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Anti-eRF1/ETF1 Antibody Picoband™ (monoclonal, 3B6)

Catalog # ABO14986

Specification

Anti-eRF1/ETF1 Antibody Picoband™ (monoclonal, 3B6) - Product Information

Application	**WB, IHC, IF, ICC, FC**
Primary Accession	P62495
Host	**Mouse**
Isotype	**Mouse IgG2a**
Reactivity	**Rat, Human, Mouse**
Clonality	**Monoclonal**
Format	**Lyophilized**

Description
Anti-eRF1/ETF1 Antibody Picoband™ (monoclonal, 3B6) . Tested in Flow Cytometry, IF, IHC, ICC, WB applications. This antibody reacts with Human, Mouse, Rat.

Reconstitution
Add 0.2ml of distilled water will yield a concentration of 500ug/ml.

Anti-eRF1/ETF1 Antibody Picoband™ (monoclonal, 3B6) - Additional Information

Gene ID 2107

Other Names
Eukaryotic peptide chain release factor subunit 1, Eukaryotic release factor 1, eRF1, Protein Cl1, TB3-1, ETF1, ERF1, RF1, SUP45L1

Calculated MW
49 kDa KDa

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

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

Immunogen
E.coli-derived human eRF1/ETF1 recombinant protein (Position: D9-K342).

Purification
Immunogen affinity purified.

Storage	**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.**

Anti-eRF1/ETF1 Antibody Picoband™ (monoclonal, 3B6) - Protein Information

Name ETF1

Synonyms ERF1, RF1, SUP45L1

Function
Component of the eRF1-eRF3-GTP ternary complex, a ternary complex that mediates translation termination in response to the termination codons (PubMed:10676813, PubMed:16777602, PubMed:24486019, PubMed:26245381, PubMed:27863242, PubMed:36638793, PubMed:7990965). The eRF1-eRF3-GTP complex binds to a stop codon in the ribosomal A-site (PubMed:26245381, PubMed:27863242, PubMed:36638793). ETF1/ERF1 is responsible for stop codon recognition and inducing hydrolysis of peptidyl-tRNA (PubMed:26245381, PubMed:27863242, PubMed:36638793). Following GTP hydrolysis, eRF3 (GSPT1/ERF3A or GSPT2/ERF3B) dissociates, permitting ETF1/eRF1 to accommodate fully in the A-site and mediate hydrolysis of peptidyl-tRNA (PubMed:10676813, PubMed:16777602, PubMed:26245381, PubMed:27863242). Component of the transient SURF complex which recruits UPF1 to stalled ribosomes in the context of nonsense-mediated decay (NMD) of mRNAs containing premature stop codons (PubMed:19417104). Required for SHFL-mediated translation termination which inhibits programmed ribosomal frameshifting (-1PRF) of mRNA from viruses and cellular genes (PubMed:30682371).

Cellular Location
Cytoplasm.

Anti-eRF1/ETF1 Antibody Picoband™ (monoclonal, 3B6) - Protocols

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

- Western Blot
- Blocking Peptides
- Dot Blot
- Immunohistochemistry
- Immunofluorescence
- Immunoprecipitation
- Flow Cytomety
- Cell Culture

Anti-eRF1/ETF1 Antibody Picoband™ (monoclonal, 3B6) - Images

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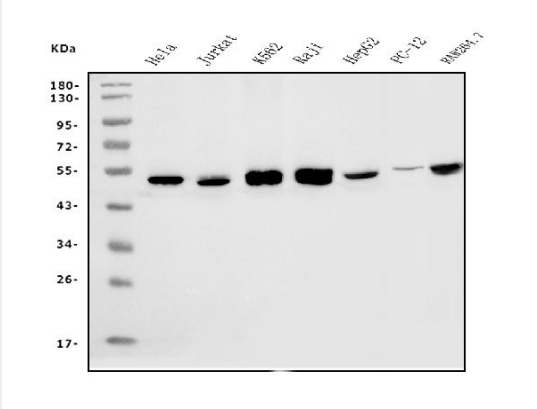

Figure 1. Western blot analysis of eRF1/ETF1 using anti-eRF1/ETF1 antibody (M04157).
Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 50ug of sample under reducing conditions.
Lane 1: human Hela whole cell lysates,
Lane 2: human Jurkat whole cell lysates,
Lane 3: human K562 whole cell lysates,
Lane 4: human Raji whole cell lysates,
Lane 5: human HEPG2 whole cell lysates,
Lane 6: rat PC-12 whole cell lysates,
Lane 7: 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-eRF1/ETF1 antigen affinity purified monoclonal antibody (Catalog # M04157) 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-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 eRF1/ETF1 at approximately 49KD. The expected band size for eRF1/ETF1 is at 49KD.

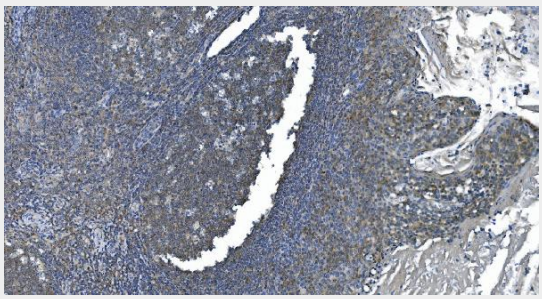

Figure 2. IHC analysis of eRF1/ETF1 using anti-eRF1/ETF1 antibody (M04157).
eRF1/ETF1 was detected in paraffin-embedded section of human tonsil tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 µg/ml mouse anti-eRF1/ETF1 Antibody (M04157) 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.

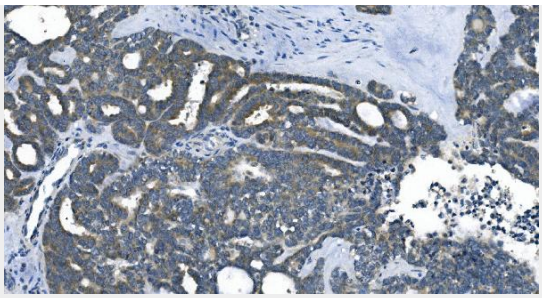

Figure 3. IHC analysis of eRF1/ETF1 using anti-eRF1/ETF1 antibody (M04157).
eRF1/ETF1 was detected in paraffin-embedded section of human ovarian cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 µg/ml mouse anti-eRF1/ETF1 Antibody (M04157) 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.

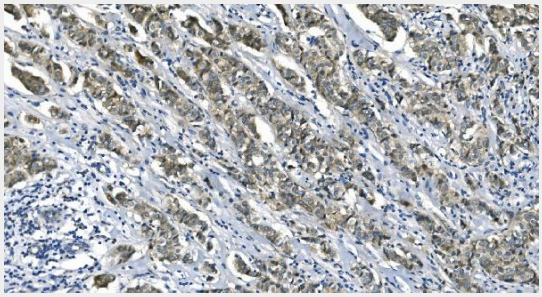

Figure 4. IHC analysis of eRF1/ETF1 using anti-eRF1/ETF1 antibody (M04157).
eRF1/ETF1 was detected in paraffin-embedded section of human breast cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 µg/ml mouse anti-eRF1/ETF1 Antibody (M04157) 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.

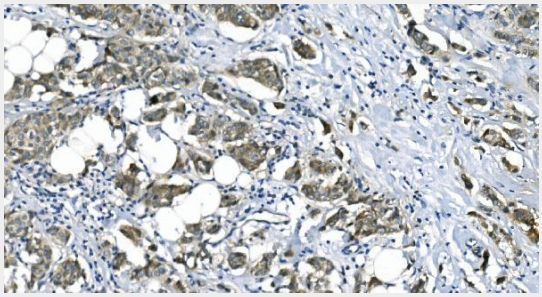

Figure 5. IHC analysis of eRF1/ETF1 using anti-eRF1/ETF1 antibody (M04157).
eRF1/ETF1 was detected in paraffin-embedded section of human breast cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 µg/ml mouse anti-eRF1/ETF1 Antibody (M04157) 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.

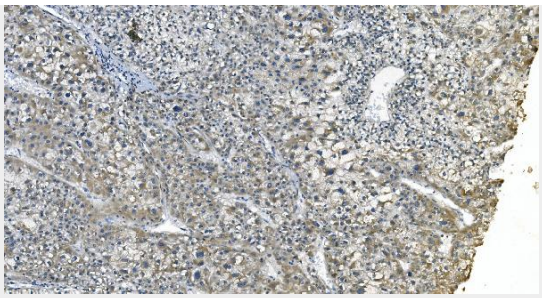

Figure 6. IHC analysis of eRF1/ETF1 using anti-eRF1/ETF1 antibody (M04157).
eRF1/ETF1 was detected in paraffin-embedded section of human liver cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 µg/ml mouse anti-eRF1/ETF1 Antibody (M04157) 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.

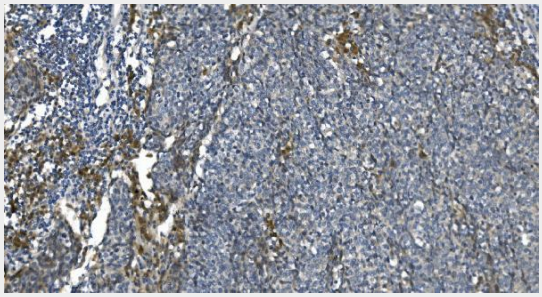

Figure 7. IHC analysis of eRF1/ETF1 using anti-eRF1/ETF1 antibody (M04157).
eRF1/ETF1 was detected in paraffin-embedded section of human lung cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 µg/ml mouse anti-eRF1/ETF1 Antibody (M04157) 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.

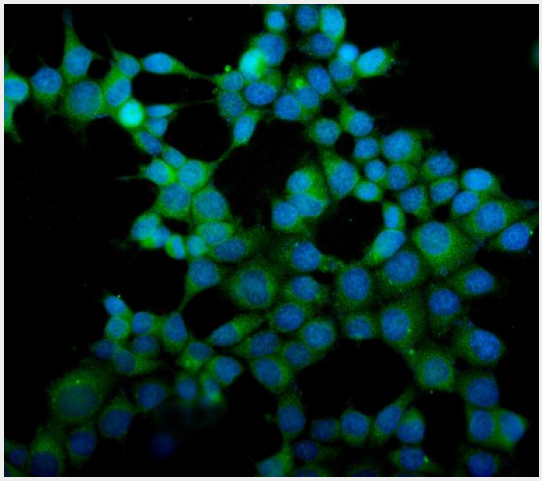

Figure 8. IF analysis of eRF1/ETF1 using anti-eRF1/ETF1 antibody (M04157).
eRF1/ETF1 was detected in immunocytochemical section of MCF-7 cells. Enzyme antigen retrieval was performed using IHC enzyme antigen retrieval reagent (AR0022) for 15 mins. The cells were blocked with 10% goat serum. And then incubated with 5 µg/mL mouse anti-eRF1/ETF1 Antibody

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(M04157) 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.

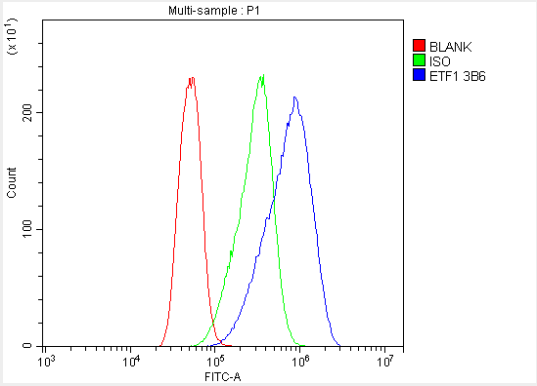

Figure 9. Flow Cytometry analysis of CACO-2 cells using anti-eRF1/ETF1 antibody (M04157). Overlay histogram showing CACO-2 cells stained with M04157 (Blue line).The cells were blocked with 10% normal goat serum. And then incubated with mouse anti-eRF1/ETF1 Antibody (M04157, 1 µg/1x10^6 cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-mouse IgG (BA1126, 5-10 µg/1x10^6 cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was mouse IgG (1 µg/1x10^6) used under the same conditions. Unlabelled sample (Red line) was also used as a control.

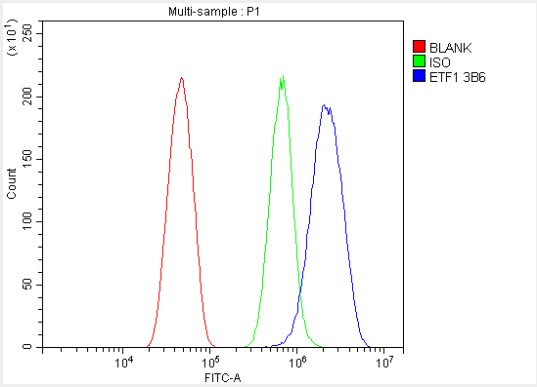

Figure 10. Flow Cytometry analysis of HEPA1-6 cells using anti-eRF1/ETF1 antibody (M04157). Overlay histogram showing HEPA1-6 cells stained with M04157 (Blue line).The cells were blocked with 10% normal goat serum. And then incubated with mouse anti-eRF1/ETF1 Antibody (M04157, 1 µg/1x10^6 cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-mouse IgG (BA1126, 5-10 µg/1x10^6 cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was mouse IgG (1 µg/1x10^6) used under the same conditions. Unlabelled sample (Red line) was also used as a control.

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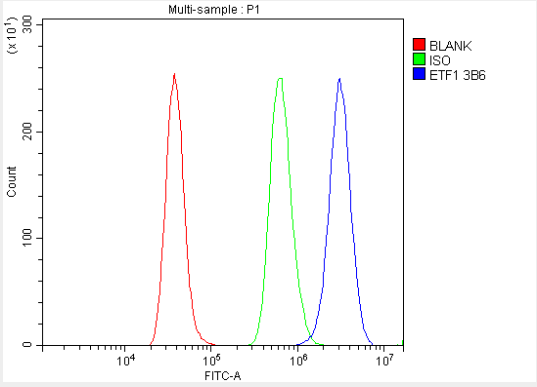

Figure 11. Flow Cytometry analysis of RH35 cells using anti-eRF1/ETF1 antibody (M04157).
Overlay histogram showing RH35 cells stained with M04157 (Blue line).The cells were blocked with 10% normal goat serum. And then incubated with mouse anti-eRF1/ETF1 Antibody (M04157, 1 µg/1x10^6 cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-mouse IgG (BA1126, 5-10 µg/1x10^6 cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was mouse IgG (1 µg/1x10^6) used under the same conditions. Unlabelled sample (Red line) was also used as a control.

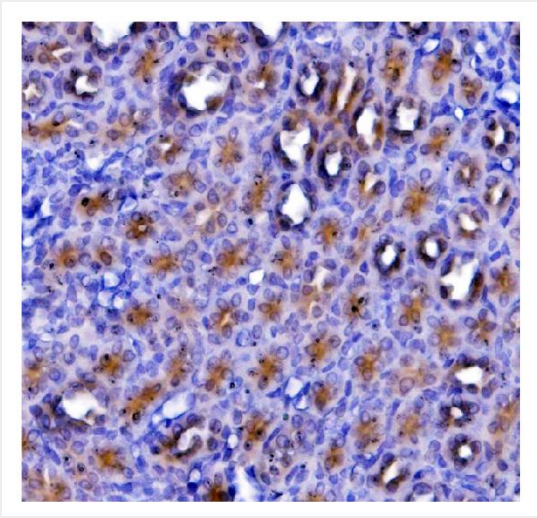

Figure 12. IHC analysis of eRF1/ETF1 using anti-eRF1/ETF1 antibody (M04157).
eRF1/ETF1 was detected in a paraffin-embedded section of rat kidney tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 µg/ml mouse anti-eRF1/ETF1 Antibody (M04157) 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.

Anti-eRF1/ETF1 Antibody Picoband™ (monoclonal, 3B6) - Background

Eukaryotic translation termination factor 1 (eRF1), also known asTB3-1, is a protein that in humans is encoded by the ETF1 gene. It is mapped to 5q31.2. This gene encodes a class-1 polypeptide chain release factor. The encoded protein plays an essential role in directing termination of mRNA translation from the termination codons UAA, UAG and UGA. This protein is a component of the SURF complex which promotes degradation of prematurely terminated mRNAs via the mechanism of nonsense-mediated mRNA decay (NMD). Alternate splicing results in multiple transcript variants. Pseudogenes of this gene are found on chromosomes 6, 7, and X.