

DESCRIPTION

Species Reactivity	Human
Specificity	Detects Human UBE2I/Ubc9 in direct Elisa.
Source	Monoclonal Mouse IgG ₁ Clone # 1050001
Purification	Protein A or G purified from hybridoma culture supernatant
Immunogen	E. coli-derived human UBE2I/Ubc9 protein Met1-Ser158 Accession # P63279
Formulation	Lyophilized from a 0.2 µm filtered solution in PBS with Trehalose. See Certificate of Analysis for details. *Small pack size (-SP) is supplied either lyophilized or as a 0.2 µm filtered solution in PBS.

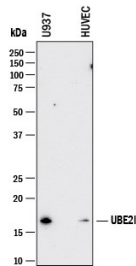
APPLICATIONS

Please Note: Optimal dilutions should be determined by each laboratory for each application. [General Protocols](#) are available in the Technical Information section on our website.

	Recommended Concentration	Sample
Western Blot	2 µg/mL	U937 human histiocytic lymphoma cell line, HUVEC human umbilical vein endothelial cells
Immunocytochemistry	8-25 µg/mL	Immersion fixed A431 cells

DATA

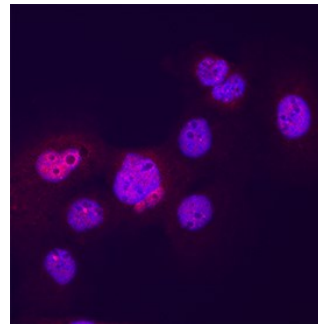
Western Blot



Detection of Human UBE2I/Ubc9 by Western Blot.

Western blot shows lysates of U937 human histiocytic lymphoma cell line, HUVEC human umbilical vein endothelial cells. PVDF membrane was probed with 2 µg/mL of Mouse Anti-Human UBE2I/Ubc9 Monoclonal Antibody (Catalog # MAB11185) followed by HRP-conjugated Anti-Mouse IgG Secondary Antibody (Catalog # HAF018). A specific band was detected for UBE2I/Ubc9 at approximately 18 kDa (as indicated). This experiment was conducted under reducing conditions and using Western Blot Buffer Group 1.

Immunocytochemistry



Detection of UBE2I/Ubc9 in A431 Human Epithelial Carcinoma Cell Line.

UBE2I/Ubc9 was detected in immersion fixed A431 cells using Mouse Anti-Human UBE2I/Ubc9 Monoclonal Antibody (Catalog # MAB11185) at 8 µg/mL for 3 hours at room temperature. Cells were stained using the NorthernLights™ 557-conjugated Anti-Rat IgG Secondary Antibody (red; Catalog # NL013) and counterstained with DAPI (blue). Specific staining was localized to Nuclei. View our protocol for [Fluorescent ICC Staining of Cells on Coverslips](#).

PREPARATION AND STORAGE

Reconstitution	Reconstitute at 0.5 mg/mL in sterile PBS.
Shipping	The product is shipped at ambient temperature. Upon receipt, store it immediately at the temperature recommended below. *Small pack size (-SP) is shipped with polar packs. Upon receipt, store it immediately at -20 to -70 °C
Stability & Storage	Use a manual defrost freezer and avoid repeated freeze-thaw cycles. <ul style="list-style-type: none"> • 12 months from date of receipt, -20 to -70 °C as supplied. • 1 month, 2 to 8 °C under sterile conditions after reconstitution. • 6 months, -20 to -70 °C under sterile conditions after reconstitution.

BACKGROUND

Ubiquitin-conjugating Enzyme E2I (UBE2I), also known as Ubiquitin-conjugating Enzyme 9 (Ubc9), is a ubiquitously expressed protein with a predicted molecular weight of 20 kDa. Human UBE2I/Ubc9 shares 100% amino acid sequence identity with the mouse and rat orthologs. UBE2I/Ubc9 catalyzes the addition of the Ubiquitin-like protein SUMO to target proteins (1,2). SUMO is transferred from a Ubiquitin-like activating (E1) enzyme heterodimer consisting of SAE1 and UBA2 to Cys93 of UBE2I/Ubc9 (3). In contrast to most Ubiquitin-conjugating (E2) enzymes that function in a complex with Ubiquitin ligases (E3s), UBE2I/Ubc9 can interact directly with protein substrates and may also play a role in substrate recognition (4). UBE2I/Ubc9 mediates the SUMOylation of a variety of proteins including RanGAP1, HDAC4, and PML (5,6). Post-translational modifications of UBE2I/Ubc9, such as auto-SUMOylation at Lys14 or Ser71 phosphorylation by CDC2/CDK1, alter UBE2I/Ubc9 catalytic activity and target protein recognition (5,7). UBE2I/Ubc9-dependent SUMOylation reduces the levels of the stem cell marker Nanog, implicating it in embryonic stem cell pluripotency maintenance (8). SUMOylation also regulates the protein levels of tumor suppressors and oncogenes, and UBE2I/Ubc9 dysregulation is thought to contribute to the pathogenesis of human cancers (9).

References:

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4. Yunus, A.A. & C.D. Lima (2006) *Nat. Struct. Mol. Biol.* 13:491.
5. Knipscheer, P. et al. (2008) *Mol. Cell* 31:371.
6. Duprez, E. et al. (1999) *J. Cell Sci.* 112:381.
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MANUFACTURING SPECIFICATIONS