Human ECM1 Antibody

Antigen Affinity-purified Polyclonal Sheep IgG
Catalog Number: AF3937

DESCRIPTION

Species Reactivity	Human
Specificity	Detects human ECM-1 in direct ELISAs and Western blots. In direct ELISAs, less than 10% cross-reactivity with recombinant mouse ECM-1 is observed.
Source	Polyclonal Sheep IgG
Purification	Antigen Affinity-purified
Immunogen	Mouse myeloma cell line NS0-derived recombinant human ECM-1 Ala20-Glu540 Accession # AAH23505
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.

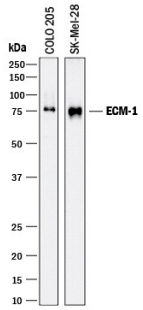
APPLICATIONS

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	1 µg/mL	See Below
Immunohistochemistry	5-15 µg/mL	See Below
Simple Western	10 µg/mL	See Below

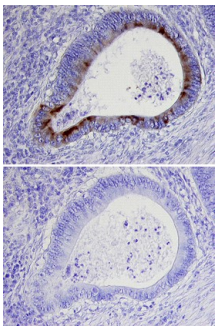
DATA

Western Blot



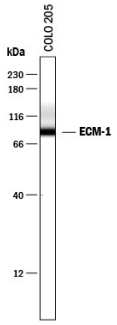
Detection of Human ECM-1 by Western Blot. Western blot shows lyates of COLO 205 human colorectal adenocarcinoma cell line and SK-MeI-28 human malignant melanoma cell line. PVDF membrane was probed with 1 µg/mL of Sheep Anti-Human ECM-1 Antigen Affinity-purified Polyclonal Antibody (Catalog # AF3937) followed by HRP-conjugated Anti-Sheep IgG Secondary Antibody (Catalog # HAF016). A specific band was detected for ECM-1 at approximately 75 kDa (as indicated). This experiment was conducted under reducing conditions and using Immunoblot Buffer Group 1.

Immunohistochemistry



ECM-1 in Human Colon Cancer Tissue. ECM-1 was detected in immersion fixed paraffin-embedded sections of human colon cancer tissue using 15 µg/mL Sheep Anti-Human ECM-1 Antigen Affinity-purified Polyclonal Antibody (Catalog # AF3937) overnight at 4 °C. Tissue was stained with the Anti-Sheep HRP-DAB Cell & Tissue Staining Kit (brown; Catalog # CTS019) and counterstained with hematoxylin (blue). Specific labeling was localized to the plasma membrane of epithelial cells. Lower panel shows a lack of labeling if primary antibodies are omitted and tissue is stained only with secondary antibody followed by incubation with detection reagents. View our protocol for Chromogenic IHC Staining of Paraffin-embedded Tissue Sections.

Simple Western



Detection of Human ECM-1 by Simple Western™. Simple Western lane view shows lyates of COLO 205 human colorectal adenocarcinoma cell line, loaded at 0.2 mg/mL. A specific band was detected for ECM-1 at approximately 90 kDa (as indicated) using 10 µg/mL of Sheep Anti-Human ECM-1 Antigen Affinity-purified Polyclonal Antibody (Catalog # AF3937) followed by 1:50 dilution of HRP-conjugated Anti-Sheep IgG Secondary Antibody (Catalog # HAF016). This experiment was conducted under reducing conditions and using the 12-230 kDa separation system.

PREPARATION AND STORAGE

Reconstitution	Reconstitute at 0.2 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.** ● 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.

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BACKGROUND

Extracellular matrix protein-1 (ECM-1) is an 85 kDa, secreted glycoprotein important in connective tissue organization (1-3). Of three identified splice variants the 540 amino acid (aa) form, ECM-1a, is the most widely expressed, with the highest expression in the placenta and heart (2). ECM-1b (415 aa) is found only in tonsil and associated with suprabasal keratinocytes (2, 4). Since ECM-1b expression is differentiation-dependent, a role in terminal keratinocyte differentiation has been suggested (4). ECM-1c (559 aa) accounts for approximately 15% of skin ECM-1 (5). Human ECM-1a contains a 19 aa signal peptide and a 521 aa secreted portion that includes an N-terminal proline-rich, cysteine-free region, two tandem repeat domains, and a C-terminal domain. There are six repeats of a $CC(X_{7-10})C$ motif (x = any aa) within the tandem repeat and C-terminal domains. These motifs are involved in ligand binding to members of the albumin family, and are expected to form two (in ECM-1b) or three (in ECM-1a) “double loop” structures (2). Mature human ECM-1a shows 69%, 71%, 72%, and 76% aa identity with corresponding isoforms of mouse, rat, canine, and bovine ECM-1, respectively. ECM-1 is over-expressed in many malignant epithelial tumors and has demonstrated angiogenic activity (6, 7). A variety of ECM-1 mutations, mainly within the first tandem repeat, are considered causative of lipoid proteinosis, a condition showing thickened and irregular extracellular matrix within connective tissue (8). In the autoimmune condition lichen sclerosis, auto-antibodies mainly recognize the second tandem repeat or the C-terminus of ECM-1 (9). These domains also bind the extracellular matrix molecules fibulin-1 and perlecan (5, 10). The phenotypes of lipoid proteinosis and lichen sclerosis support a role for ECM-1 as a “biological glue” in the dermis (1).

References:

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