

Anti-DFNA5 / GSDME Antibody [A1H3]

EM1901-48



Product Type:	Mouse monoclonal IgG1, primary antibodies
Species reactivity:	Human
Applications:	WB, IF-Cell, IHC-P, FC
Molecular Wt:	Predicted band size: 55 kDa
Clone number:	A1H3

Description: Non-syndromic hearing impairment protein 5 is a protein that in humans is encoded by the DFNA5 gene. Hearing impairment is a heterogeneous condition with over 40 loci described. The protein encoded by this gene is expressed in fetal cochlea, however, its function is not known. Nonsyndromic hearing impairment is associated with a mutation in this gene. The observation that DFNA5 is epigenetically inactivated in a large number of cancers of frequent types (gastric, colorectal, and breast) is another important finding and is in line with its apoptosis-inducing properties. Indeed, if apoptosis is an intrinsic feature of DFNA5, shutting the gene down in tumor cells makes them more susceptible to uncontrolled cellular growth. Moreover, the fact that DFNA5 is regulated by P53 strongly suggests that DFNA5 is a tumor suppressor gene.

Immunogen: Recombinant protein within Human DFNA5 aa 34-214 / 496.

Positive control: Hela cell lysates, HepG2 cell lysates, SiHa, rat testis tissue, human skin tissue, human breast carcinoma tissue, human placenta tissue, SH-SY5Y.

Subcellular location: Cell membrane, cytosol.

Database links: SwissProt: O60443 Human

Recommended Dilutions:

WB	1:500-1:2,000
IF-Cell	1:50-1:100
IHC-P	1:50-1:200
FC	1:50-1:100

Storage Buffer: 1*PBS (pH7.4), 0.2% BSA, 50% Glycerol. Preservative: 0.05% Sodium Azide.

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

Purity: Protein G affinity purified.

Hangzhou Huaan Biotechnology Co., Ltd.

Orders:0086-571-88062880

Technical:0086-571-89986345

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Images

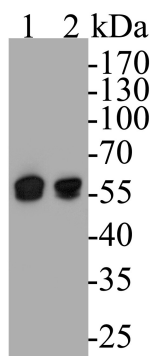


Fig1: Western blot analysis of DFNA5 / GSDME on different lysates. Proteins were transferred to a PVDF membrane and blocked with 5% BSA in PBS for 1 hour at room temperature. The primary antibody (EM1901-48, 1/500) was used in 5% BSA at room temperature for 2 hours. Goat Anti-Mouse IgG - HRP Secondary Antibody (HA1006) at 1:5,000 dilution was used for 1 hour at room temperature.

Positive control:

Lane 1: Hela cell lysate

Lane 2: HepG2 cell lysate

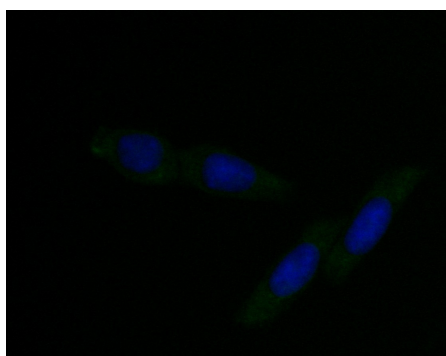


Fig2: ICC staining of DFNA5 / GSDME in SiHa cells (green). Formalin fixed cells were permeabilized with 0.1% Triton X-100 in TBS for 10 minutes at room temperature and blocked with 1% Blocker BSA for 15 minutes at room temperature. Cells were probed with the primary antibody (EM1901-48, 1/50) for 1 hour at room temperature, washed with PBS. Alexa Fluor®488 Goat anti-Mouse IgG was used as the secondary antibody at 1/1,000 dilution. The nuclear counter stain is DAPI (blue).

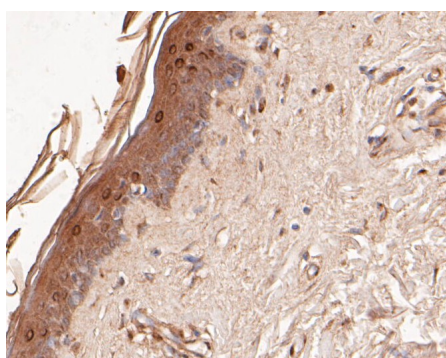


Fig3: Immunohistochemical analysis of paraffin-embedded human skin tissue using anti-DFNA5 / GSDME antibody. The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 8.0-8.4) for 20 minutes. The tissues were blocked in 5% BSA for 30 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (EM1901-48, 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.

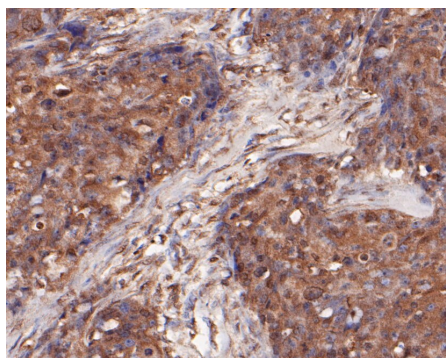


Fig4: Immunohistochemical analysis of paraffin-embedded human breast carcinoma tissue using anti-DFNA5 / GSDME antibody. The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 8.0-8.4) for 20 minutes. The tissues were blocked in 5% BSA for 30 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (EM1901-48, 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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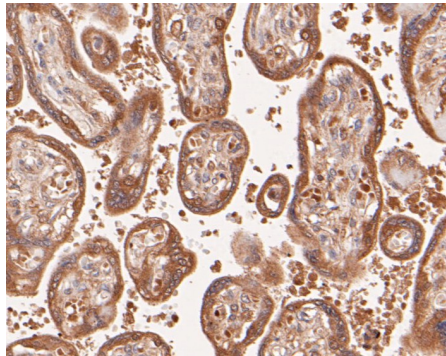


Fig5: Immunohistochemical analysis of paraffin-embedded human placenta tissue using anti-DFNA5 / GSDME antibody. The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 8.0-8.4) for 20 minutes. The tissues were blocked in 5% BSA for 30 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (EM1901-48, 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.

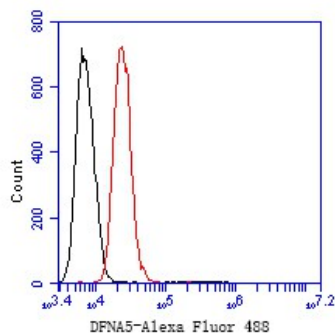


Fig6: Flow cytometric analysis of DFNA5 / GSDME was done on SH-SY5Y cells. The cells were fixed, permeabilized and stained with the primary antibody (EM1901-48, 1/50) (red). After incubation of the primary antibody at room temperature for an hour, the cells were stained with a Alexa Fluor 488-conjugated Goat anti-Mouse IgG Secondary antibody at 1/1000 dilution for 30 minutes. 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. Rogers C. et. al. Cleavage of DFNA5 by caspase-3 during apoptosis mediates progression to secondary necrotic/pyroptotic cell death. *Nat. Commun.* 8:14128-14128(2017).
2. Van Rossom S. et. al. The deafness gene DFNA5 induces programmed cell death through mitochondria and MAPK-related pathways. *Front. Cell. Neurosci.* 9:231-231(2015).

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