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Anti-TRIM25 Monoclonal Antibody
Catalog # ABO14553

Specification

Anti-TRIM25 Monoclonal Antibody - Product Information

Application	**WB, IHC, IF, ICC, IP, FC**
Primary Accession	Q14258
Host	**Rabbit**
Isotype	**Rabbit IgG**
Reactivity	**Rat, Human, Mouse**
Clonality	**Monoclonal**
Format	**Liquid**

Description
Anti-TRIM25 Monoclonal Antibody . Tested in WB, IHC, ICC/IF, IP, Flow Cytometry applications. This antibody reacts with Human, Mouse, Rat.

Anti-TRIM25 Monoclonal Antibody - Additional Information

Gene ID 7706

Other Names
E3 ubiquitin/ISG15 ligase TRIM25, 6.3.2.n3, Estrogen-responsive finger protein, RING finger protein 147, RING-type E3 ubiquitin transferase, 2.3.2.27, RING-type E3 ubiquitin transferase TRIM25, Tripartite motif-containing protein 25, Ubiquitin/ISG15-conjugating enzyme TRIM25, Zinc finger protein 147, TRIM25, EFP {ECO:0000303|PubMed:8248217}, RNF147, ZNF147

Application Details
WB 1:1000-1:5000
IHC 1:50-1:200
ICC/IF 1:100-1:500
IP 1:50
FC 1:60

Contents
Rabbit IgG in phosphate buffered saline, pH 7.4, 150mM NaCl, 0.02% sodium azide and 50% glycerol, 0.4-0.5mg/ml BSA.

Immunogen
A synthesized peptide derived from human TRIM25 Functions as an ubiquitin E3 ligase and as an ISG15 E3 ligase. Involved in innate immune defense against viruses by mediating ubiquitination of DDX58. Mediates 'Lys-63'-linked polyubiquitination of the DDX58 N-terminal CARD-like region which is crucial for triggering the cytosolic signal transduction that leads to the production of interferons in response to viral infection.

Purification
Affinity-chromatography

Storage	**Store at -20°C for one year. For short term storage and frequent use, store at 4°C for up to one month. Avoid repeated freeze-thaw cycles.**

Anti-TRIM25 Monoclonal Antibody - Protein Information

Name TRIM25

Synonyms EFP {ECO:0000303|PubMed:8248217}, RNF147

Function
Functions as a ubiquitin E3 ligase and as an ISG15 E3 ligase (PubMed:16352599). Involved in innate immune defense against viruses by mediating ubiquitination of RIGI and IFIH1 (PubMed:17392790, PubMed:29357390, PubMed:30193849, PubMed:31710640, PubMed:33849980, PubMed:36045682). Mediates 'Lys-63'-linked polyubiquitination of the RIGI N-terminal CARD-like region and may play a role in signal transduction that leads to the production of interferons in response to viral infection (PubMed:17392790, PubMed:23950712). Mediates 'Lys-63'- linked polyubiquitination of IFIH1 (PubMed:30193849). Promotes ISGylation of 14-3-3 sigma (SFN), an adapter protein implicated in the regulation of a large spectrum signaling pathway (PubMed:16352599, PubMed:17069755). Mediates estrogen action in various target organs (PubMed:22452784). Mediates the ubiquitination and subsequent proteasomal degradation of ZFHX3 (PubMed:22452784). Plays a role in promoting the restart of stalled replication forks via interaction with the KHDC3L-OOEP scaffold and subsequent ubiquitination of BLM, resulting in the recruitment and retainment of BLM at DNA replication forks (By similarity). Plays an essential role in the antiviral activity of ZAP/ZC3HAV1; an antiviral protein which inhibits the replication of certain viruses. Mechanistically, mediates 'Lys-63'-linked polyubiquitination of ZAP/ZC3HAV1 that is required for its optimal binding to target mRNA (PubMed:28060952, PubMed:28202764). Also mediates the ubiquitination of various substrates implicated in stress granule formation, nonsense-mediated mRNA decay, nucleoside synthesis and mRNA translation and stability (PubMed:36067236).

Cellular Location
Cytoplasm. Cytoplasm, Stress granule. Nucleus {ECO:0000250|UniProtKB:Q61510}

Tissue Location
Expressed in breast tumors (at protein level). Ubiquitous.

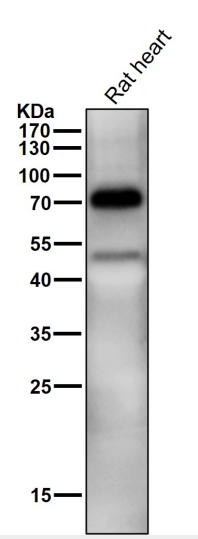
Anti-TRIM25 Monoclonal Antibody - Protocols

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-TRIM25 Monoclonal Antibody - Images



All lanes use the Antibody at 1:1W dilution for 1 hour at room temperature.

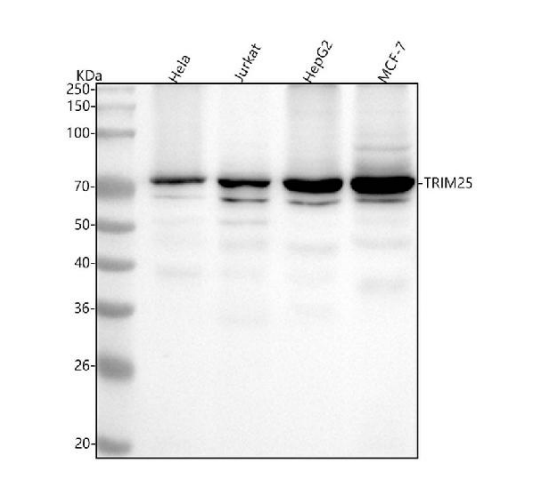


Figure 1. Western blot analysis of TRIM25 using anti-TRIM25 antibody (M03232).
Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 30 ug of sample under reducing conditions.
Lane 1: human Hela whole cell lysates,
Lane 2: human Jurkat whole cell lysates,
Lane 3: human HepG2 whole cell lysates,
Lane 4: human MCF-7 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-TRIM25 antigen affinity purified monoclonal antibody (Catalog # M03232) at 1:1000 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 1:500 for 1.5 hour at RT. The signal is developed using an Enhanced Chemiluminescent detection (ECL) kit (Catalog # EK1002) with Tanon 5200 system. A specific band was detected for TRIM25 at

approximately 71 kDa. The expected band size for TRIM25 is at 71 kDa.

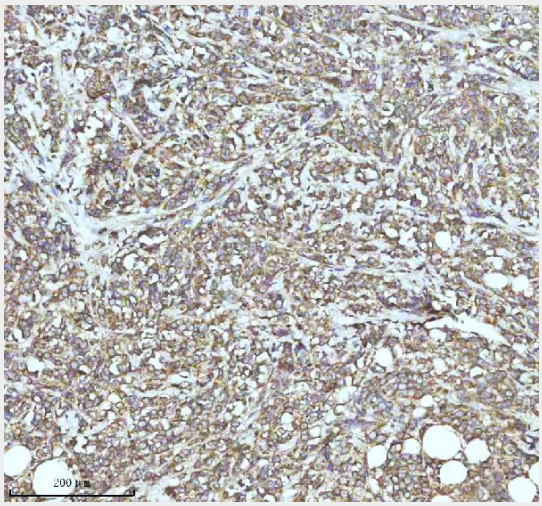


Figure 2. IHC analysis of TRIM25 using anti-TRIM25 antibody (M03232).
TRIM25 was detected in a paraffin-embedded section of human breast cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1:50 rabbit anti-TRIM25 Antibody (M03232) overnight at 4°C. Peroxidase Conjugated Goat Anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Rabbit IgG Super Vision Assay Kit (Catalog # SV0002) with DAB as the chromogen.

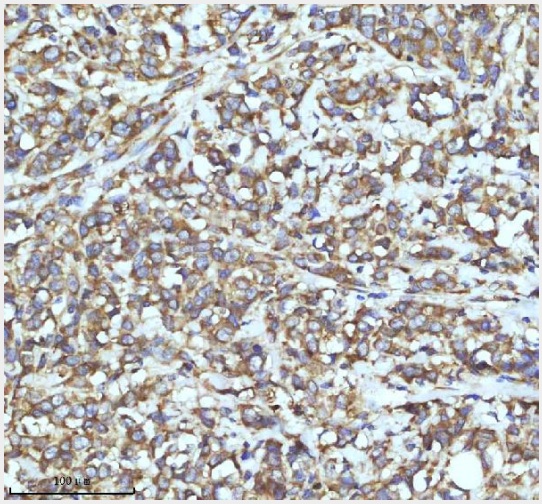


Figure 3. IHC analysis of TRIM25 using anti-TRIM25 antibody (M03232).
TRIM25 was detected in a paraffin-embedded section of human breast cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1:50 rabbit anti-TRIM25 Antibody (M03232) overnight at 4°C. Peroxidase Conjugated Goat Anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Rabbit IgG Super Vision Assay Kit (Catalog # SV0002) with DAB as the chromogen.

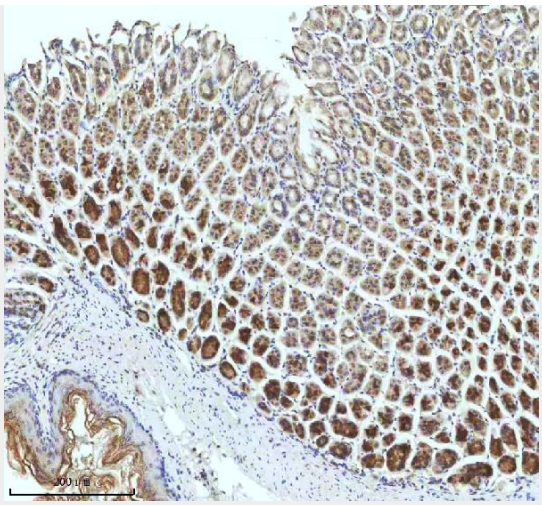


Figure 4. IHC analysis of TRIM25 using anti-TRIM25 antibody (M03232).
TRIM25 was detected in a paraffin-embedded section of mouse stomach tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1:50 rabbit anti-TRIM25 Antibody (M03232) overnight at 4°C. Peroxidase Conjugated Goat Anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Rabbit IgG Super Vision Assay Kit (Catalog # SV0002) with DAB as the chromogen.

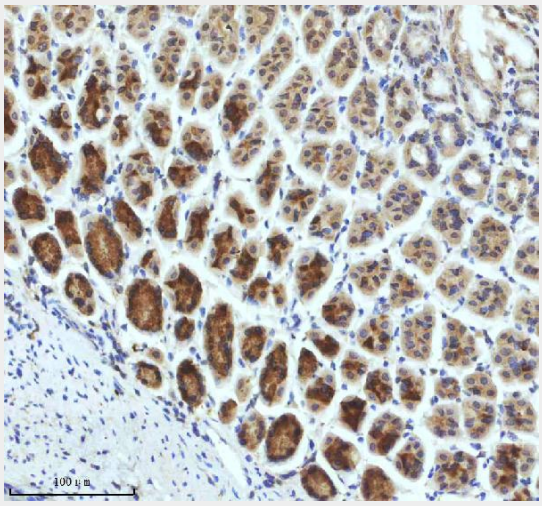


Figure 5. IHC analysis of TRIM25 using anti-TRIM25 antibody (M03232).
TRIM25 was detected in a paraffin-embedded section of mouse stomach tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1:50 rabbit anti-TRIM25 Antibody (M03232) overnight at 4°C. Peroxidase Conjugated Goat Anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Rabbit IgG Super Vision Assay Kit (Catalog # SV0002) with DAB as the chromogen.

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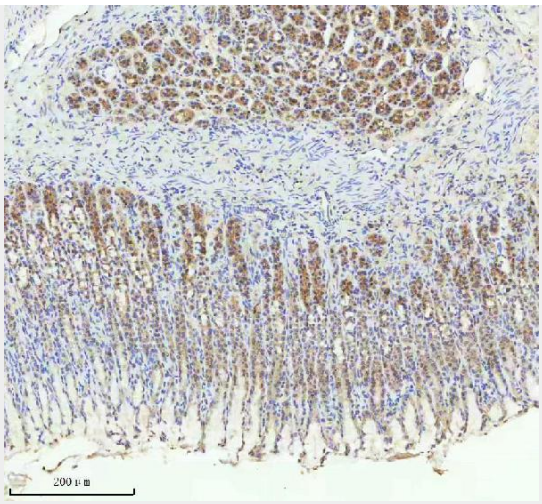

Figure 6. IHC analysis of TRIM25 using anti-TRIM25 antibody (M03232).
TRIM25 was detected in a paraffin-embedded section of rat stomach tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1:50 rabbit anti-TRIM25 Antibody (M03232) overnight at 4°C. Peroxidase Conjugated Goat Anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Rabbit IgG Super Vision Assay Kit (Catalog # SV0002) with DAB as the chromogen.

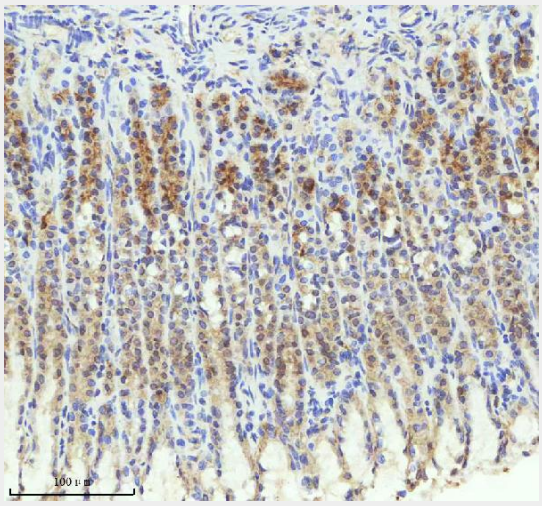

Figure 7. IHC analysis of TRIM25 using anti-TRIM25 antibody (M03232).
TRIM25 was detected in a paraffin-embedded section of rat stomach tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1:50 rabbit anti-TRIM25 Antibody (M03232) overnight at 4°C. Peroxidase Conjugated Goat Anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Rabbit IgG Super Vision Assay Kit (Catalog # SV0002) with DAB as the chromogen.