# biotechne<sup>®</sup> RDSYSTEMS

## Human Nidogen-1/Entactin Antibody

Monoclonal Mouse IgG<sub>1</sub> Clone # 302117 Catalog Number: MAB2570

DESCRIPTION	
Species Reactivity	Human
Specificity	Detects human Nidogen-1/Entactin in ELISAs and Western blots. In direct ELISAs and Western blots, no cross-reactivity with recombinant human Nidogen-2 is observed.
Source	Monoclonal Mouse IgG <sub>1</sub> Clone # 302117
Purification	Protein A or G purified from hybridoma culture supernatant
Immunogen	Mouse myeloma cell line NS0-derived recombinant human Nidogen-1/Entactin Leu29-Lys1114 (Gln1113Arg) Accession # AAH45606.1
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	Recombinant Human Nidogen-1/Entactin (Catalog # 2570-ND)
Immunohistochemistry	3-25 μg/mL	Immersion fixed paraffin-embedded sections of human heart
Human Nidogen-1/Entactin Sandwich Immund	oassay	Reagent
ELISA Capture	2-8 µg/mL	Human Nidogen-1/Entactin Antibody (Catalog # MAB2570)
ELISA Detection	0.1-0.4 µg/mL	Human Nidogen-1/Entactin Biotinylated Antibody (Catalog # BAF2570)
Standard		Recombinant Human Nidogen-1/Entactin (Catalog # 2570-ND)
Immunocytochemistry	3-25 μg/mL	Immersion fixed human mesenchymal stem cells differentiated into chondrocytes

### DATA

#### Immunohistochemistry



Detection of Nidogen-1/Entactin in Human Heart. Nidogen-1/Entactin was detected in immersion fixed paraffinembedded sections of human heart using Mouse Anti-Human Nidogen-1/Entactin Monoclonal Antibody (Catalog # MAB2570) at 5 µg/ml for 1 hour at room temperature followed by incubation with the HRP-conjugated Anti-Mouse IgG Secondary Antibody (Catalog # HAF007) or the Anti-Mouse IgG VisUCyte™ HRP Polymer Antibody (Catalog # VC001). Before incubation with the primary antibody, tissue was subjected to heat-induced epitope retrieval using VisUCyte Antigen Retrieval Reagent-Basic (Catalog # VCTS021). Tissue was stained using DAB (brown) and counterstained with hematoxylin (blue). Specific staining was localized to the membrane. View our protocol for Chromogenic IHC Staining of Paraffin-embedded Tissue Sections.





Nidogen-1/Entactin in Human Chondrocytes. Nidogen-1/Entactin was detected in immersion fixed human mesenchymal stem cells differentiated into chondrocytes using Mouse Anti-Human Nidogen-1/Entactin Monoclonal Antibody (Catalog # MAB2570) at 10 µg/mL for 3 hours at room temperature. Cells were stained using the NorthernLights™ 557conjugated Anti-Mouse IgG Secondary Antibody (yellow; Catalog # NL007) and counterstained with DAPI (blue). View our protocol for Fluorescent ICC Staining of Cells on Coverslips.

PREPARATION AND S	STORAGE			
Reconstitution	Reconstitute at 0.5 mg/mL in sterile PBS. For liquid material, refer to CoA for concentration.			
Shipping	Lyophilized product is shipped at ambient temperature. Liquid small pack size (-SP) is shipped with polar packs. Upon receipt, store immediately at the temperature recommended below.			
Stability & Storage	<ul> <li>Use a manual defrost freezer and avoid repeated freeze-thaw cycles.</li> <li>12 months from date of receipt, -20 to -70 °C as supplied.</li> <li>1 month, 2 to 8 °C under sterile conditions after reconstitution.</li> <li>6 months, -20 to -70 °C under sterile conditions after reconstitution.</li> </ul>			
Rev. 7/2/2024 Pa	ge 1 of 2			
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#### BACKGROUND

Nidogen-1 (also entactin) is a 150 kDa, secreted, monomeric glycoprotein that serves as a major linking component of basement membranes (1-4). It is synthesized as a 1247 amino acid (aa) precursor with a 28 as signal sequence and a 1219 aa mature protein. The molecule is modular in structure with five distinct regions. There are three globular domains (G1-3) separated by a mucin region and an extended rod-shaped segment (5-7). The N-terminal globular domain (G1) is 200 aa in length and seemingly unrelated to any known motif (8). The mucin region is nearly 160 aa in length and presumably O-glycosylated (2, 8). G2 and G3 are both approximately 300 aa in length. G2 is described as a Nidogen ( $\beta$ -barrel) domain, while C-terminal G3 assumes a  $\beta$ -propeller configuration (1). The 250 aa rod-shaped segment has multiple EGF-like motifs and two thyroglobulin type 1 domains. Functionally, G1 is reported to bind type IV collagen (2, 7). The mucin region contains a short peptide that ligates  $\alpha_3\beta_1$  integrins (9, 10). G2 interacts with perlecan, and an RGD motif in the rod-shaped segment serves as a binding site for  $\alpha_k\beta_3$  integrins (9, 10). Finally, G3 is associated with laminin binding (2, 7). As a full-length molecule, the multiple extracellular matrix-binding sites of Nidogen-1 are well positioned to serve as anchor sites for basement membrane molecules. Nidogen-1 also undergoes proteolytic processing by at least two MMPs, MMP-7 and MMP-19 (10, 11). While this destroys the integrity of Nidogen-associated matrices, it also generates peptide fragments that are capable of inducing neutrophil chemotaxis and phagocytosis (10). Nidogen-2 is related to Nidogen-1 ( $\approx$  50% aa identity) and shares many of the same adhesive properties as Nidogen-1 (12). Both bind perlecan plus collagens I and IV. Nidogen-2, however, does not bind fibulin-1 or 2, and shows only modest interaction with laminin. Thus, although coexpressed, Nidogen-2 serves as only a partial substitute for Nidogen-1 (2, 12). Human Nidogen-1 shares 85% aa sequence

#### References:

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