

## DESCRIPTION

<b>Species Reactivity</b>	Human
<b>Specificity</b>	Detects human Laminin $\alpha 4$ in direct ELISAs and Western blots. In direct ELISAs, approximately 15% cross-reactivity with recombinant mouse Laminin $\alpha 4$ is observed.
<b>Source</b>	Polyclonal Sheep IgG
<b>Purification</b>	Antigen Affinity-purified
<b>Immunogen</b>	Chinese hamster ovary cell line CHO-derived recombinant human Laminin $\alpha 4$ Gln826-Ala1816, predicted Accession # EAW48267
<b>Endotoxin Level</b>	<0.10 EU per 1 $\mu$ g of the antibody by the LAL method.
<b>Formulation</b>	Lyophilized from a 0.2 $\mu$ m 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 $\mu$ m filtered solution in PBS.

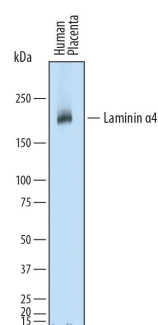
## APPLICATIONS

**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
<b>Western Blot</b>	2 $\mu$ g/mL	See Below
<b>Immunocytochemistry</b>	5-15 $\mu$ g/mL	See Below
<b>Immunohistochemistry</b>	3-25 $\mu$ g/mL	See Below
<b>Simple Western</b>	20 $\mu$ g/mL	See Below
<b>Neutralization</b>	Measured by its ability to neutralize Laminin $\alpha 4$ -induced adhesion in the HT1080 human fibrosarcoma cell line. The Neutralization Dose (ND <sub>50</sub> ) is typically 1.5-7.5 $\mu$ g/mL in the presence of 5 $\mu$ g/mL Recombinant Human Laminin $\alpha 4$ .	

## DATA

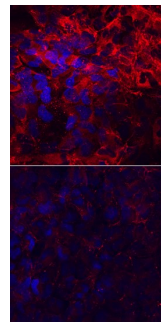
### Western Blot



#### Detection of Human Laminin $\alpha 4$ by Western Blot.

Western blot shows lysates of human placenta tissue. PVDF membrane was probed with 2  $\mu$ g/mL of Sheep Anti-Human Laminin  $\alpha 4$  Antigen Affinity-purified Polyclonal Antibody (Catalog # AF7340) followed by HRP-conjugated Anti-Sheep IgG Secondary Antibody (Catalog # HAF016). A specific band was detected for Laminin  $\alpha 4$  at approximately 200-220 kDa (as indicated). This experiment was conducted under reducing conditions and using [Immunoblot Buffer Group 1](#).

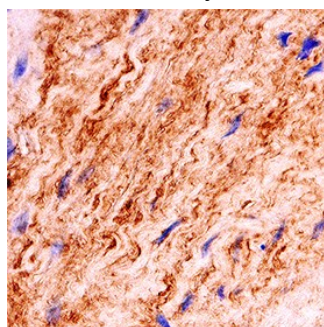
### Immunocytochemistry



#### Laminin $\alpha 4$ in T98G Human Cell Line.

Laminin  $\alpha 4$  was detected in immersion fixed T98G human glioblastoma cell line, induced (upper panel) or not (lower panel) with StemXVivo EMT Inducing Media Supplement (Catalog # CCM017), using Sheep Anti-Human Laminin  $\alpha 4$  Antigen Affinity-purified Polyclonal Antibody (Catalog # AF7340) at 10  $\mu$ g/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 and cell surfaces. View our protocol for [Fluorescent ICC Staining of Cells on Coverslips](#).

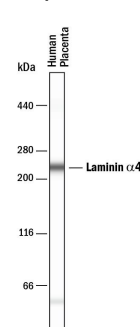
### Immunohistochemistry



#### Laminin $\alpha 4$ in Human Placenta.

Laminin  $\alpha 4$  was detected in immersion fixed paraffin-embedded sections of human placenta using Sheep Anti-Human Laminin  $\alpha 4$  Antigen Affinity-purified Polyclonal Antibody (Catalog # AF7340) at 3  $\mu$ g/mL for 1 hour at room temperature followed by incubation with the Anti-Sheep IgG VisUCyte™ HRP Polymer Antibody (Catalog # VC006). Before incubation with the primary antibody, tissue was subjected to heat-induced epitope retrieval using Antigen Retrieval Reagent-Basic (Catalog # CTS013). Tissue was stained using DAB (brown) and counterstained with hematoxylin (blue). Specific staining was localized to Golgi apparatus. View our protocol for [IHC Staining with VisUCyte HRP Polymer Detection Reagents](#).

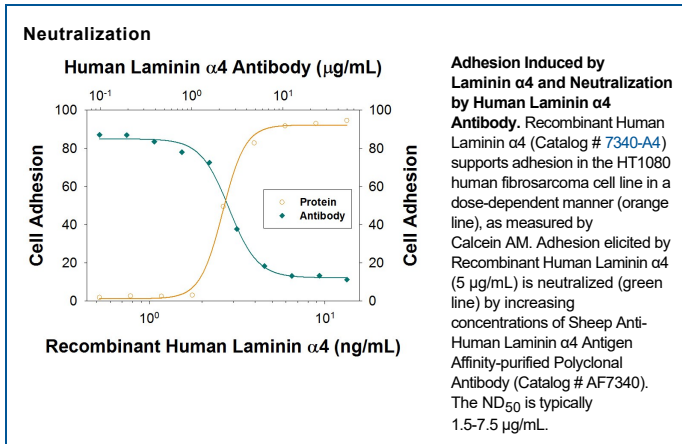
### Simple Western



#### Detection of Human Laminin $\alpha 4$ by Simple Western™.

Simple Western lane view shows lysates of human placenta tissue, loaded at 0.2 mg/mL. A specific band was detected for Laminin  $\alpha 4$  at approximately 233 kDa (as indicated) using 20  $\mu$ g/mL of Sheep Anti-Human Laminin  $\alpha 4$  Antigen Affinity-purified Polyclonal Antibody (Catalog # AF7340) 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 66-440 kDa separation system.





## PREPARATION AND STORAGE

<b>Reconstitution</b>	Sterile PBS to a final concentration of 0.2 mg/mL.
<b>Shipping</b>	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
<b>Stability &amp; Storage</b>	<p><b>Use a manual defrost freezer and avoid repeated freeze-thaw cycles.</b></p> <ul style="list-style-type: none"> <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>

## BACKGROUND

Laminin  $\alpha 4$  (LAMA-4 [Laminin subunit alpha 4]; also Laminin-14/423, -8/411 and -9/421 subunit alpha) is a 180-230 kDa secreted member of the laminin family of molecules. It is found in the basement membranes of adipocytes, endothelial cells, cardiac and visceral smooth muscle cells, fibroblasts, keratinocytes, and pancreatic acinar cells. Laminin is a heterotrimeric glycoprotein that attaches cells to basement membranes. It is composed of covalently-linked  $\alpha$ -,  $\beta$ - and  $\gamma$ -chains, and is known to attach cells to basement membranes. In the case of the  $\alpha$ -subunit, this is mediated through binding to multiple integrins ( $\alpha 3\beta 1$ ,  $\alpha \nu\beta 3$  and  $\alpha 6\beta 1$ ), plus dystroglycan and the syndecans-2 and -4. Mature human Laminin  $\alpha 4$  is 1799 amino acids (aa) in length (aa 25-1823). It contains multiple domains, including four EGF-like domains (aa 82-255) and five Laminin G-like domains (aa 833-1820). There is a chondroitin sulfate attachment at the N-terminus that accounts for 20-30 kDa of MW. Laminin  $\alpha 4$  is cleaved between the Laminin G-like 3 and 4 domains, creating a soluble 44 kDa fragment that possesses antibacterial activity. There are two potential splice variants, one that shows a deletion of aa 266-272, and another that contains a 55 aa substitution for aa 66-1823. Over aa 826-1816, human Laminin  $\alpha 4$  shares 91% aa sequence identity with mouse Laminin  $\alpha 4$ .