# **Anti-UBC9 Antibody [SC0534]**

### ET1610-21



Product Type: Recombinant Rabbit monoclonal IgG, primary antibodies

Species reactivity: Human, Mouse, Rat

Applications: WB, IF-Cell, IF-Tissue, IHC-P, IP

Molecular Wt: 18 kDa
Clone number: SC0534

**Description:** UBC9 is a component of the ubiquitin-mediated proteolytic pathway, which targets proteins

for degradation by the 26S proteasome, mediates endocytosis and directs protein subcellular localization. Ub and Ub-like molecules are systematically transferred from E2 conjugating enzymes to the targeted substrate by way of an E3 ubiquitin ligase. UBC9 functions as an E2 ubiquitin conjugating enzyme that preferentially associates with the ubiquitin homolog designated SUMO-1 or sentrin, a component of the sentrinization complex. Characteristic of the E2 family members, UBC9 contains a conserved cysteine residue that is required for the thio ester formation between Ub-like proteins and the E2 member, and it shares a conserved UBC domain. Substrates for UBC9 include transcription factors E12 and E47 and mitotic regulators RanBP2 and RanGAP1, which indicates that UBC9 may regulate various cellular

processes including cell cycle progression and differentiation.

**Immunogen:** Synthetic peptide within Human UBC9 aa 15-60 / 158.

Positive control: HUVEC cell lysate, HepG2 cell lysate, Hela cell lysate, SH-SY5Y cell lysate, A431, HepG2,

human kidney tissue, mouse testis tissue.

Subcellular location: Nucleus, Cytoplasm.

Database links: SwissProt: P63279 Human | P63280 Mouse | P63281 Rat

Recommended Dilutions:

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

**IP** Use at an assay dependent concentration.

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

**Storage Instruction:** Shipped at  $4^{\circ}$ C. Store at  $+4^{\circ}$ 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.

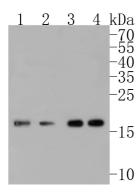
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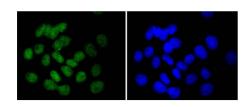
#### **Images**



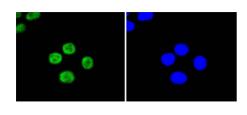
**Fig1:** Western blot analysis of UBC9 on different lysates. Proteins were transferred to a PVDF membrane and blocked with 5% BSA in PBS for 1 hour at room temperature. The primary antibody (ET1610-21, 1/500) was used in 5% BSA at room temperature for 2 hours. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1:5,000 dilution was used for 1 hour at room temperature.

#### Positive control:

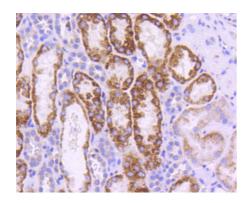
Lane 1: HUVEC cell lysate Lane 2: HepG2 cell lysate Lane 3: Hela cell lysate Lane 4: SH-SY5Y cell lysate



**Fig2:** ICC staining of UBC9 in A431 cells (green). Formalin fixed cells were permeabilized with 0.1% Triton X-100 in TBS for 10 minutes at room temperature and blocked with 1% Blocker BSA for 15 minutes at room temperature. Cells were probed with the primary antibody (ET1610-21, 1/50) for 1 hour at room temperature, washed with PBS. Alexa Fluor®488 Goat anti-Rabbit IgG was used as the secondary antibody at 1/1,000 dilution. The nuclear counter stain is DAPI (blue).



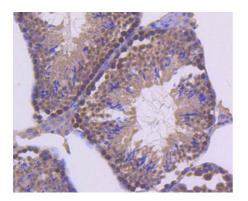
**Fig3:** ICC staining of UBC9 in HepG2 cells (green). Formalin fixed cells were permeabilized with 0.1% Triton X-100 in TBS for 10 minutes at room temperature and blocked with 1% Blocker BSA for 15 minutes at room temperature. Cells were probed with the primary antibody (ET1610-21, 1/50) for 1 hour at room temperature, washed with PBS. Alexa Fluor®488 Goat anti-Rabbit IgG was used as the secondary antibody at 1/1,000 dilution. The nuclear counter stain is DAPI (blue).



**Fig4:** Immunohistochemical analysis of paraffin-embedded human kidney tissue using anti-UBC9 antibody. The section was pretreated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 8.0-8.4) for 20 minutes. The tissues were blocked in 5% BSA for 30 minutes at room temperature, washed with ddH<sub>2</sub>O and PBS, and then probed with the primary antibody (ET1610-21, 1/50) for 30 minutes 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.

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**Fig5:** Immunohistochemical analysis of paraffin-embedded mouse testis tissue using anti-UBC9 antibody. The section was pretreated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 8.0-8.4) for 20 minutes. The tissues were blocked in 5% BSA for 30 minutes at room temperature, washed with ddH<sub>2</sub>O and PBS, and then probed with the primary antibody (ET1610-21, 1/50) for 30 minutes 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.

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

#### **Background References**

- 1. Hsieh YL et al. Ubc9 acetylation modulates distinct SUMO target modification and hypoxia response. EMBO J 32:791-804 (2013).
- 2. Santos A et al. SUMOylation affects the interferon blocking activity of the influenza A nonstructural protein NS1 without affecting its stability or cellular localization. J Virol 87:5602-20 (2013).