Human Metadherin Antibody

Antigen Affinity-purified Polyclonal Sheep IgG

Catalog Number: AF7644

DESCRIPTION

Species Reactivity	Human
Specificity	Detects human Metadherin in direct ELISAs and Western blots. In direct ELISAs, 100% cross-reactivity with recombinant mouse Metadherin is observed.
Source	Polyclonal Sheep IgG
Purification	Antigen Affinity-purified
Immunogen	*E. coli*-derived recombinant human Metadherin Lys168-Ser298 Accession # Q86UE4
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 as a 0.2 μm filtered solution in PBS.

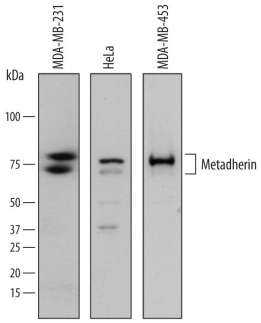
APPLICATIONS

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	0.2 µg/mL	See Below
Immunocytochemistry	5-15 µg/mL	See Below

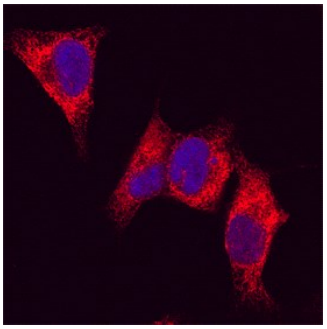
DATA

Western Blot



Detection of Human Metadherin by Western Blot. Western blot shows lysates of MDA-MB-231 human breast cancer cell line, HeLa human cervical epithelial carcinoma cell line, and MDA-MB-453 human breast cancer cell line. PVDF membrane was probed with 0.2 µg/mL of Sheep Anti-Human Metadherin Antigen Affinity-purified Polyclonal Antibody (Catalog # AF7644) followed by HRP-conjugated Anti-Sheep IgG Secondary Antibody (Catalog # HAF016). Specific bands were detected for Metadherin at approximately 80 and 75 kDa (as indicated). This experiment was conducted under reducing conditions and using Immunoblot Buffer Group 1.

Immunocytochemistry



Metadherin in HeLa Human Cell Line. Metadherin was detected in immersion fixed HeLa human cervical epithelial carcinoma cell line using Sheep Anti-Human Metadherin Antigen Affinity-purified Polyclonal Antibody (Catalog # AF7644) at 5 µg/mL for 3 hours at room temperature. Cells were stained using the NorthernLights™ 557-conjugated Anti-Sheep IgG Secondary Antibody (red; Catalog # NL010) and counterstained with DAPI (blue). Specific staining was localized to cytoplasm. View our protocol for Fluorescent ICC Staining of Cells on Coverslips.

PREPARATION AND STORAGE

Reconstitution	Sterile PBS to a final concentration of 0.2 mg/mL.
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.** ● 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

MTDH (Metadherin; also LYRIC and Astrocyte Elevated Gene product-1/AEG1) is an 80 kDa membrane-bound protein that belongs to no known structural protein family. It has restricted expression, being found in breast ductal epithelium, striated muscle, select neurons and astrocytes, as well as multiple tumor types of diverse origin. MTDH is considered a mediator of Myc and Ha-Ras, and its activity is associated with stimulation of the β-catenin/Wnt and NFkB signaling pathways. It is also believed to contribute to tight junction formation via an interaction with ZO-1. MTDH is also found in the nucleus where it complexes with CBP and p65. Human MTDH is 582 amino acids (aa) in length. It appears to be a type III transmembrane protein (N-terminus luminal/extracellular with no signal sequence) that is found embedded in both the plasma membrane and ER. It contains an N-terminal luminal region (aa 1-48), plus a large 513 aa cytoplasmic domain (aa 70-582) that possesses at least 11 utilized Ser/Thr phosphorylation sites. There is the potential for ubiquitination, SUMOylation, acetylation and proteolytic processing, and this may account for multiple bands in SDS-PAGE that represent MWs of 20, 35, 50-55, 70-75, and 86 kDa. Over aa 169-298, human MTDH shares 97% aa identity with mouse MDTH.