

DESCRIPTION

Source Chinese Hamster Ovary cell line, CHO-derived
Leu31-Lys1375 (Gly832Ala), with an N-terminal 9-His tag
Accession # Q14112

N-terminal Sequence Analysis His

Predicted Molecular Mass 149.4 kDa

SPECIFICATIONS

SDS-PAGE 140-190 kDa, reducing conditions

Activity Measured by the ability of the immobilized protein to support the adhesion of SVEC4-10 mouse vascular endothelial cells.
When 4×10^4 cells/well are added to Recombinant Human Nidogen-2 coated plates (30 $\mu\text{g}/\text{mL}$ with 100 $\mu\text{L}/\text{well}$), approximately 30-50% will adhere after one hour at 37° C.
Optimal dilutions should be determined by each laboratory for each application.

Endotoxin Level <1.0 EU per 1 μg of the protein by the LAL method.

Purity >90%, by SDS-PAGE under reducing conditions and visualized by silver stain.

Formulation Lyophilized from a 0.2 μm filtered solution in PBS. See Certificate of Analysis for details.

PREPARATION AND STORAGE

Reconstitution Reconstitute at 100 $\mu\text{g}/\text{mL}$ in sterile PBS.

Shipping The product is shipped at ambient temperature. Upon receipt, store it immediately at the temperature recommended below.

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.
- 3 months, -20 to -70 °C under sterile conditions after reconstitution.

BACKGROUND

Nidogen-2 (also named entactin-2) is a 200 kDa, secreted, monomeric basement membrane glycoprotein (1). Nidogens-1 and 2 are expressed in nearly all basement membranes (1-3) where they interact with laminins, collagen type IV and proteoglycan family members to form structural scaffolds (4, 5). In mouse, Nidogens 1 and 2 appear to substitute for each other. Deletion of one nidogen gives a mild phenotype, but deletion of both nidogens is lethal (6, 7). Affinity of laminin binding is much lower for human Nidogen-2 than that of mouse Nidogen-2, indicating that human Nidogen-2 may not be a strict substitute for Nidogen-1 (1). Both nidogens bind perlecan and collagens I and IV, but only Nidogen-1 binds fibulins (1, 3). The two nidogens show approximately 50% amino acid (aa) identity in human and are structurally similar (1, 4, 6). Cleavage of a 28 aa signal sequence from human Nidogen-2 produces a 1219 aa mature protein containing three globular domains (G1-3) separated by a link region and an extended rod-shaped segment. The G1 domain is reported to bind type IV collagen, the G2 Nidogen (β -barrel) domain interacts with perlecan, and the C-terminal G3 β -propeller structure is associated with laminin binding. The mucin-like link region is longer in Nidogen-2 than nidogen-1, and contains both N- and O-glycosylation (2, 8). There is one EGF-like motif and a short peptide that ligates $\alpha_3\beta_1$ integrins. The rod-shaped segment contains four additional EGF-like motifs, two of which bind calcium, and two thyroglobulin type 1 domains that serve as a binding site for $\alpha_3\beta_3$ integrins. Mature human Nidogen-2 is 80% aa identical to both mouse and rat Nidogen-2, and 73% aa identical to both canine and bovine Nidogen-2.

References:

1. Kohfeldt, K. *et al.* (1998) *J. Mol. Biol.* **282**:99.
2. Miosge, N. *et al.* (2001) *Histochem. J.* **33**:523.
3. Salmivirta, K. *et al.* (2002) *Exp. Cell Res.* **279**:188.
4. Hohenester, E. and J. Engel (2002) *Matrix Biol.* **21**:115.
5. Charonis, A. *et al.* (2005) *Curr. Med. Chem.* **12**:1495.
6. Schymeinsky, J. *et al.* (2002) *Mol. Cell. Biol.* **22**:6820.
7. Bader, B.L. *et al.* (2005) *Mol. Cell. Biol.* **25**:6846.