

# Biotin Conjugated Anti-Human CSPG4 Antibody [PSH10-79] - Detector

## HA723247B



<b>Product Type:</b>	Recombinant Rabbit monoclonal IgG, primary antibodies
<b>Species reactivity:</b>	Human
<b>Applications:</b>	ELISA(Det), ELISA
<b>Clone number:</b>	PSH10-79

**Description:** Proteoglycan playing a role in cell proliferation and migration which stimulates endothelial cells motility during microvascular morphogenesis. May also inhibit neurite outgrowth and growth cone collapse during axon regeneration. Cell surface receptor for collagen alpha 2(VI) which may confer cells ability to migrate on that substrate. Binds through its extracellular N-terminus growth factors, extracellular matrix proteases modulating their activity. May regulate MMP16-dependent degradation and invasion of type I collagen participating in melanoma cells invasion properties. May modulate the plasminogen system by enhancing plasminogen activation and inhibiting angiostatin. Functions also as a signal transducing protein by binding through its cytoplasmic C-terminus scaffolding and signaling proteins. May promote retraction fiber formation and cell polarization through Rho GTPase activation. May stimulate alpha-4, beta-1 integrin-mediated adhesion and spreading by recruiting and activating a signaling cascade through CDC42, ACK1 and BCAR1. May activate FAK and ERK1/ERK2 signaling cascades.

<b>Conjugate:</b>	Biotin-conjugated
<b>Immunogen:</b>	Recombinant protein within Human CSPG4 aa 1356-2224 (HA211047).
<b>Positive control:</b>	Recombinant Human CSPG4 protein (HA211047).
<b>Subcellular location:</b>	Cell membrane.
<b>Database links:</b>	SwissProt: Q6UVK1 Human

**Recommended Dilutions:**  
**ELISA(Det)** Use at an assay dependent concentration. Can be paired for Sandwich ELISA with Rabbit monoclonal [PSH10-77] to Human CSPG4 antibody (Capture) (HA723244) or Rabbit monoclonal [PSH10-78] to Human CSPG4 antibody (Capture) (HA723245) and Recombinant Human CSPG4 protein (HA211047) as the standard. The reference range value is 156-20,000 pg/ml.  
**ELISA** Use at an assay dependent concentration.

<b>Storage Buffer:</b>	PBS (pH7.4), 0.1% BSA, 40% Glycerol. Preservative: 0.05% ProClin300.
<b>Storage Instruction:</b>	Shipped at 4°C. Store at +4°C short term (1-2 weeks). It is recommended to aliquot into single-use upon delivery. Store at -20°C long term.
<b>Purity:</b>	Protein A affinity purified.

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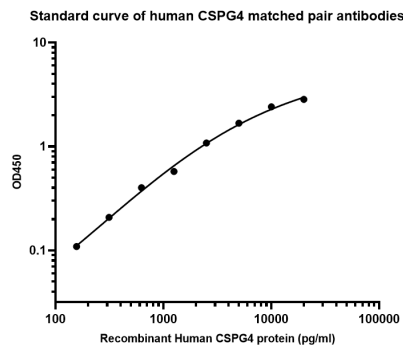
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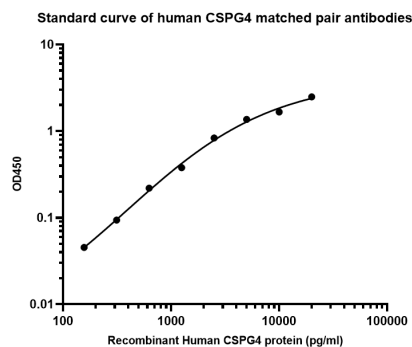
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## Images



**Fig1:** Sandwich ELISA analysis of Human CSPG4 matched pair antibodies

Elisa assay was performed by coating wells of a 96-well plate with 50  $\mu$ l per well of capture antibody (HA723244) diluted in carbonate/bicarbonate buffer, at a concentration of 5  $\mu$ g/mL overnight at 4°C. Wells of the plate were washed, blocked with 150  $\mu$ l 0.05% tween-20 1% BSA blocking buffer, and incubated with serial diluted Recombinant Human CSPG4 protein (HA211047) starting from 20,000 pg/ml to 0 pg/ml and detect antibody (HA723247B, 0.2  $\mu$ g/ml) for 1 hour at 30°C with shaking. Then the plate was washed and incubated with 50  $\mu$ l per well of SA-HRP for 0.5 hour at 30°C with shaking. Detection was performed using an Ultra TMB Substrate for 10 minutes at room temperature in the dark. The reaction was stopped with sulfuric acid and absorbances were read on a spectrophotometer at 450 nm.



**Fig2:** Sandwich ELISA analysis of Human CSPG4 matched pair antibodies

Elisa assay was performed by coating wells of a 96-well plate with 50  $\mu$ l per well of capture antibody (HA723245) diluted in carbonate/bicarbonate buffer, at a concentration of 5  $\mu$ g/mL overnight at 4°C. Wells of the plate were washed, blocked with 150  $\mu$ l 0.05% tween-20 1% BSA blocking buffer, and incubated with serial diluted Recombinant Human CSPG4 protein (HA211047) starting from 20,000 pg/ml to 0 pg/ml and detect antibody (HA723247B, 0.2  $\mu$ g/ml) for 1 hour at 30°C with shaking. Then the plate was washed and incubated with 50  $\mu$ l per well of SA-HRP for 0.5 hour at 30°C with shaking. Detection was performed using an Ultra TMB Substrate for 10 minutes at room temperature in the dark. The reaction was stopped with sulfuric acid and absorbances were read on a spectrophotometer at 450 nm.

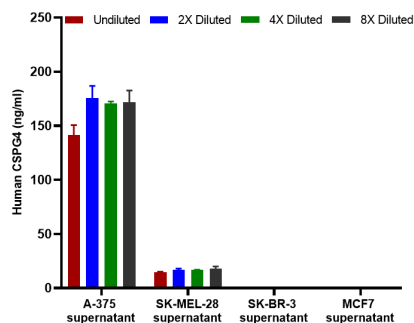
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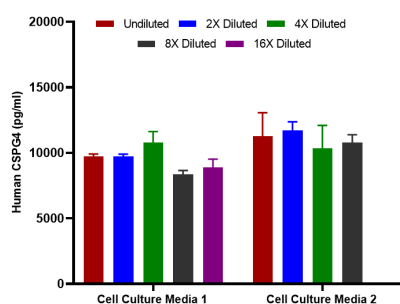
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**Fig3:** Interpolated concentrations of native CSPG4 in A-375, SK-MEL-28, SK-BR-3 and MCF7 cell culture supernatant.

The concentrations of CSPG4 were measured in duplicates, interpolated from the CSPG4 standard curve and corrected for sample dilution. Undiluted samples are A-375 cell culture supernatant 25%, SK-MEL-28 cell culture supernatant 100%, SK-BR-3 cell culture supernatant 100% and MCF7 cell culture supernatant 100%. The interpolated dilution factor corrected values are plotted (mean  $\pm$  SD, n=2). The mean CSPG4 concentration was determined to be 164.9 ng/ml in A-375 cell culture supernatant, 17.2 ng/ml in SK-MEL-28 cell culture supernatant and undetectable in SK-BR-3 and MCF7 cell culture supernatant.



**Fig4:** Interpolated concentrations of spiked CSPG4 in human cell culture media samples.

The concentrations of CSPG4 were measured in duplicates, interpolated from the CSPG4 standard curves and corrected for sample dilution. Undiluted samples are as follows: cell culture media 50%. The interpolated dilution factor corrected values are plotted (mean  $\pm$  SD, n=2).

**Note:** All products are "FOR RESEARCH USE ONLY AND ARE NOT INTENDED FOR DIAGNOSTIC OR THERAPEUTIC USE".

## Background References

1. Yang J., Price M.A., Neudauer C.L., Wilson C., Ferrone S., Xia H., Iida J., Simpson M.A., McCarthy J.B. Melanoma chondroitin sulfate proteoglycan enhances FAK and ERK activation by distinct mechanisms. *J. Cell Biol.* 165:881-891 (2004)
2. Iida J., Pei D., Kang T., Simpson M.A., Herlyn M., Furcht L.T., McCarthy J.B. Melanoma chondroitin sulfate proteoglycan regulates matrix metalloproteinase-dependent human melanoma invasion into type I collagen. *J. Biol. Chem.* 276:18786-18794 (2001)

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