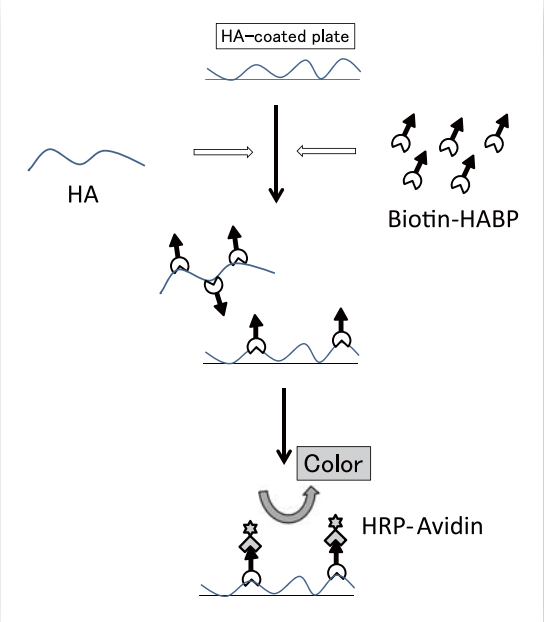
Highly Specific for Hyaluronan

Hyaluronan Quantification Kit

Background



Measurement principle (Competition Method)
A sample containing HA and Biotin-HABP are added to an HA-coated plate. HRP-Avidin is then added, and the HA binding Biotin-HABP is detected by chromophoric substrate.
HA in sample displaces Biotin-HABP from HA coated well plates.

Hyaluronan (HA) is an unbranched glycosaminoglycan composed of repeating disaccharide units of D-glucronic acid and *N*-acetyl-D-glucosamine. HA is abundant in synovial fluid, skin, umbilical cord, and vitreous bodies. HA is a prominent component of the extracellular matrix and contributes to tissue water retention and to cellular growth, differentiation, and migration. HA exists in the body in a wide range of molecular weights (MW) derived from several distinct HA synthases and degradation enzymes. For example, the average molecular weights of HA in synovial fluid, blood and urine are 4-6 MDa, 100-300 kDa and less 10 kDa, respectively.

This product is a competitive ELISA-like kit using an HA binding protein optimized to quantify HA polymers of average molecular weight between 7.4 kDa and 2,400 kDa in samples such as serum, plasma and culture supernatant.

Features

- **Hardly affected by HA molecular size**
- **Measures HA concentrations accurately in serum, plasma and culture supernatant where HA molecular weight is above 7.4 kDa**
- **Measures HA concentrations between 3.13 ～ 200 ng/mL**
- **Negligable cross-reaction with other glycosaminoglycans**

Kit Components

Description	Volume	Size
96 well plate	8 x 12 wells	1
HA Coating Solution	11 mL	1
Wash Buffer (20X)	50 mL	1
Sample Buffer (2X) *	50 mL	1
Blocking Buffer (2X) *	11 mL	1
HA Standard (10μg/mL) *	0.1 mL	1
Biotin-HABP *	10 ～ 40 μL	1
HRP-Avidin (100X) *	0.2 mL	1
Substrate Solution A	1 mL	1
Substrate Solution B	10 mL	1
Stop Solution	11 mL	1
Plate Seals	5 mL	2

Materials Required But Not Provided

- Microplate reader capable of measuring absorbance at 450 nm
- Horizontal orbital microplate shaker

* : contains 0.025 ～ 0.05% Proclin 300
Storage: -20℃ or below

Ordering Information

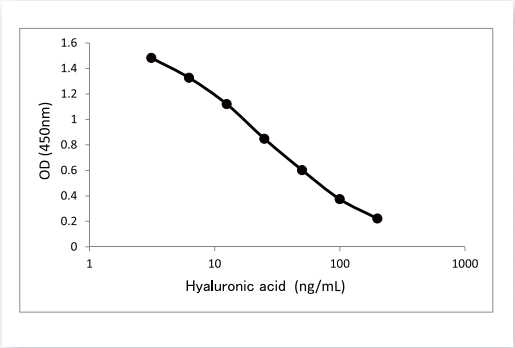
Description	Cat. No.	Size
Hyaluronan Quantification Kit	CSR-HA-96KIT	1 kit

Related Products

Fluoresceinamine labeled Sodium Hyaluronate ····· page 2
Hyaluronan Binding Protein (HABP) ····· page 3
Fluoresceinamine labeled Glycosaminoglycans ····· page 4
Hyaluronan Acid Oligosaccharides ····· page 4

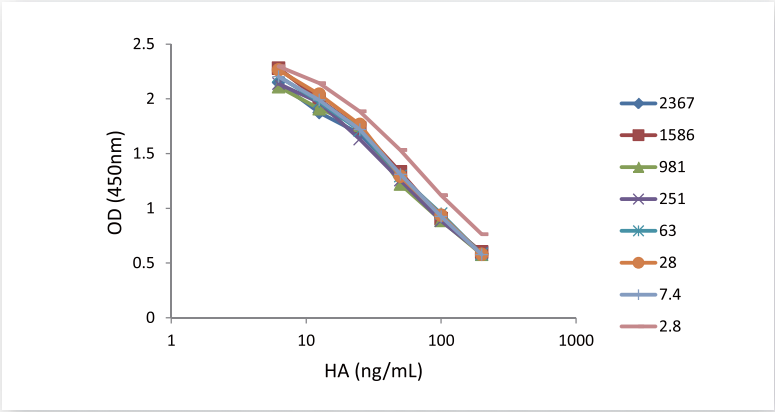
Hyaluronan Quantification Kit

Measuring Range



Typical calibration curve
The measuring range of HA using Hyaluronan Quantification Kit is 3.13 ～ 200 ng/mL.

Product Data: Reactivity & Specificity



The reactivity with various molecular weights of HA（HA with molecular weights of from 2.8 to 2367 kDa were compared）

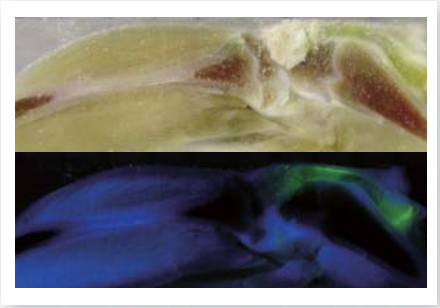
The reactivity with various molecular weights of HA was evaluated using Hyaluronan Quantification Kit . When using HA with molecular weights of from 2.8 to 2367 kDa, this kit reacted with molecular weights of over 7.4 kDa HA in a similar manner. HA with a molecular weight of 2.8 kDa showed slightly weaker reactivity.

Related Products

Fluoresceinamine labeled Sodium Hyaluronate

Background

This product is prepared by the fluorescent labeling of HA. Fluoresceinamine molecules are chemically attached to carboxyl groups of the GlcUA of HA. This solution is dissolved in PBS (−) and sterilized by filtration.



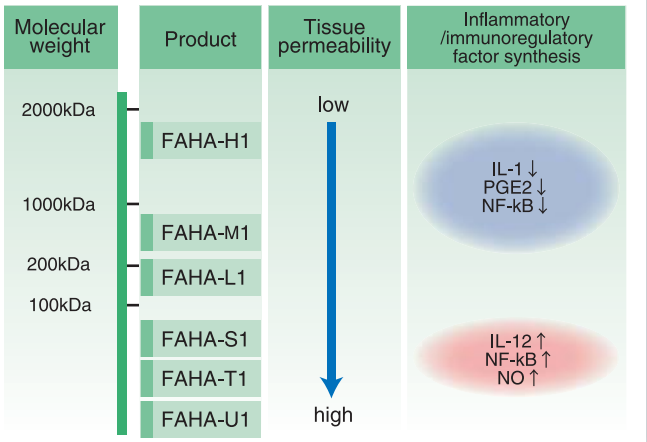
Fluorescence photograph of rat knee joint after intra-articular injection of FAHA-M1

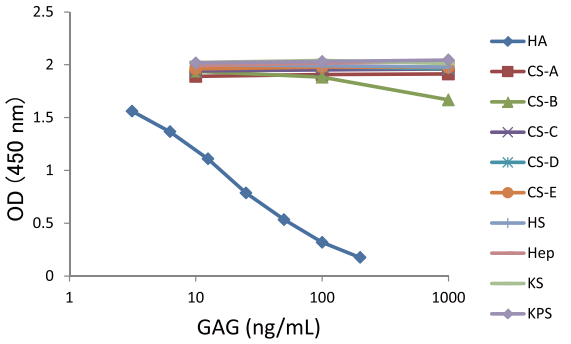
Specification

	Specification
Appearance	yellow green solution or yellow green lyophilizate
Concentration of FAHA (PBS(-) solution)	0.90 - 1.10 mg/mL
Mw calculated by Intrinsic Viscosity	1211 × 10^3 - 1603 × 10^3 dalton (FAHA-H1, H2)
	602 × 10^3 - 1118 × 10^3 dalton (FAHA-M1, M2)
	102 × 10^3 - 304 × 10^3 dalton (FAHA-L1, L2)
	40 × 10^3 - 80 × 10^3 dalton (FAHA-S1)
	20 × 10^3 - 30 × 10^3 dalton (FAHA-T1)
	5 × 10^3 - 10 × 10^3 dalton (FAHA-U1)
Wavelength of Excitation	490 - 500 nm
Wavelength of Emission	515 - 525 nm
Degree of Substitution*	0.600 - 1.100%
Fluorescent Purity on Electrophoresis	one band at same mobility with original NaHA
Endotoxin Level (PBS(-) solution)	< 0.25 EU/mL
Storage	Store protected from light at -20° C or below. We recommend storing in aliquots appropriate for anticipated usage.

NaHA : Sodium Hyaluronate
*Degree of Substitution is expressed as the molar percentage of bound fluoresceinamine groups to disaccharide units.

Effects of the different molecular weights of sodium hyaluronate on the tissue permeability and inflammatory/immunoregulatory factor synthesis





The cross-reactivity with other glycosaminoglycans (GAGs)

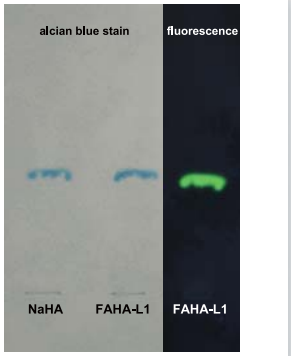
Cross-reactivity with other glycosaminoglycans (GAGs) was evaluated using the Hyaluronan Quantification Kit. This kit did not show cross-reactivity with chondroitin sulfate-A (CS-A), CS-C, CS-D, CS-E, heparan sulfate (HS), heparin (Hep), keratan sulfate (KS) and keratan polysulfate (KPS) at the concentrations of up to 1000 ng/mL. Weak reactivity was observed at high concentrations of CS-B (dermatan sulfate).

Samples	Number of Samples	Mean (ng/mL)
Human plasma	30	18.1
Human serum	3	29.2
Rabbit serum	8	37.1
Rat serum	2	213.3

HA contents in plasma and serum

References

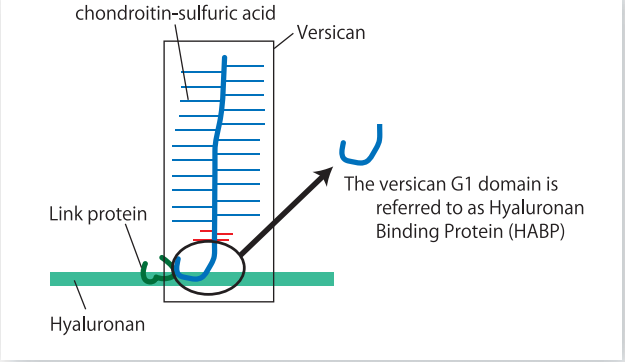
1. Maeda H, *et al.*, A competitive enzyme-linked immunosorbent assay-like method for the measurement of urinary hyaluronan. *Biosci Biotechnol Biochem.* 1999 May; **63**(5): 892-5.



Cellulose acetate membrane electrophoresis of FAHA-L1

Description	Cat. No.	Size
Fluoresceinamine Labeled Sodium Hyaluronate (H1) Specification: 1211 × 10^3- 1603 × 10^3 dalton	CSR-FAHA-H1	3 mL
Fluoresceinamine Labeled Sodium Hyaluronate (H2) Specification: 1211 × 10^3- 1603 × 10^3 dalton lyophilized product	CSR-FAHA-H2	3 mg
Fluoresceinamine Labeled Sodium Hyaluronate (M1) Specifi cation: 602 × 10^3- 1118 × 10^3 dalton	CSR-FAHA-M1	3 mL
Fluoresceinamine Labeled Sodium Hyaluronate (M2) Specification: 602 × 10^3- 1118 × 10^3 dalton lyophilized product	CSR-FAHA-M2	3 mg
Fluoresceinamine Labeled Sodium Hyaluronate (L1) Specification: 102 × 10^3- 304 × 10^3 dalton	CSR-FAHA-L1	3 mL
Fluoresceinamine Labeled Sodium Hyaluronate (L2) Specification: 102 × 10^3- 304 × 10^3 dalton lyophilized product	CSR-FAHA-L2	3 mg
Fluoresceinamine Labeled Sodium Hyaluronate (S1) Specification: 40 × 10^3- 80 × 10^3 dalton	CSR-FAHA-S1	3 mL
Fluoresceinamine Labeled Sodium Hyaluronate (T1) Specification: 20 × 10^3 - 30 × 10^3 dalton	CSR-FAHA-T1	3 mL
Fluoresceinamine Labeled Sodium Hyaluronate (U1) Specification: 5 × 10^3- 10 × 10^3 dalton	CSR-FAHA-U1	3 mL
Fluoresceinamine Labeled Sodium Hyaluronate(U2) Specification: 5x10^3- 10x10^3 dalton	CSR-FAHA-U2	3 mg

Hyaluronan Binding Protein (HABP)



Association of Versican and Hyaluronan

References

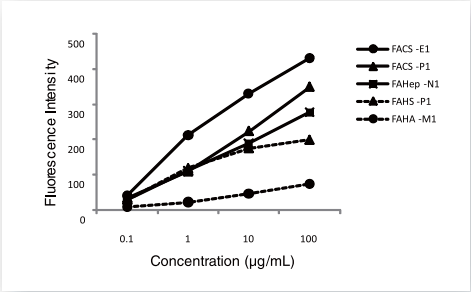
1. Tengblad A, *et al.*, A comparative study of the binding of cartilage link protein and the hyaluronate-binding region of the cartilage proteoglycan to hyaluronate-substituted Sepharose gel. *Biochem J.* 1981 Nov 1; **199**(2): 297-305.

2. Tengblad A, *et al.*, Quantitative analysis of hyaluronate in nanogram amounts. *Biochem J.* 1980 Jan 1; **185**(1): 101-5.

Description	Cat. No.	Size
Hyaluronan Binding Protein [HABP], Recombinant	HKD-BC40	50 µg
Biotinylated Hyaluronan Binding Protein [Biotin-HABP],Recombinant	HKD-BC41	50 µg

Fluoresceinamine labeled Glycosaminoglycans

- Research interactions with cells, growth factors or cytokines
- Analyze GAG degradation enzymes



Analysis of binding activities with bFGF

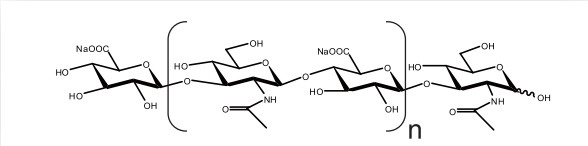
All Products
Ex: 490 - 500 nm
Em: 515 - 525nm

Description	Cat. No.	Size
Chondroitin Sulfate		
Fluoresceinamine Labeled Sodium Chondroitin Sulfate A (A1)	CSR-FACS-A1	3 mL
Fluoresceinamine-labeled Sodium Chondroitin Sulfate C	CSR-FACS-C2(SHC)3	3 mg
Fluoresceinamine Labeled Sodium Chondroitin Sulfate (C1)	CSR-FACS-C1	3 mL
Fluoresceinamine Labeled Sodium Chondroitin Sulfate D (D1)	CSR-FACS-D1	3 mL
Fluoresceinamine Labeled Sodium Chondroitin Sulfate E (E1)	CSR-FACS-E1	3 mL
Fluoresceinamine-labeled Sodium Chondroitin Sulfate E	CSR-FACS-E2(SQC)3	3 mg
Fluoresceinamine Labeled Sodium Chondroitin Poly-Sulfate (P1)	CSR-FACS-P1	3 mL
Dermatan Sulfate		
Fluoresceinamine Labeled Sodium Dermatan Sulfate (B1)	CSR-FADS-B1	3 mL
Heparin		
Fluoresceinamine Labeled Sodium Heparin (N1)	CSR-FAHEP-N1	3 mL
Heparan Sulfate		
Fluoresceinamine-labeled Sodium Heparan Sulfate	CSR-FAHS-P2(PGK)1	1 mg
	CSR-FAHS-P2(PGK)3	3 mg
Fluoresceinamine Labeled Sodium Heparan Sulfate (P1)	CSR-FAHS-P2(PGK)1	1 mL

Specification: 1 mg/ml PBS(-)

Hyaluronan Acid Oligosaccharides

Low molecular weight hyaluronans may exhibit activities not observed with larger species, or even exhibit opposite effects. For example, HA6 has been shown to induce NO and MMPs in chodrocytes. The HA oligosaccharides shown here are also available as endotoxin free for work with sensitive cell-based systems.



	Hexamer	Octamer	Decamer	Dodecamer
n	2	3	4	5
M. W.	1221.29	1607.21	1992.51	2393.81

Description	Cat. No.	Size
Hyaluronan Oligosaccharide 4 mer (HA4)	CSR-11001	3 mg
Hyaluronan Oligosaccharide 4 mer (HA4), Endotoxin Free	CSR-11006	3 mg
Hyaluronan Oligosaccharide 6 mer (HA6)	CSR-11002	3 mg
Hyaluronan Oligosaccharide 6 mer (HA6), Endotoxin Free	CSR-11007	3 mg
Hyaluronan Oligosaccharide 8 mer (HA8)	CSR-11003	3 mg
Hyaluronan Oligosaccharide 8 mer (HA8), Endotoxin Free	CSR-11008	3 mg
Hyaluronan Oligosaccharide 10 mer (HA10)	CSR-11004	3 mg
Hyaluronan Oligosaccharide 10 mer (HA10), Endotoxin Free	CSR-11009	3 mg
Hyaluronan Oligosaccharide 12 mer (HA12)	CSR-11005	1 mg
Hyaluronan Oligosaccharide 12 mer (HA12), Endotoxin Free	CSR-11010	1mg
Hyaluronan oligosaccharide assortment (HA4, HA6, HA8, HA10, HA12)	CSR-93001	1 set
Hyaluronan oligosaccharide assortment (HA4, HA6, HA8, HA10, HA12), Endotoxin Free	CSR-93002	1 set

References

1. Stern R, *et al.*, Hyaluronan fragments: an information-rich system. *Eur J Cell Biol.* 2006 Aug; **85**(8): 699-715.
2. Tawada A, *et al.*, Large-scale preparation, purification, and characterization of hyaluronan oligosaccharides from 4-mers to 52-mers. *Glycobiology.* 2002 Jul; **12**(7): 421-6.

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