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Anti-TIF1 gamma/TRIM33 Antibody Picoband™ (monoclonal, 8I8)

Catalog # ABO14964

Specification

Anti-TIF1 gamma/TRIM33 Antibody Picoband™ (monoclonal, 8I8) - Product Information

Application	**WB, IHC, IF, ICC, FC**
Primary Accession	Q9UPN9
Host	**Mouse**
Isotype	**Mouse IgG2b**
Reactivity	**Human**
Clonality	**Monoclonal**
Format	**Lyophilized**

Description
Anti-TIF1 gamma/TRIM33 Antibody Picoband™ (monoclonal, 8I8) . Tested in Flow Cytometry, IF, IHC, ICC, WB applications. This antibody reacts with Human.

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

Anti-TIF1 gamma/TRIM33 Antibody Picoband™ (monoclonal, 8I8) - Additional Information

Gene ID 51592

Other Names
E3 ubiquitin-protein ligase TRIM33, 2.3.2.27, Ectodermin homolog, RET-fused gene 7 protein, Protein Rfg7, RING-type E3 ubiquitin transferase TRIM33, Transcription intermediary factor 1-gamma, TIF1-gamma, Tripartite motif-containing protein 33, TRIM33, KIAA1113, RFG7, TIF1G

Calculated MW
150 kDa KDa

Application Details
Western blot, 0.1-0.5 µg/ml, Human
Immunohistochemistry (Paraffin-embedded Section), 0.5-1 µg/ml, Human
Immunocytochemistry/Immunofluorescence, 4 µg/ml, Human
Flow Cytometry, 1-3 µg/1x10^6 cells, Human

Contents
Each vial contains 4mg Trehalose, 0.9mg NaCl, 0.2mg Na2HPO4, 0.01mg NaN3.

Immunogen
E.coli-derived human TIF1 gamma recombinant protein (Position: M1001-K1127). Human TIF1 gamma shares 96.1% amino acid (aa) sequence identity with mouse TIF1 gamma.

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-TIF1 gamma/TRIM33 Antibody Picoband™ (monoclonal, 8I8) - Protein Information

Name TRIM33

Synonyms KIAA1113, RFG7, TIF1G

Function
Acts as an E3 ubiquitin-protein ligase. Promotes SMAD4 ubiquitination, nuclear exclusion and degradation via the ubiquitin proteasome pathway. According to PubMed:16751102, does not promote a decrease in the level of endogenous SMAD4. May act as a transcriptional repressor. Inhibits the transcriptional response to TGF-beta/BMP signaling cascade. Plays a role in the control of cell proliferation. Its association with SMAD2 and SMAD3 stimulates erythroid differentiation of hematopoietic stem/progenitor (By similarity). Monoubiquitinates SMAD4 and acts as an inhibitor of SMAD4-dependent TGF-beta/BMP signaling cascade (Monoubiquitination of SMAD4 hampers its ability to form a stable complex with activated SMAD2/3 resulting in inhibition of TGF-beta/BMP signaling cascade).

Cellular Location
Nucleus. Note=In discrete nuclear dots resembling nuclear bodies (By similarity). Localizes to sites of DNA damage (PubMed:25593309). {ECO:0000250|UniProtKB:Q99PP7, ECO:0000269|PubMed:25593309}

Tissue Location
Expressed in stem cells at the bottom of the crypts of the colon (at protein level). Expressed in colon adenomas and adenocarcinomas (at protein level). Expressed in brain, lung, liver, spleen, thymus, prostate, kidney, testis, heart, placenta, pancreas, small intestine, ovary, colon, skeletal muscle and hematopoietic progenitors

Anti-TIF1 gamma/TRIM33 Antibody Picoband™ (monoclonal, 8I8) - 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-TIF1 gamma/TRIM33 Antibody Picoband™ (monoclonal, 8I8) - Images

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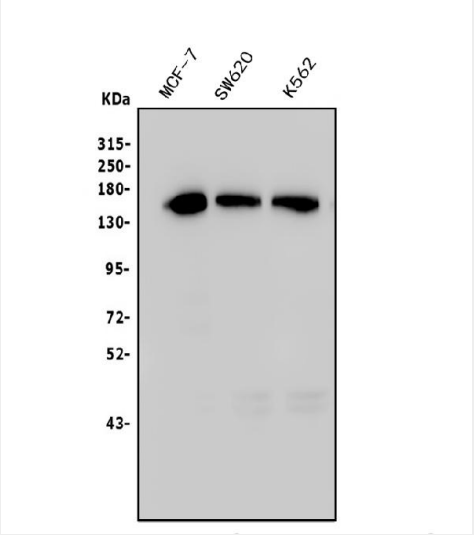

Figure 1. Western blot analysis of TIF1 gamma/TRIM33 using anti-TIF1 gamma/TRIM33 antibody (M03133-2).
Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 50ug of sample under reducing conditions.
Lane 1: human MCF-7 whole cell lysates,
Lane 2: human SW620 whole cell lysates,
Lane 3: human K562 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-TIF1 gamma/TRIM33 antigen affinity purified monoclonal antibody (Catalog # M03133-2) 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 TIF1 gamma/TRIM33 at approximately 150KD. The expected band size for TIF1 gamma/TRIM33 is at 150KD.

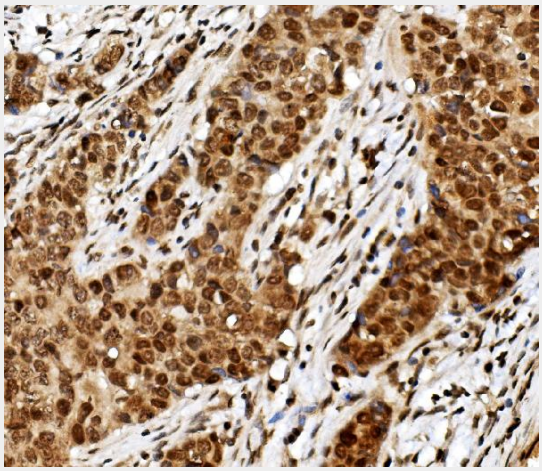

Figure 2. IHC analysis of TIF1 gamma/TRIM33 using anti TIF1 gamma/TRIM33 antibody (M03133-2).
TIF1 gamma/TRIM33 was detected in paraffin-embedded section of human mammary cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1 µg/ml mouse anti-TIF1 gamma/TRIM33 Antibody (M03133-2) 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.

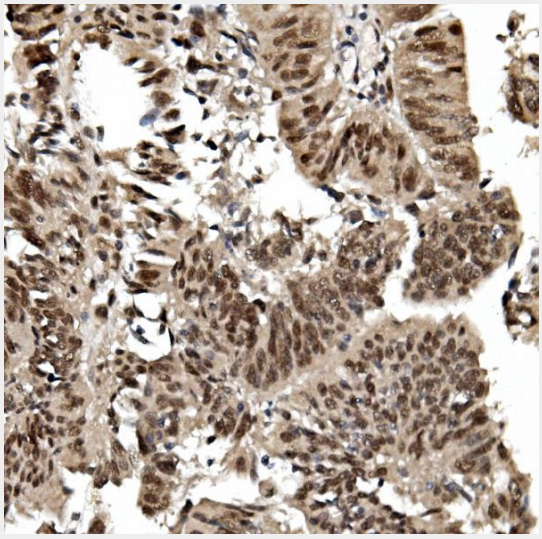

Figure 3. IHC analysis of TIF1 gamma/TRIM33 using anti TIF1 gamma/TRIM33 antibody (M03133-2).
TIF1 gamma/TRIM33 was detected in paraffin-embedded section of human rectal cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1 µg/ml mouse anti-TIF1 gamma/TRIM33 Antibody (M03133-2) 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.

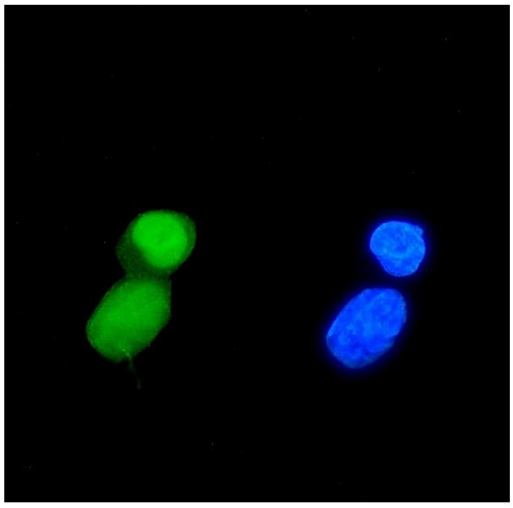

Figure 4. IF analysis of TIF1 gamma/TRIM33 using anti-TIF1 gamma/TRIM33 antibody (M03133-2). TIF1 gamma/TRIM33 was detected in immunocytochemical section of Hela cells. Enzyme antigen retrieval was performed using IHC enzyme antigen retrieval reagent (AR0022) for 15 mins. The cells were blocked with 10% goat serum. And then incubated with 4 µg/mL mouse anti-TIF1 gamma/TRIM33 Antibody (M03133-2) 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.

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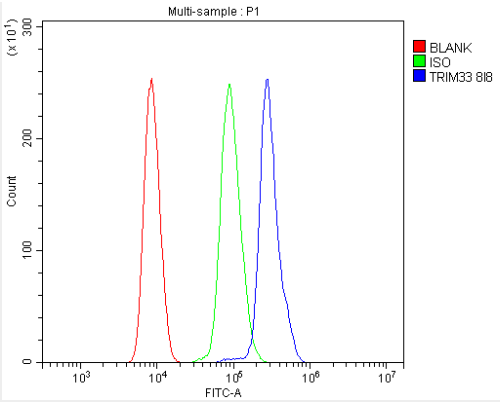

Figure 5. Flow Cytometry analysis of HL-60 cells using anti-TIF1 gamma/TRIM33 antibody (M03133-2).
Overlay histogram showing HL-60 cells stained with M03133-2 (Blue line).The cells were blocked with 10% normal goat serum. And then incubated with mouse anti-TIF1 gamma/TRIM33 Antibody (M03133-2, 1 µg/1x10^6 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.

Anti-TIF1 gamma/TRIM33 Antibody Picoband™ (monoclonal, 8I8) - Background

Tripartite motif-containing 33 (TRIM33), also known as transcriptional intermediary factor 1 gamma (TIF1-γ), is a human gene. The TRIM33 gene is mapped to chromosome 1p13 by FISH. The protein encoded by this gene is thought to be a transcriptional corepressor. However, molecules that interact with this protein have not yet been identified. The protein is a member of the tripartite motif family. This motif includes three zinc-binding domains, a RING, a B-box type 1 and a B-box type 2, and a coiled-coil region. Three alternatively spliced transcript variants for this gene have been described; however, the full-length nature of one variant has not been determined.