

Anti-p63 Antibody [SC06-31]

ET1610-44



Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human, Mouse, Rat
Applications:	WB, IF-Cell, IF-Tissue, IHC-P, FC
Molecular Wt:	Predicted band size: 77 kDa
Clone number:	SC06-31

Description: The p53 tumor suppressor protein plays a major role in cellular response to DNA damage and other genomic aberrations. Activation of p53 can lead to either cell cycle arrest and DNA repair or apoptosis. In addition to p53, mammalian cells contain two p53 family members, p63 and p73, which are similar to p53 in both structure and function. While p63 can induce p53-responsive genes and apoptosis, mutation of p63 rarely results in tumors. Research investigators frequently observe amplification of the p63 gene in squamous cell carcinomas of the lung, head and neck. The p63 gene contains an alternative transcription initiation site that yields a 40 kDa δ Np63 lacking the transactivation domain, and alternative splicing at the carboxy-terminus yields the α , β , and γ isoforms.

Immunogen: Recombinant protein within human p63 aa 1-250.

Positive control: A431 cell lysate, MCF7 cell lysate, HaCaT cell lysate, Mouse skin tissue lysate, Rat skin tissue lysate, A431, human skin tissue, human breast tissue, human esophagus tissue, mouse esophagus tissue, rat esophagus tissue.

Subcellular location: Nucleus.

Database links: SwissProt: Q9H3D4 Human | O88898 Mouse | Q9JJP6 Rat

Recommended Dilutions:

WB	1:1,000-1:2,000
IF-Cell	1:500-1:2,000
IF-Tissue	1:50-1:200
IHC-P	1:800-1:2,000
FC	1:1,000

Storage Buffer: 1*TBS (pH7.4), 0.05% BSA, 40% Glycerol. Preservative: 0.05% Sodium Azide.

Storage Instruction: Shipped at 4°C. Store at +4°C short term (1-2 weeks). It is recommended to aliquot into single-use upon delivery. Store at -20°C long term.

Purity: Protein A affinity purified.

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Orders:0086-571-88062880

Technical:0086-571-89986345

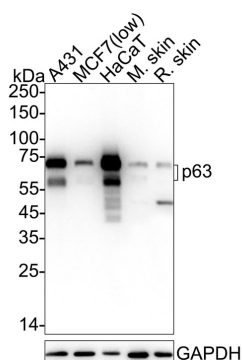
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Applications:WB=Western blot IHC-P=Immunohistochemistry (paraffin) IF-Cell=Immunofluorescence (Cell) IF-Tissue=Immunofluorescence (Tissue) FC=Flow cytometry IP=Immunoprecipitation

Images

Fig1: Western blot analysis of p63 on different lysates with Rabbit anti-p63 antibody (ET1610-44) at 1/1,000 dilution.



Lane 1: A431 cell lysate (15 µg/Lane)
 Lane 2: MCF7 cell lysate (low expression) (15 µg/Lane)
 Lane 3: HaCaT cell lysate (15 µg/Lane)
 Lane 4: Mouse skin tissue lysate (30 µg/Lane)
 Lane 5: Rat skin tissue lysate (30 µg/Lane)

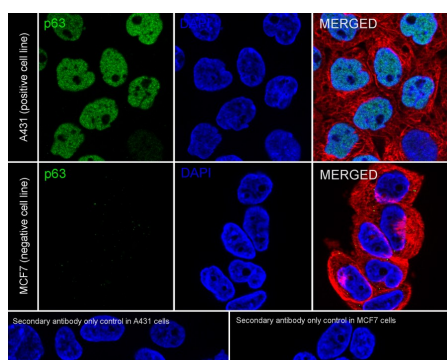
Predicted band size: 77 kDa
 Observed band size: 60-70 kDa

Exposure time: 10 seconds; ECL: K1801;

4-20% SDS-PAGE gel.

Proteins were transferred to a PVDF membrane and blocked with 5% NFDM/TBST for 1 hour at room temperature. The primary antibody (ET1610-44) at 1/1,000 dilution was used in 5% NFDM/TBST at 4°C overnight. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1/50,000 dilution was used for 1 hour at room temperature.

Fig2: Immunocytochemistry analysis of A431 (positive) and MCF7 (negative) labeling p63 with Rabbit anti-p63 antibody (ET1610-44) at 1/2,000 dilution.



Cells were fixed in 4% paraformaldehyde for 20 minutes at room temperature, permeabilized with 0.1% Triton X-100 in PBS for 5 minutes at room temperature, then blocked with 1% BSA in 10% negative goat serum for 1 hour at room temperature. Cells were then incubated with Rabbit anti-p63 antibody (ET1610-44) at 1/2,000 dilution in 1% BSA in PBST overnight at 4 °C. Goat Anti-Rabbit IgG H&L (iFluor™ 488, HA1121) was used as the secondary antibody at 1/1,000 dilution. PBS instead of the primary antibody was used as the secondary antibody only control. Nuclear DNA was labelled in blue with DAPI.

Beta tubulin (M1305-2, red) was stained at 1/100 dilution overnight at +4°C. Goat Anti-Mouse IgG H&L (iFluor™ 594, HA1126) was used as the secondary antibody at 1/1,000 dilution.

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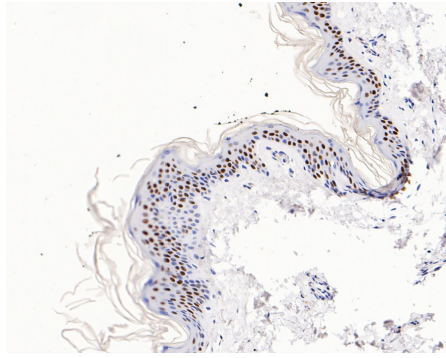


Fig3: Immunohistochemical analysis of paraffin-embedded human skin tissue with Rabbit anti-p63 antibody (ET1610-44) at 1/800 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) (high pressure) for 2 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (ET1610-44) at 1/800 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

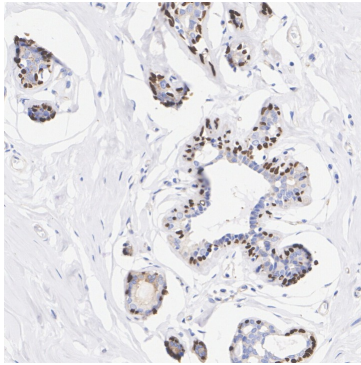


Fig4: Immunohistochemical analysis of paraffin-embedded human breast tissue with Rabbit anti-p63 antibody (ET1610-44) at 1/2,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) (high pressure) for 2 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (ET1610-44) at 1/2,000 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

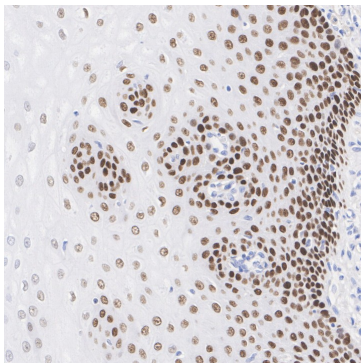


Fig5: Immunohistochemical analysis of paraffin-embedded human esophagus tissue with Rabbit anti-p63 antibody (ET1610-44) at 1/2,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) (high pressure) for 2 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (ET1610-44) at 1/2,000 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

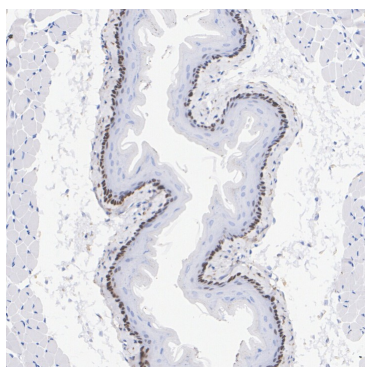


Fig6: Immunohistochemical analysis of paraffin-embedded mouse esophagus tissue with Rabbit anti-p63 antibody (ET1610-44) at 1/2,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) (high pressure) for 2 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (ET1610-44) at 1/2,000 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

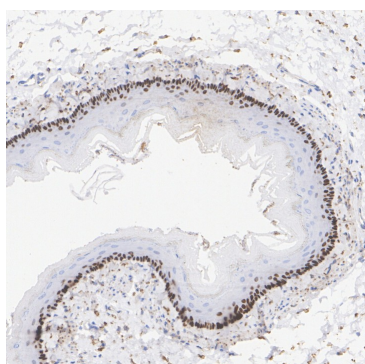


Fig7: Immunohistochemical analysis of paraffin-embedded rat esophagus tissue with Rabbit anti-p63 antibody (ET1610-44) at 1/2,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) (high pressure) for 2 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (ET1610-44) at 1/2,000 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

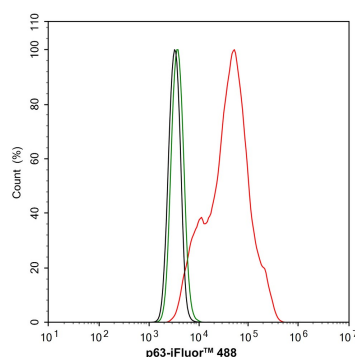


Fig8: Flow cytometric analysis of A431 cells labeling p63.

Cells were fixed and permeabilized. Then stained with the primary antibody (ET1610-44, 1/1,000) (red) compared with Rabbit IgG Isotype Control (green). After incubation of the primary antibody at +4°C for an hour, the cells were stained with a iFluor™ 488 conjugate-Goat anti-Rabbit IgG Secondary antibody (HA1121) at 1/1,000 dilution for 30 minutes at +4°C. Unlabelled sample was used as a control (cells without incubation with primary antibody; black).

Note: All products are "FOR RESEARCH USE ONLY AND ARE NOT INTENDED FOR DIAGNOSTIC OR THERAPEUTIC USE".

Background References

1. Bag S et al. Connecting cyto-nano-architectural attributes and epithelial molecular expression in oral submucous fibrosis progression to cancer. J Clin Pathol 68:605-13 (2015).
2. Yu XM et al. Reduced growth and proliferation dynamics of nasal epithelial stem/progenitor cells in nasal polyps in vitro. Sci Rep 4:4619 (2014).

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