Anti-E-Cadherin Antibody [A0-G11-2]

EM0502

Product Type:	Mouse monoclonal IgG1, primary antibodies
Species reactivity:	Human, Mouse, Rat
Applications:	WB, IF-Cell, IHC-P
Molecular Wt:	Predicted band size: 97 kDa
Clone number:	A0-G11-2

Description: E-cadherin (epithelial) is the most well-studied member of the cadherin family. It consists of 5 cadherin repeats (EC1 ~ EC5) in the extracellular domain, one transmembrane domain, and an intracellular domain that binds p120-catenin and beta-catenin. The intracellular domain contains a highly-phosphorylated region vital to beta-catenin binding and, therefore, to E-cadherin function. Loss of E-cadherin function or expression has been implicated in cancer progression and metastasis. E-cadherin downregulation decreases the strength of cellular adhesion within a tissue, resulting in an increase in cellular motility. This in turn may allow cancer cells to cross the basement membrane and invade surrounding tissues. E-cadherin is also used by pathologists to diagnose different kinds of breast cancer.

Immunogen: Recombinant protein within mouse E-Cadherin aa 350-550.

Positive control: SW480 cell lysate, A431 cell lysate, A431, human liver carcinoma tissue, human colon carcinoma tissue.

Subcellular location: Cell membrane.

Database links: SwissProt: P12830 Human | P09803 Mouse

Recommended Dilutions:

WB	1:1,000
IF-Cell	1:200
IHC-P	1:200-1:1,000

Storage Buffer: 1*PBS (pH7.4), 0.2% BSA, 40% Glycerol. Preservative: 0.05% Sodium Azide.

Storage Instruction: Store at +4℃ after thawing. Aliquot store at -20℃ or -80℃. Avoid repeated freeze / thaw cycles.

Purity: Protein A affinity purified.

Images

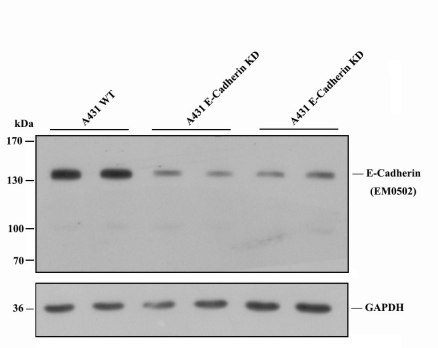

Fig1: All lanes: Western blot analysis of E-Cadherin with anti-E-Cadherin antibody [A0-G11-2] (EM0502) at 1:1,000 dilution.
Lane 1/2: Wild-type A431 whole cell lysate (20 µg).
Lane 3/4: E-Cadherin fragment 1 knockdown A431 whole cell lysate (20 µg).
Lane 5/6: E-Cadherin fragment 2 knockdown A431 whole cell lysate (20 µg).

EM0502 was shown to specifically react with E-Cadherin in wild-type A431 cells. Weakened bands were observed when E-Cadherin knockdown samples were tested. Wild-type and E-Cadherin knockdown samples were subjected to SDS-PAGE. Proteins were transferred to a PVDF membrane and blocked with 5% NFDM in TBST for 1 hour at room temperature. The primary antibody (EM0502, 1/1,000) and Loading control antibody (Rabbit anti-GAPDH , ET1601-4, 1/10,000) were used in 5% BSA at room temperature for 2 hours. Goat Anti-Mouse IgG-HRP Secondary Antibody (HA1006) at 1:20,000 dilution was used for 1 hour at room temperature.

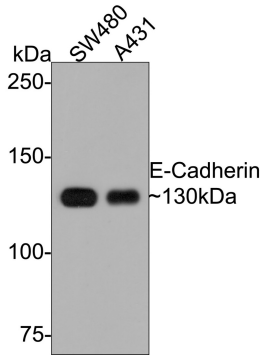

Fig2: Western blot analysis of E-Cadherin on different lysates with Mouse anti-E-Cadherin antibody (EM0502) at 1/500 dilution.

Lane 1: SW480 cell lysate
Lane 2: A431 cell lysate

Lysates/proteins at 10 µg/Lane.

Predicted band size: 97 kDa
Observed band size: 130 kDa

Exposure time: 2 minutes;

6% 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 (EM0502) at 1/500 dilution was used in 5% NFDM/TBST at room temperature for 2 hours. Goat Anti-Mouse IgG - HRP Secondary Antibody (HA1006) at 1:100,000 dilution was used for 1 hour at room temperature.

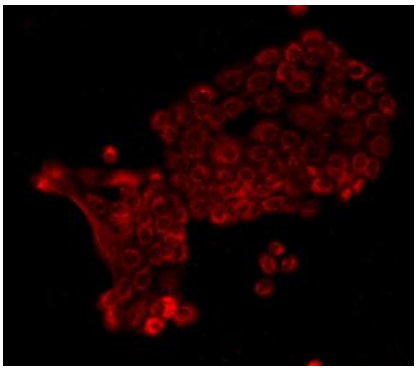

Fig3: ICC staining of E-Cadherin in A431 cells (red). Formalin fixed cells were permeabilized with 0.1% Triton X-100 in TBS for 10 minutes at room temperature and blocked with 10% negative goat serum for 15 minutes at room temperature. Cells were probed with the primary antibody (EM0502, 1/50) for 1 hour at room temperature, washed with PBS. Alexa Fluor®555 conjugate-Goat anti-Mouse IgG was used as the secondary antibody at 1/1,000 dilution.

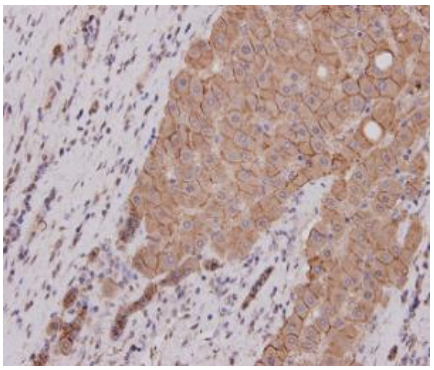

Fig4: Immunohistochemical analysis of paraffin-embedded human liver carcinoma tissue using anti-E-Cadherin antibody. The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes.The tissues were blocked in 1% BSA for 30 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (EM0502, 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.

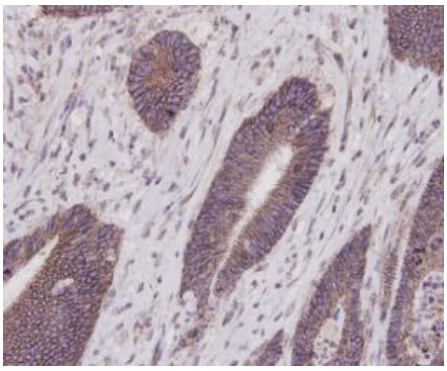

Fig5: Immunohistochemical analysis of paraffin-embedded human colon carcinoma tissue using anti-E-Cadherin antibody. The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes.The tissues were blocked in 1% BSA for 30 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (EM0502, 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.

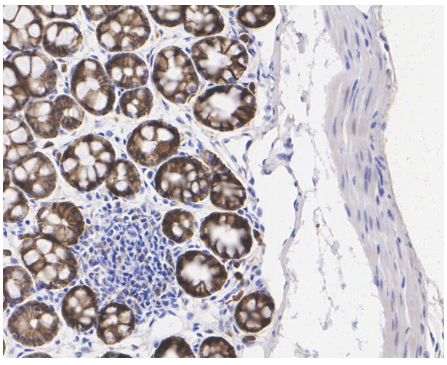

Fig6: Immunohistochemical analysis of paraffin-embedded mouse colon tissue with Mouse anti-E-Cadherin antibody (EM0502) at 1/1,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (EM0502) at 1/1,000 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.

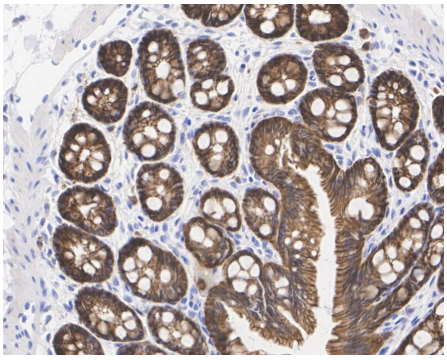

Fig7: Immunohistochemical analysis of paraffin-embedded rat colon tissue with Mouse anti-E-Cadherin antibody (EM0502) at 1/1,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (EM0502) at 1/1,000 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.

Background References

1. Eger A, et al. (Mar 2005). "DeltaEF1 is a transcriptional repressor of E-cadherin and regulates epithelial plasticity in breast cancer cells." Oncogene 24 (14): 2375-85.
2. Liu YN, et al. (Dec 2005). "Regulatory mechanisms controlling human E-cadherin gene expression." Oncogene 24 (56): 8277-90.
3. Lombaerts M, et al. (Mar 2006). "E-cadherin transcriptional downregulation by promoter methylation but not mutation is related to epithelial-to-mesenchymal transition in breast cancer cell lines." Br J Cancer 94 (5): 661-71.