

QUANTITATIVE DETERMINATION OF HUMAN CARTILAGE OLIGOMERIC MATRIX PROTEIN (COMP) ELISA

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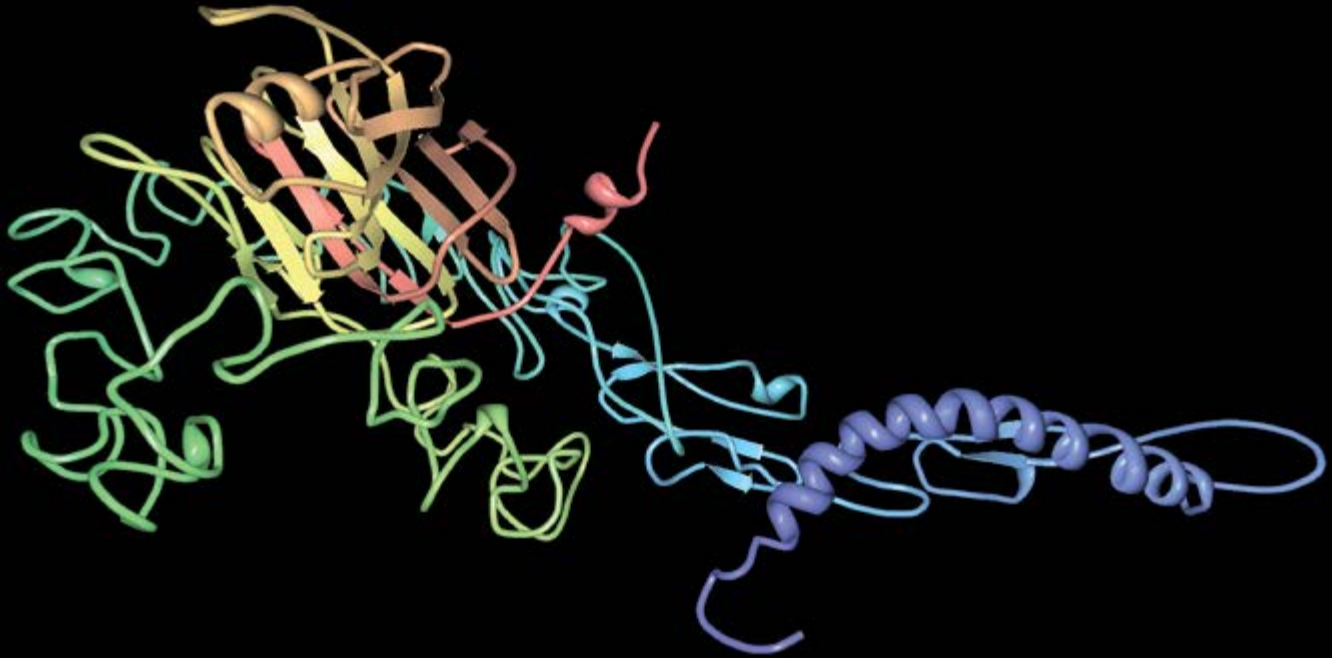
Human CARTILAGE OLIGOMERIC MATRIX PROTEIN (COMP) ELISA

- › High sensitivity (0.4 ng/ml)
- › Excellent analytical characteristics
- › Validated for human serum and plasma (EDTA, citrate, heparin)



**BONE AND CARTILAGE METABOLISM
JOINT DISEASES
OSTEOARTHRITIS
RHEUMATOID ARTHRITIS**

HUMAN CARTILAGE OLIGOMERIC MATRIX PROTEIN (COMP) ELISA



Introduction

Cartilage oligomeric matrix protein (COMP), also designated thrombospondin 5 (TSP 5), is a non-collagenous glycoprotein and is a member of the thrombospondin family of extracellular proteins. COMP is a calcium-binding protein of high molecular weight (>500kDa) present in the extracellular matrix of articular, nasal and tracheal cartilage. COMP is not only cartilage-derived but was found widely in other tissues, including synovium and tendon.

Intact COMP is pentameric, with five identical subunits and the carboxy-terminal globular domain of native COMP binds to collagens I, II, and IX. It has been proposed that COMP molecules are important for maintaining the properties and integrity of collagen network. In addition COMP may have a storage and delivery function for hydrophobic cell-signaling molecules such as vitamin D. The significance of COMP for normal development and function of cartilage has been

underscored by the discovery that mutations of the COMP gene result in pseudoachondroplasia and some forms of multiple epiphyseal dysplasia.

Most published studies have shown that serum levels of COMP provide important information about metabolic changes occurring in the cartilage matrix in joint disease. These studies describe that serum COMP level correlated with cartilage degradation and is a potential prognostic marker in inflammatory joint diseases such as osteoarthritis (OA) and rheumatoid arthritis (RA). Results have demonstrated an association of increasing serum COMP levels with progressive destruction of articular cartilage monitored radiographically. OA and RA are a common disease causing pain and disability in a significant proportion of the adult population and early diagnostics of these diseases is very important for future therapy.

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BioVendor Human COMP ELISA (RD194080200)

Intended use

The RD194080200 Human Cartilage Oligomeric Matrix Protein ELISA is a sandwich enzyme immunoassay for the quantitative measurement of human cartilage oligomeric matrix protein (COMP).

- The total assay time is less than 3.5 hours
- The kit measures COMP in serum and plasma (EDTA, citrate, heparin)
- Assay format is 96 wells
- Quality Controls are human serum based
- Standard is recombinant protein based
- Components of the kit are provided ready to use, concentrated or lyophilized

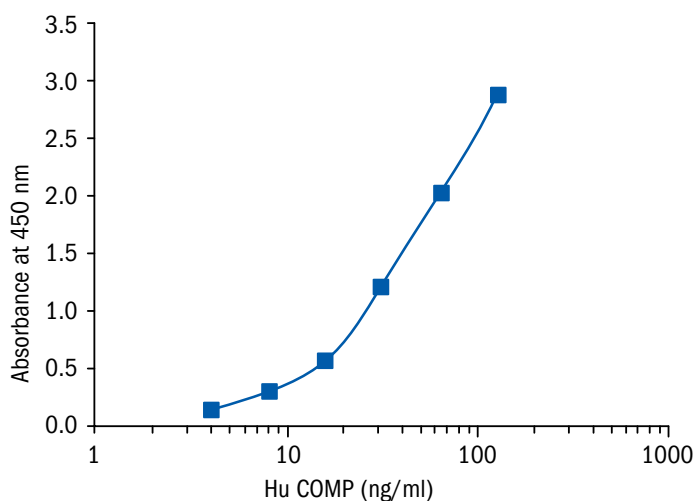
Clinical application

- Bone and cartilage metabolism
- Joint diseases
- Osteoarthritis
- Rheumatoid arthritis

Test principle

In the BioVendor Human Cartilage Oligomeric Matrix Protein ELISA, standards, quality controls and samples are incubated in microplate wells pre-coated with monoclonal anti-human COMP antibody. After 60 minutes incubation and washing, biotin labelled second monoclonal anti-human COMP antibody is added and incubated with captured COMP for 60 minutes. After another washing, streptavidin-HRP conjugate is added. After 30 minutes incubation and the last washing step, the remaining conjugate is allowed to react with the substrate solution (TMB). The reaction is stopped by addition of acidic solution and absorbance of the resulting yellow product is measured. The absorbance is proportional to the concentration of COMP. A standard curve is constructed by plotting absorbance values against concentrations of standards, and concentrations of unknown samples are determined using this standard curve.

HUMAN COMP ELISA CAT. NO.: RD194080200	
Assay format	Sandwich ELISA, Biotin-labelled antibody, 96 wells/kit
Samples	Serum, plasma
Standards	4 to 128 ng/ml
Limit of detection	0.4 ng/ml



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Precision

Intra-assay (Within-Run) (n=8)

Sample	Mean (ng/ml)	SD (ng/ml)	CV (%)
1	495	18.5	4.0
2	1 290	102.5	8.0

Inter-assay (Run-to-Run) (n=8)

Sample	Mean (ng/ml)	SD (ng/ml)	CV (%)
1	625	19.1	3.1
2	1 604	105.1	6.6

Spiking recovery

Serum samples were spiked with different amounts of human COMP and assayed.

Sample	Observed (ng/ml)	Expected (ng/ml)	Recovery O/E (%)
1	495	-	-
	2 200	2 495	88.2
	1 405	1 495	94.0
	910	995	91.5
2	1 220	-	-
	3 630	3 220	112.7
	2 230	2 220	100.5
	1 700	1 720	98.8

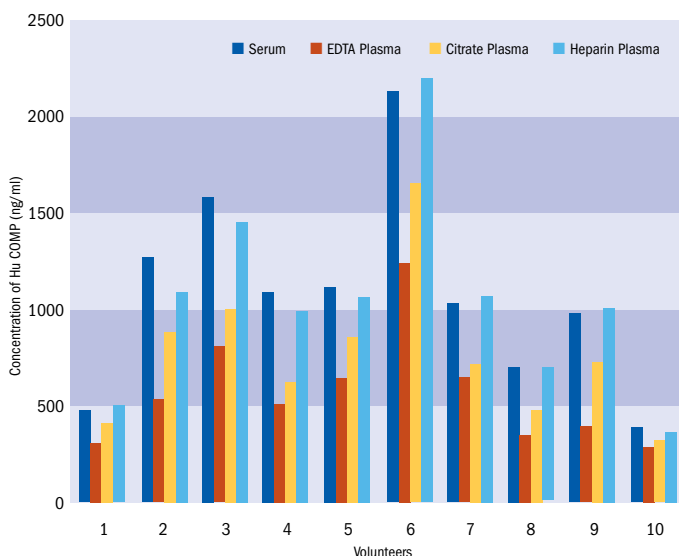
Linearity

Serum samples were serially diluted with Dilution Buffer and assayed.

Sample	Dilution	Observed (ng/ml)	Expected (ng/ml)	Recovery O/E (%)
1	-	1 817	-	-
	2x	937	909	103.9
	4x	500	454	105.4
	8x	207	227	83.7
2	-	3 970	-	-
	2x	1 940	1 985	97.7
	4x	990	992	99.7
	8x	420	496	84.6

Effect of sample matrix

EDTA, citrate and heparin plasmas were compared to respective serum samples from the same 10 individuals. Results are shown below:



Summary of protocol

- Reconstitute QCs and Master Standard and prepare set of standards
- Dilute samples 50x
- Add Standards, QCs and samples 100 µl
- Incubate at RT for 1 hours with shaking 300 rpm
- Wash plate 3 times
- Add Biotin Labelled Antibody 100 µl
- Incubate at RT for 1 hour with shaking 300 rpm
- Wash plate 3 times
- Add 100 µl Streptavidin-HRP Conjugate
- Incubate at RT for 30 min with shaking 300 rpm
- Wash plate 3 times
- Add 100 µl Substrate Solution
- Incubate at RT for 10 min
- Add 100 µl stop solution
- Read absorbance and calculate results

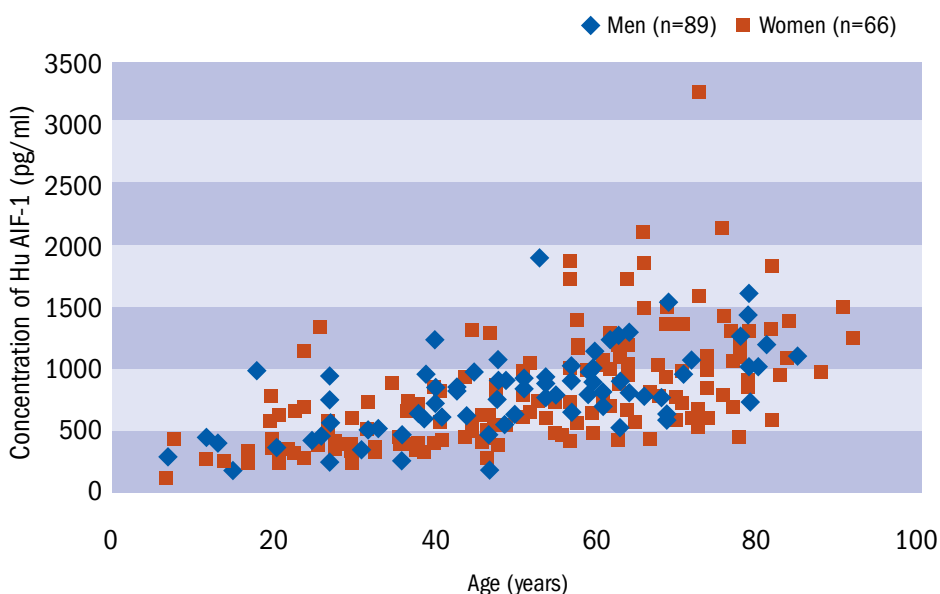
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Preliminary Population Data

The following results were obtained when serum samples from 246 unselected donors (165 female + 81 male) 7-92 years old were assayed with the Biovendor Human Cartilage Oligomeric Matrix Protein ELISA in our laboratory.

Age and sex dependent distribution of COMP

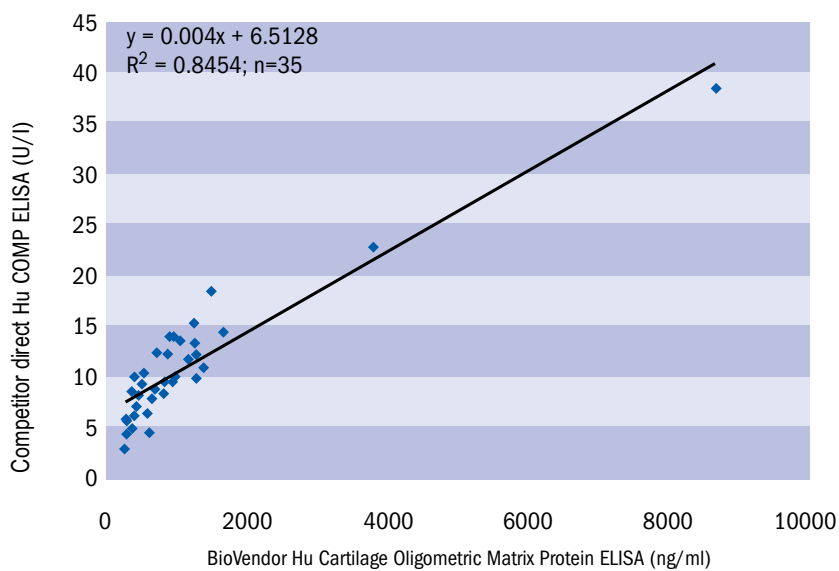
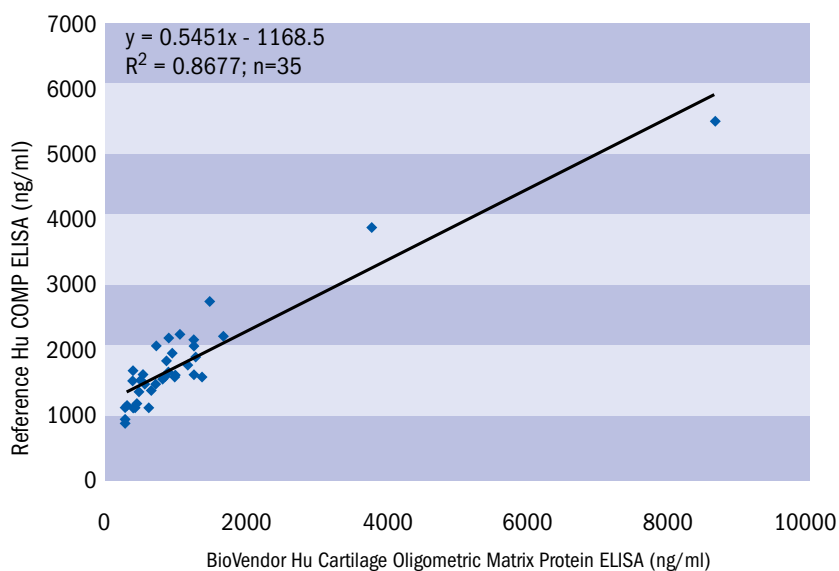
Sex	Age (years)	n	Mean COMP (ng/ml)	SD COMP (ng/ml)	Min. COMP (ng/ml)	Max. COMP (ng/ml)
Male	7-19	5	453	314	180	987
	20-29	9	523	221	234	939
	30-39	8	530	212	249	963
	40-49	15	763	268	260	1 242
	50-59	13	915	320	516	1 911
	60-69	20	925	272	519	1 551
	70-92	11	1 136	244	729	1 620
Female	7-19	7	257	98	93	414
	20-29	25	488	268	255	1 329
	30-39	20	487	206	204	888
	40-49	24	621	269	270	1 299
	50-59	22	867	407	408	1 884
	60-69	29	1 018	429	423	2 111
	70-92	38	1 091	527	432	3 250



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Method Comparison

The BioVendor Human Cartilage Oligomeric Matrix Protein ELISA was compared to the other commercial immunoassay, by measuring 35 serum samples. The following correlation graph was obtained:



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Related products

- RD172080100 Cartilage Oligomeric Matrix Protein Human HEK293
- RD182080100C4 Cartilage Oligomeric Matrix Protein Human, Mouse Monoclonal Antibody, Clone: 12C4
- RD182080100F1 Cartilage Oligomeric Matrix Protein Human, Mouse Monoclonal Antibody, Clone: 16F12
- RD182080100C1 Cartilage Oligomeric Matrix Protein Human, Mouse Monoclonal Antibody, Clone: 17C10

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