

Anti-Ubiquitin Polyclonal Antibody

Product Details

Ig Type:	IgG
Reactivity:	Human,Mouse,Rat (predicted:Pig)
Molecular Weight:	Theoretical: 8.5 kDa. Actual: 8.5 kDa.
Purification:	Protein A purified

Applications

1. Blank control (black line): HepG2. Primary Antibody (green line): Rabbit Anti-Ubiquitin antibody (TMAB-01928)

Dilution: 1 µg/Test;

Secondary Antibody: Goat anti-rabbit IgG-AF488

Dilution: 0.5 µg/Test.

Negative control (white blue line): PBS

Isotype control (orange line): Normal Rabbit IgG

Protocol

The cells were fixed with 4% PFA (10 min at room temperature) and then permeabilized with 90% ice-cold methanol for 20 min at -20°C. The cells were then incubated in 5% BSA to block non-specific protein-protein interactions for 30 min at room temperature. Cells stained with Primary Antibody for 30 min at room temperature. The secondary antibody used for 40 min at room temperature.

2. Blank control (Black line): Molt4 (Black).

Primary Antibody (green line): Rabbit Anti-Ubiquitin antibody (TMAB-01928)

Dilution: 1 µg/10⁶ cells;

Isotype Control Antibody (orange line): Rabbit IgG.

Secondary Antibody (white blue line): Goat anti-rabbit IgG-AF647

Dilution: 1 µg/test.

Verified Activity:

Protocol

The cells were fixed with 4% PFA (10 min at room temperature) and then permeabilized with 90% ice-cold methanol for 20 min at room temperature. The cells were then incubated in 5% BSA to block non-specific protein-protein interactions for 30 min at room temperature. Cells stained with Primary Antibody for 30 min at room temperature. The secondary antibody used for 40 min at room temperature.

3. Sample: Ubiquitin protein at 100ng;

Primary: Anti-Ubiquitin (TMAB-01928) at 1:300 dilution;

Secondary: HRP conjugated Goat-Anti-rabbit IgG (secondary antibody) at 1: 5000 dilution;

Predicted band size: 8.5 kDa

Observed band size: 8.5 kDa

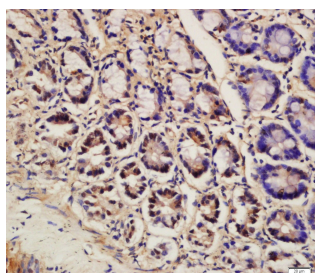
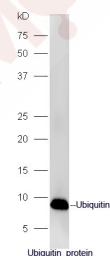
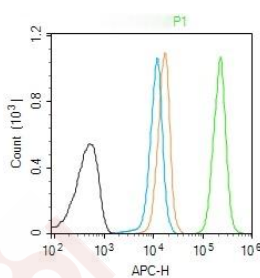
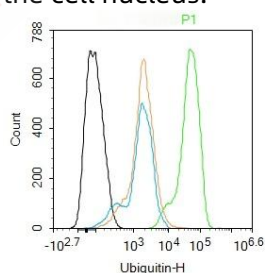
4. Paraformaldehyde-fixed, paraffin embedded (rat colon tissue); Antigen retrieval by boiling in sodium citrate buffer (pH6.0) for 15 min; Block endogenous peroxidase by 3% hydrogen peroxide for 20 min; Blocking buffer (normal goat serum) at 37°C for 30 min; Antibody incubation with (Ubiquitin) Polyclonal Antibody, Unconjugated (TMAB-01928) at 1:400 overnight at 4°C, followed by a conjugated secondary for 20 min and DAB staining.

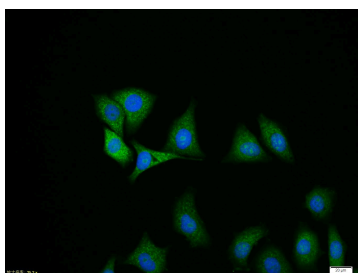
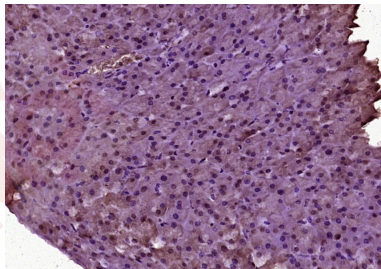
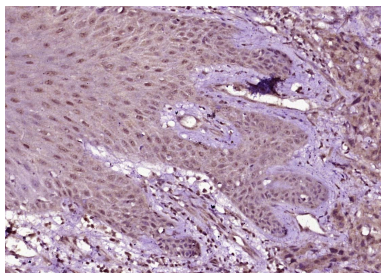
5. Paraformaldehyde-fixed, paraffin embedded (Human skin carcinoma); Antigen retrieval by boiling in sodium citrate buffer (pH6.0) for 15 min; Block endogenous peroxidase by 3% hydrogen peroxide for 20 min; Blocking buffer (normal goat serum) at 37°C for 30 min;

Antibody incubation with (Ubiquitin) Polyclonal Antibody, Unconjugated (TMAB-01928) at 1:400 overnight at 4°C, followed by operating according to SP Kit (Rabbit) instructions and DAB staining.

6. Paraformaldehyde-fixed, paraffin embedded (Mouse pancreas); Antigen retrieval by boiling in sodium citrate buffer (pH6.0) for 15 min; Block endogenous peroxidase by 3% hydrogen peroxide for 20 min; Blocking buffer (normal goat serum) at 37°C for 30 min; Antibody incubation with (Ubiquitin) Polyclonal Antibody, Unconjugated (TMAB-01928) at 1:400 overnight at 4°C, followed by operating according to SP Kit (Rabbit) instructions and DAB staining.

7. HepG2 cell; 4% Paraformaldehyde-fixed; Triton X-100 at room temperature for 20 min; Blocking buffer (normal goat serum) at 37°C for 20 min; Antibody incubation with (Ubiquitin) polyclonal Antibody, Unconjugated (TMAB-01928) 1:100, 90 minutes at 37°C; followed by a conjugated Goat Anti-Rabbit IgG antibody at 37°C for 90 minutes, DAPI (blue) was used to stain the cell nucleus.





Application: FCM,ICC/IF,IF,IHC-Fr,IHC-P,WB

Recommended FCM=1 µg/Test; ICC/IF=1:100-500; IF=1:100-500; IHC-Fr=1:100-500; IHC-P=1:100-500; WB=1:500-2000

Properties

Stability & Storage: Store at -20°C or -80°C for 12 months. Avoid repeated freeze-thaw cycles.

Shipping: Shipping with blue ice.

Antigen Details

Immunogen: KLH conjugated synthetic peptide: human Ubiquitin

Antigen Species: Human

Gene ID: 7314

Uniprot ID: P0CG47

Synonyms: UBA 52;MGC8385;Polyubiquitin-B;ubiquitin B;UBCEP 2;UBCEP 1;UB;UBCEP2;UBCEP1;UBC;UBA 80;FLJ25987;Polyubiquitin-C;UBA80;RPS 27A;HMG20;UBB;RS27A;RPS27A;Ubiquitin carboxyl extension protein 80;Ubiquitin;UBA52

Biology Area: Proteasome,Ubiquitin

Research Background

This gene encodes ubiquitin, one of the most conserved proteins known. Ubiquitin has a major role in targeting cellular proteins for degradation by the 26S proteasome. It is also involved in the maintenance of chromatin structure, the regulation of gene expression, and the stress response. Ubiquitin is synthesized as a precursor protein consisting of either polyubiquitin chains or a single ubiquitin moiety fused to an unrelated protein. This gene consists of three direct repeats of the ubiquitin coding sequence with no spacer sequence. Consequently, the protein is expressed as a polyubiquitin precursor with a final amino acid after the last repeat. An aberrant form of this

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protein has been detected in patients with Alzheimer's disease and Down syndrome. Pseudogenes of this gene are located on chromosomes 1, 2, 13, and 17. Alternative splicing results in multiple transcript variants. [provided by RefSeq, Aug 2013]

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