Biotin Conjugated Anti-Human Osteopontin Antibody [PSH08-60] - Detector

HA723008B

Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human
Applications:	ELISA(Det), ELISA
Clone number:	PSH08-60

Description: Major non-collagenous bone protein that binds tightly to hydroxyapatite. Appears to form an integral part of the mineralized matrix. Probably important to cell-matrix interaction. Acts as a cytokine involved in enhancing production of interferon-gamma and interleukin-12 and reducing production of interleukin-10 and is essential in the pathway that leads to type I immunity. The protein encoded by this gene is involved in the attachment of osteoclasts to the mineralized bone matrix. The encoded protein is secreted and binds hydroxyapatite with high affinity. The osteoclast vitronectin receptor is found in the cell membrane and may be involved in the binding to this protein. This protein is also a cytokine that upregulates expression of interferon-gamma and interleukin-12. Several transcript variants encoding different isoforms have been found for this gene.

Conjugate: Biotin-conjugated

Immunogen: Recombinant protein within Human Osteopontin aa 17-314 (HA210993).

Positive control: Recombinant Human Osteopontin protein (HA210993).

Subcellular location: Secreted.

Database links: SwissProt: P10451 Human

Recommended Dilutions:

ELISA(Det)
ELISA

Use at an assay dependent concentration. Can be paired for Sandwich ELISA with Rabbit monoclonal [PSH08-58] to Human Osteopontin antibody (Capture) (HA723004) and recombinant Human Osteopontin protein (HA210993) as the standard. The reference range value is 61.7-5000 pg/ml.
Use at an assay dependent concentration.

Storage Buffer: PBS (pH7.4), 0.1% BSA, 40% Glycerol. Preservative: 0.05% ProClin300.

Storage Instruction: Shipped at 4℃. Store at +4℃ short term (1-2 weeks). It is recommended to aliquot into single-use upon delivery. Store at -20℃ long term.

Purity: Protein A affinity purified.

Images

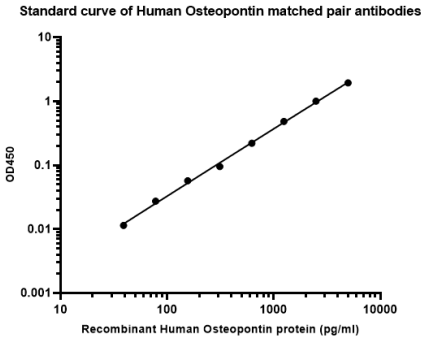

Fig1: Sandwich ELISA analysis of Human Osteopontin matched pair antibodies

Elisa assay was performed by coating wells of a 96-well plate with 100 µl per well of capture antibody (HA723004) diluted in carbonate/bicarbonate buffer, at a concentration of 5ug/ml overnight at 4℃. Wells of the plate were washed, blocked with 150 µl 0.05% tween-20 1%BSA blocking buffer, and incubated with serial diluted Recombinant Human Osteopontin protein (HA210993) starting from 5,000 pg/ml to 0 pg/ml and detect antibody (HA723008B, 0.2 µg/ml) for 1 hour at 30℃ with shaking. Then the plate was washed and incubated with 100 µl per well of SA-HRP for 0.5 hour at 30℃ 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.

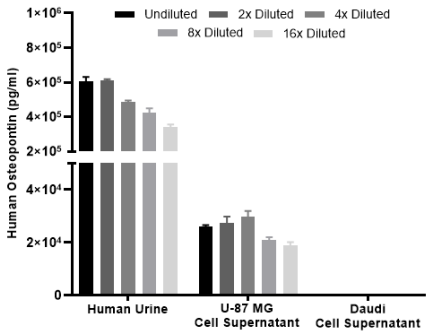

Fig2: Interpolated concentrations of native Osteopontin in human samples.

Interpolated concentration of native Osteopontin was measured in duplicate at different sample concentrations. The interpolated dilution factor corrected values were plotted (mean +/- SD, n=2). The mean Osteopontin concentration was determined to be 493 ng/mL in human urine, 25 ng/ml in U-87 MG cell culture supernatant. There was no detectable signal in Daudi cell supernatant.

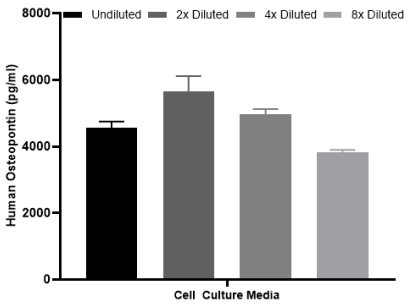

Fig3: Interpolated concentrations of spiked Osteopontin in cell culture media samples.

The concentrations of Osteopontin were measured in duplicates, interpolated from the Osteopontin 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 +/- SD, n=2).

Background References

1. Young M.F., Kerr J.M., Termine J.D., Wewer U.M., Wang M.G., McBride O.W., Fisher L.W. cDNA cloning, mRNA distribution and heterogeneity, chromosomal location, and RFLP analysis of human osteopontin (OPN). Genomics 7:491-502 (1990)
2. Kiefer M.C., Bauer D.M., Barr P.J. The cDNA and derived amino acid sequence for human osteopontin. Nucleic Acids Res. 17:3306-3306 (1989)