonal, 3I11F2) - Product Information

Application Primary Accession Host Isotype Reactivity Clonality Format Description WB, IHC, IF, ICC, FC <u>014974</u> Mouse Mouse IgG2b Rat, Human, Mouse Monoclonal Lyophilized

Anti-KPNB1 Antibody Picoband[™] (monoclonal, 3I11F2). Tested in Flow Cytometry, IF, IHC, ICC, WB applications. This antibody reacts with Human, Mouse, Rat.

Reconstitution Adding 0.2 ml of distilled water will yield a concentration of 500 μ g/ml.

Anti-KPNB1 Antibody Picoband[™] (monoclonal, 3I11F2) - Additional Information

Gene ID 3837

Other Names Importin subunit beta-1, Importin-90, Karyopherin subunit beta-1, Nuclear factor p97, Pore targeting complex 97 kDa subunit, PTAC97, KPNB1, NTF97

Calculated MW 97 kDa KDa

Application Details Western blot, 0.25-0.5 μ g/ml, Human, Mouse
 Immunohistochemistry(Paraffin-embedded Section), 2-5 μ g/ml, Human, Mouse, Rat
 Immunocytochemistry/Immunofluorescence, 5 μ g/ml, Human
 Flow Cytometry, 1-3 μ g/1x10^6 cells, Human

Contents Each vial contains 4 mg Trehalose, 0.9 mg NaCl and 0.2 mg Na2HPO4.

Immunogen E.coli-derived human KPNB1 recombinant protein (Position: E8-A876).

Purification Immunogen affinity purified.

Storage

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 freezing and thawing.



Anti-KPNB1 Antibody Picoband[™] (monoclonal, 3I11F2) - Protein Information

Name KPNB1

Synonyms NTF97

Function

Functions in nuclear protein import, either in association with an adapter protein, like an importin-alpha subunit, which binds to nuclear localization signals (NLS) in cargo substrates, or by acting as autonomous nuclear transport receptor (PubMed:10228156, PubMed:11682607, PubMed:11891849, PubMed:19386897, PubMed:20818336, PubMed:24699649, PubMed:7615630, PubMed:9687515). Acting autonomously, serves itself as NLS receptor (PubMed:10228156, PubMed:11682607, PubMed:11891849, PubMed:19386897, PubMed:20818336, PubMed:24699649, PubMed:7615630, PubMed:9687515). Docking of the importin/substrate complex to the nuclear pore complex (NPC) is mediated by KPNB1 through binding to nucleoporin FxFG repeats and the complex is subsequently translocated through the pore by an energy requiring, Ran-dependent mechanism (PubMed: 10228156, PubMed:11682607, PubMed:11891849, PubMed:19386897, PubMed:20818336, PubMed:24699649, PubMed:7615630, PubMed:9687515). At the nucleoplasmic side of the NPC. Ran binds to importin-beta and the three components separate and importin-alpha and -beta are re-exported from the nucleus to the cytoplasm where GTP hydrolysis releases Ran from importin (PubMed:10228156, PubMed:11682607, PubMed:11891849, PubMed:19386897, PubMed:20818336, PubMed:24699649, PubMed:7615630, PubMed:9687515). The directionality of nuclear import is thought to be conferred by an asymmetric distribution of the GTP- and GDP-bound forms of Ran between the cytoplasm and nucleus (PubMed: 10228156, PubMed:11682607, PubMed:11891849, PubMed:19386897, PubMed:<a href="http://www.uniprot.org/citations/24699649"



target="_blank">24699649, PubMed:7615630, PubMed:9687515). Mediates autonomously the nuclear import of ribosomal proteins RPL23A, RPS7 and RPL5 (PubMed:11682607, PubMed:9687515). In essociation with IPO7, mediates the nuclear import of H1 histone (PubMed:10228156). In vitro, mediates nuclear import of H2A, H2B, H3 and H4 histones (By similarity). Imports MRTFA, SNAI1 and PRKCI into the nucleus (PubMed:11891849, PubMed:10386897, PubMed:10386897, PubMed:10386897, PubMed:10386897, PubMed:20818336, PubMed:20818336, PubMed:24699649).

Cellular Location Cytoplasm. Nucleus envelope

Anti-KPNB1 Antibody Picoband™ (monoclonal, 3I11F2) - Protocols

Provided below are standard protocols that you may find useful for product applications.

- <u>Western Blot</u>
- <u>Blocking Peptides</u>
- Dot Blot
- Immunohistochemistry
- Immunofluorescence
- Immunoprecipitation
- Flow Cytomety
- <u>Cell Culture</u>

Anti-KPNB1 Antibody Picoband™ (monoclonal, 3I11F2) - Images



Figure 1. Western blot analysis of KPNB1 using anti-KPNB1 antibody (M01851-2).

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 CACO-2 whole cell lysates,

Lane 2: human K562 whole cell lysates,



Lane 3: human A549 whole cell lysates,

Lane 4: human A549 whole cell lysates,

Lane 5: mouse testis tissue lysates,

Lane 6: mouse lung 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 mouse anti-KPNB1 antigen affinity purified monoclonal antibody (Catalog # M01851-2) 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-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 KPNB1 at approximately 97 kDa.



Figure 2. IHC analysis of KPNB1 using anti-KPNB1 antibody (M01851-2).

KPNB1 was detected in a paraffin-embedded section of human thyroiditis tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 μ g/ml mouse anti-KPNB1 Antibody (M01851-2) overnight at 4°C. Peroxidase Conjugated Goat Anti-mouse IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Mouse IgG Super Vision Assay Kit (Catalog # SV0001) with DAB as the chromogen.



Figure 3. IHC analysis of KPNB1 using anti-KPNB1 antibody (M01851-2).

KPNB1 was detected in a paraffin-embedded section of human squamous cell lung carcinoma tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 μ g/ml mouse anti-KPNB1 Antibody (M01851-2) overnight at 4°C. Peroxidase Conjugated Goat Anti-mouse IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Mouse IgG Super Vision Assay Kit (Catalog # SV0001) with DAB as the chromogen.





Figure 4. IHC analysis of KPNB1 using anti-KPNB1 antibody (M01851-2).

KPNB1 was detected in a paraffin-embedded section of human renal carcinoma tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 µg/ml mouse anti-KPNB1 Antibody (M01851-2) overnight at 4°C. Peroxidase Conjugated Goat Anti-mouse IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Mouse IgG Super Vision Assay Kit (Catalog # SV0001) with DAB as the chromogen.



Figure 5. IHC analysis of KPNB1 using anti-KPNB1 antibody (M01851-2).

KPNB1 was detected in a paraffin-embedded section of human breast cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 µg/ml mouse anti-KPNB1 Antibody (M01851-2) overnight at 4°C. Peroxidase Conjugated Goat Anti-mouse IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Mouse IgG Super Vision Assay Kit (Catalog # SV0001) with DAB as the chromogen.





Figure 6. IHC analysis of KPNB1 using anti-KPNB1 antibody (M01851-2).

KPNB1 was detected in a paraffin-embedded section of human colorectal adenocarcinoma tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 μ g/ml mouse anti-KPNB1 Antibody (M01851-2) overnight at 4°C. Peroxidase Conjugated Goat Anti-mouse IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Mouse IgG Super Vision Assay Kit (Catalog # SV0001) with DAB as the chromogen.



Figure 7. IHC analysis of KPNB1 using anti-KPNB1 antibody (M01851-2).

KPNB1 was detected in a paraffin-embedded section of human laryngeal squamous cell carcinoma tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 μ g/ml mouse anti-KPNB1 Antibody (M01851-2) overnight at 4°C. Peroxidase Conjugated Goat Anti-mouse IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Mouse IgG Super Vision Assay Kit (Catalog # SV0001) with DAB as the chromogen.





Figure 8. IHC analysis of KPNB1 using anti-KPNB1 antibody (M01851-2).

KPNB1 was detected in a paraffin-embedded section of human ovary tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 µg/ml mouse anti-KPNB1 Antibody (M01851-2) overnight at 4°C. Peroxidase Conjugated Goat Anti-mouse IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Mouse IgG Super Vision Assay Kit (Catalog # SV0001) with DAB as the chromogen.



Figure 9. IHC analysis of KPNB1 using anti-KPNB1 antibody (M01851-2).

KPNB1 was detected in a paraffin-embedded section of human placenta tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 μ g/ml mouse anti-KPNB1 Antibody (M01851-2) overnight at 4°C. Peroxidase Conjugated Goat Anti-mouse IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Mouse IgG Super Vision Assay Kit (Catalog # SV0001) with DAB as the chromogen.





Figure 10. IHC analysis of KPNB1 using anti-KPNB1 antibody (M01851-2).

KPNB1 was detected in a paraffin-embedded section of human spleen tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 μ g/ml mouse anti-KPNB1 Antibody (M01851-2) overnight at 4°C. Peroxidase Conjugated Goat Anti-mouse IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Mouse IgG Super Vision Assay Kit (Catalog # SV0001) with DAB as the chromogen.



Figure 11. IHC analysis of KPNB1 using anti-KPNB1 antibody (M01851-2).

KPNB1 was detected in a paraffin-embedded section of mouse lymphaden tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 μ g/ml mouse anti-KPNB1 Antibody (M01851-2) overnight at 4°C. Peroxidase Conjugated Goat Anti-mouse IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Mouse IgG Super Vision Assay Kit (Catalog # SV0001) with DAB as the chromogen.





Figure 12. IHC analysis of KPNB1 using anti-KPNB1 antibody (M01851-2).

KPNB1 was detected in a paraffin-embedded section of rat colon tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 μ g/ml mouse anti-KPNB1 Antibody (M01851-2) overnight at 4°C. Peroxidase Conjugated Goat Anti-mouse IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Mouse IgG Super Vision Assay Kit (Catalog # SV0001) with DAB as the chromogen.



Figure 13. IF analysis of KPNB1 using anti-KPNB1 antibody (M01851-2).

KPNB1 was detected in an immunocytochemical section of CACO-2 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 5 μ g/mL mouse anti-KPNB1 Antibody (M01851-2) overnight at 4°C. DyLight®594 Conjugated Goat Anti-Mouse IgG (BA1141) 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.

Anti-KPNB1 Antibody Picoband™ (monoclonal, 3l11F2) - Background

Importin subunit beta-1 is a protein that in humans is encoded by the KPNB1 gene. Nucleocytoplasmic transport, a signal- and energy-dependent process, takes place through nuclear pore complexes embedded in the nuclear envelope. The import of proteins containing a nuclear localization signal (NLS) requires the NLS import receptor, a heterodimer of importin alpha and beta subunits also known as karyopherins. Importin alpha binds the NLS-containing cargo in the cytoplasm and importin beta docks the complex at the cytoplasmic side of the nuclear pore complex. In the presence of nucleoside triphosphates and the small GTP binding protein Ran, the complex moves into the nuclear pore complex and the importin subunits dissociate. Importin alpha enters the nucleoplasm with its passenger protein and importin beta remains at the pore. Interactions between importin beta and the FG repeats of nucleoporins are essential in translocation through the pore complex. The protein encoded by this gene is a member of the importin beta family. Two transcript variants encoding different isoforms have been found for this gene.