

Human Osteopontin ELISA Kit ab269374

Recombinant SimpleStep ELISA

9 Images

Overview

Product name Human Osteopontin ELISA Kit

Detection method Colorimetric

Precision

Intra-assay

Sample	n	Mean	SD	CV%
Serum	3			2.1%

Inter-assay

Sample	n	Mean	SD	CV%
Serum	8			2.8%

Sample type Cell culture supernatant, Milk, Urine, Serum, Hep Plasma, EDTA Plasma, Cit plasma, Cerebral Spinal Fluid

Assay type Sandwich (quantitative)

Sensitivity 88 pg/ml

Range 109.38 pg/ml - 7000 pg/ml

Recovery

Sample specific recovery

Sample type	Average %	Range
Cell culture supernatant	108	102% - 115%
Milk	96	93% - 98%
Urine	98	96% - 100%
Serum	102	99% - 104%
Hep Plasma	93	90% - 96%
EDTA Plasma	99	95% - 102%

Sample type	Average %	Range
Cit plasma	92	91% - 93%
Cerebral Spinal Fluid	86	82% - 93%

**Assay time**

1h 30m

**Assay duration**

One step assay

**Species reactivity**

**Reacts with:** Human

**Product overview**

Human Osteopontin ELISA Kit (ab269374) is a single-wash 90 min sandwich ELISA designed for the quantitative measurement of Osteopontin protein in edta plasma, hep plasma, milk, serum, urine, cit plasma, and cell culture supernatant. It uses our proprietary SimpleStep ELISA® technology. Quantitate Human Osteopontin with 88 pg/ml sensitivity.

SimpleStep ELISA® technology employs capture antibodies conjugated to an affinity tag that is recognized by the monoclonal antibody used to coat our SimpleStep ELISA® plates. This approach to sandwich ELISA allows the formation of the antibody-analyte sandwich complex in a single step, significantly reducing assay time. See the SimpleStep ELISA® protocol summary in the image section for further details. Our SimpleStep ELISA® technology provides several benefits:

- Single-wash protocol reduces assay time to 90 minutes or less
- High sensitivity, specificity and reproducibility from superior antibodies
- Fully validated in biological samples
- 96-wells plate breakable into 12 x 8 wells strips

A 384-well SimpleStep ELISA® microplate (**ab203359**) is available to use as an alternative to the 96-well microplate provided with SimpleStep ELISA® kits.

**Notes**

Osteopontin (OPN) is a 35kDa secreted adhesive glycoposphoprotein expressed mainly in bone, teeth, kidney, epithelial lining tissues, activated T cells, macrophages and breast milk. It is processed by matrix metalloproteases 3 and 4 as well as thrombin. It can be present as a cytokine-like soluble protein or as an immobilized extracellular matrix protein. It is important in multiple physiologic and pathologic processes including survival, adhesion, migration, angiogenesis, apoptosis and inflammation. High levels of Osteopontin is used as a biomarker of poor prognosis in patients with breast cancer, multiple myeloma and prostate cancer. Osteopontin has a role in the immune response as a chemotactic factor for macrophages, dendritic cells and T cells. It influences both cell-mediated and humoral immunity and Th1-cytokine functions as well as enhancing B lymphocytes proliferation and immunoglobulin production.

**Platform**

Pre-coated microplate (12 x 8 well strips)

**Properties**

**Storage instructions**

Store at +4°C. Please refer to protocols.

Components	1 x 96 tests
10X Human Osteopontin Capture Antibody	1 x 600µl

Components	1 x 96 tests
10X Human Osteopontin Detector Antibody	1 x 600µl
10X Wash Buffer PT (ab206977)	1 x 20ml
Antibody Diluent 4BI	1 x 6ml
Human Osteopontin Lyophilized Recombinant Protein	2 vials
Plate Seals	1 unit
Sample Diluent NS (ab193972)	1 x 12ml
SimpleStep Pre-Coated 96-Well Microplate (ab206978)	1 unit
Stop Solution	1 x 12ml
TMB Development Solution	1 x 12ml

## Function

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.

## Tissue specificity

Bone. Found in plasma.

## Sequence similarities

Belongs to the osteopontin family.

## Post-translational modifications

Extensively phosphorylated on clustered serine residues.

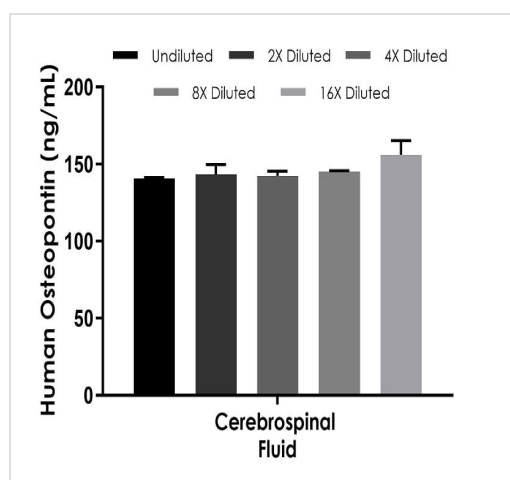
N- and O-glycosylated.

Phosphorylation sites are present in the extracellular medium.

## Cellular localization

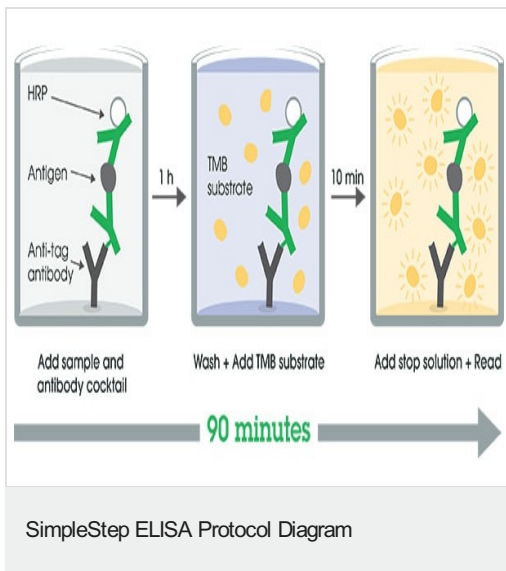
Secreted.

## Images

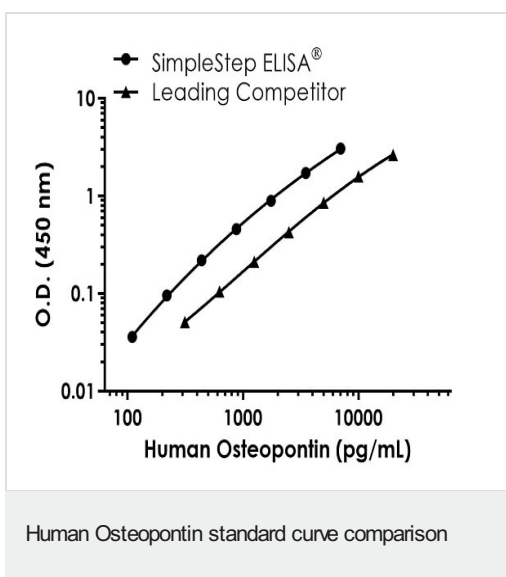


Interpolated concentrations of human Osteopontin in CSF.

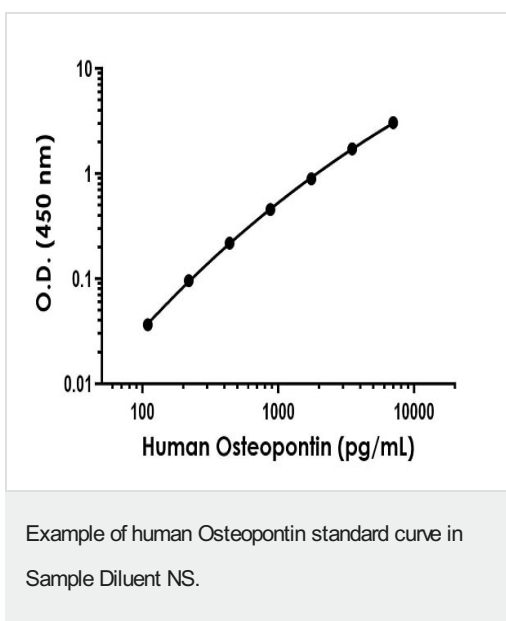
Interpolated concentration of native Osteopontin was measured in duplicate at different sample concentrations. Undiluted samples are as follows: CSF 2.5%. The interpolated dilution factor corrected values are plotted (mean  $\pm$  SD, n=2). Sample dilutions are made in Sample Diluent NS.



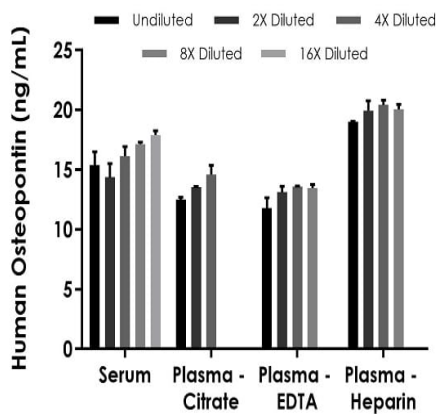
SimpleStep ELISA technology allows the formation of the antibody-antigen complex in one single step, reducing assay time to 90 minutes. Add samples or standards and antibody mix to wells all at once, incubate, wash, and add your final substrate. See protocol for a detailed step-by-step guide.



Standard Curve comparison between human Osteopontin SimpleStep ELISA kit and traditional ELISA kit from leading competitor. SimpleStep ELISA kit shows increased sensitivity.

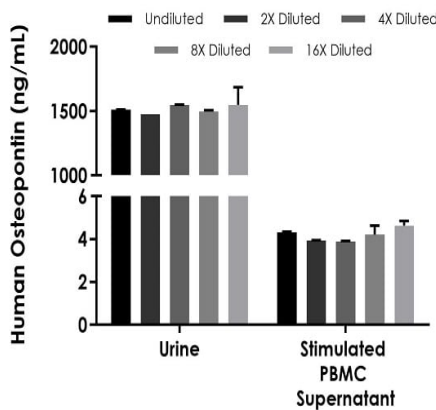


The Osteopontin standard curve was prepared as described in Section 10. Raw data values are shown in the table. Background-subtracted data values (mean  $\pm$  SD) are graphed.



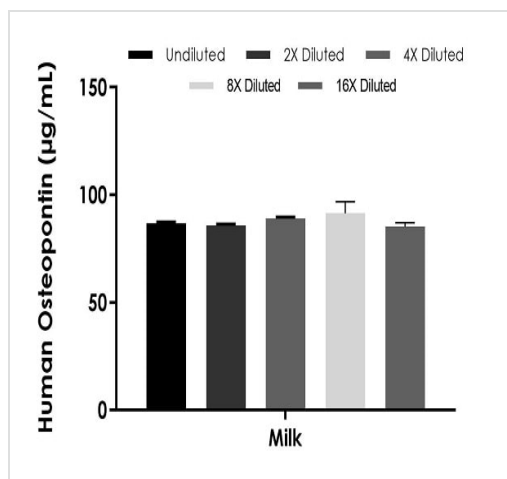
Interpolated concentrations of native Osteopontin in human serum and plasma samples.

The concentrations of Osteopontin were measured in duplicates, interpolated from the Osteopontin standard curves and corrected for sample dilution. Undiluted samples are as follows: serum 50%, plasma (citrate) 12.5%, plasma (EDTA) 12.5%, and plasma (heparin) 12.5%. The interpolated dilution factor corrected values are plotted (mean  $\pm$  SD,  $n=2$ ). The mean Osteopontin concentration was determined to be 16.2 ng/mL in serum, 13.5 ng/mL in plasma (citrate), 13.0 ng/mL in plasma (EDTA), and 19.9 ng/mL in plasma (heparin).



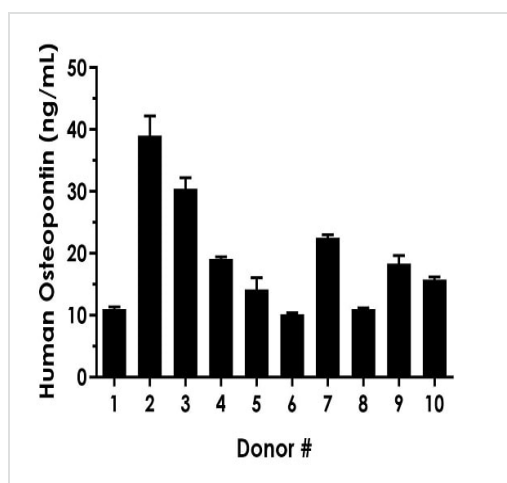
Interpolated concentrations of native Osteopontin in human urine and stimulated PBMC cell culture supernatant samples.

The concentrations of Osteopontin were measured in duplicates, interpolated from the Osteopontin standard curves and corrected for sample dilution. Undiluted samples are as follows: urine 1:400 and stimulated PBMC cell culture supernatant 50%. The interpolated dilution factor corrected values are plotted (mean  $\pm$  SD,  $n=2$ ). The mean Osteopontin concentration was determined to be 1,516 ng/mL in urine and 4.2 ng/mL in stimulated PBMC cell culture supernatant. No detectable signal was observed in unstimulated PBMC cell culture supernatant.



Interpolated concentrations of native Osteopontin in human milk 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: milk 1:20,000. The interpolated dilution factor corrected values are plotted (mean  $\pm$  SD,  $n=2$ ). The mean Osteopontin concentration was determined to be 88  $\mu\text{g/mL}$  in urine.



Serum from ten individual healthy human female donors was measured in duplicate

Interpolated dilution factor corrected values are plotted (mean  $\pm$  SD,  $n=2$ ). The mean Osteopontin concentration was determined to be 19 ng/mL with a range of 10 – 39 ng/mL.

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Sandwich ELISA - Human Osteopontin ELISA Kit  
(ab269374)

To learn more about the advantages of recombinant antibodies see [here](#).

**Please note:** All products are "FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES"

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