abcam

Product datasheet

Human Fibronectin ELISA Kit ab219046

יילצעבע SimpleStep ELISA

5 References 画像数 18

製品の概要

製品名

Human Fibronectin ELISA Kit

検出方法

Colorimetric

再現性

Intra-Assay(同時再現性)

| サンプル | N | 平均值 | SD | CV% |
|-------|---|-----|----|-----|
| Serum | 3 | | | 5% |

Inter-Assay(日差再現性)

| サンプル | N | 平均值 | SD | CV% |
|-------|---|-----|----|------|
| Serum | 5 | | | 8.3% |

サンプルの種類

Cell culture supernatant, Milk, Serum, Cell culture extracts, Tissue Extracts, EDTA Plasma, Cit

plasma

アッセイタイプ

Sandwich (quantitative)

検出感度

20.6 pg/ml

検出範囲

125 pg/ml - 8000 pg/ml

添加回収試験

特定サンプルでの回収試験

| サンプルの種類 | 平均 % | 測定範囲 |
|--------------------------|------|------------|
| Cell culture supernatant | 102 | 90% - 117% |
| Milk | 102 | 91% - 111% |
| Serum | 93 | 87% - 102% |
| Cell culture extracts | 89 | 80% - 105% |
| Tissue Extracts | 90 | 80% - 131% |
| EDTA Plasma | 86 | 81% - 94% |

| サンプルの種類 | 平均 % | 測定範囲 |
|------------|------|------------|
| Cit plasma | 99 | 87% - 120% |

全工程の試験時間

ステップ

種交差性

製品の概要

1h 30m

One step assay

交差種: Human, Rhesus monkey

非交差種: Cow

Human Fibronectin ELISA Kit (ab219046) is a single-wash 90 min sandwich ELISA designed for the quantitative measurement of Fibronectin protein in cell culture supernatant, cit plasma, edta plasma, milk, serum, tissue extracts, and cell culture extracts. It uses our proprietary SimpleStep ELISA® technology. Quantitate Human Fibronectin with 20.6 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 (<u>ab203359</u>) is available to use as an alternative to the 96-well microplate provided with SimpleStep ELISA® kits.

ASSAY SPECIFICITY This kit recognizes both native and recombinant human Fibronectin protein in serum, plasma (citrate and EDTA), milk, cell culture supernatant, and cell and tissue extract samples only.

CROSS REACTIVITY Recombinant human Anastelin was prepared within the working range of the assay and assayed for cross reactivity. No cross-reactivity was observed.

SPECIES REACTIVITY This kit recognizes human Fibronectin protein.

Other species reactivity was determined by measuring 1/100,000 serum (mouse, rat, cow) or plasma (Rhesus macaque) samples of various species, interpolating the protein concentrations from the human standard curve, and expressing the interpolated concentrations as a percentage of the protein concentration in human serum or plasma assayed at the same dilution.

特記事項

Fibronectin is a large glycoprotein present in the extracellular matrix and circulating plasma. Fibronectin is important in many cell adhesion and migration related processes, including wound healing, embryogenesis and nerve regeneration. Differential expression of fibronectin is seen in coronary heart disease, glomerulopathy and tumor cell metastasis. The protein contains binding

sites for collagen, heparin and fibrin and is a specific ligand for several integrin adhesion receptors. Fibronectin exists as a dimer or multimer.

Abcam has not and does not intend to apply for the REACH Authorisation of customers' uses of products that contain European Authorisation list (Annex XIV) substances.

It is the responsibility of our customers to check the necessity of application of REACH Authorisation, and any other relevant authorisations, for their intended uses.

試験プラットフォーム

Pre-coated microplate (12 x 8 well strips)

製品の特性

保存方法

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

| 内容 | 1 x 96 tests |
|---|--------------|
| 10X Wash Buffer PT (ab206977) | 1 x 20ml |
| 50X Cell Extraction Enhancer Solution (ab193971) | 1 x 1ml |
| 5X Cell Extraction Buffer PTR (ab193970) | 1 x 10ml |
| Antibody Diluent 4BR | 1 x 6ml |
| 10X Human Fibronectin Capture Antibody | 1 x 600µl |
| 10X Human Fibronectin Detector Antibody | 1 x 600µl |
| Human Fibronectin Lyophilized Recombinant Protein | 2 vials |
| Plate Seals | 1 unit |
| Sample Diluent NS (ab193972) | 1 x 50ml |
| SimpleStep Pre-Coated 96-Well Microplate (ab206978) | 1 unit |
| Stop Solution | 1 x 12ml |
| TMB Development Solution | 1 x 12ml |

機能

Fibronectins bind cell surfaces and various compounds including collagen, fibrin, heparin, DNA, and actin. Fibronectins are involved in cell adhesion, cell motility, opsonization, wound healing, and maintenance of cell shape. Involved in osteoblast compaction through the fibronectin fibrillogenesis cell-mediated matrix assembly process, essential for osteoblast mineralization. Participates in the regulation of type I collagen deposition by osteoblasts.

Anastellin binds fibronectin and induces fibril formation. This fibronectin polymer, named superfibronectin exhibits enhanced adhesive properties. Both anastellin and superfibronecting

superfibronectin, exhibits enhanced adhesive properties. Both anastellin and superfibronectin inhibit tumor growth, angiogenesis and metastasis. Anastellin activates p38 MAPK and inhibits lysophospholipid signaling.

組織特異性

Plasma FN (soluble dimeric form) is secreted by hepatocytes. Cellular FN (dimeric or cross-linked multimeric forms), made by fibroblasts, epithelial and other cell types, is deposited as fibrils in the extracellular matrix. Ugl-Y1, Ugl-Y2 and Ugl-Y3 are found in urine.

関連疾患 Glomerulopathy with fibronectin deposits 2

配列類似性 Contains 12 fibronectin type-I domains.

Contains 2 fibronectin type-II domains.
Contains 16 fibronectin type-III domains.

発生段階 Ugl-Y1, Ugl-Y2 and Ugl-Y3 are present in the urine from 0 to 17 years of age.

翻訳後修飾 Sulfated.

It is not known whether both or only one of Thr-2064 and Thr-2065 are/is glycosylated.

Forms covalent cross-links mediated by a transglutaminase, such as F13A or TGM2, between a

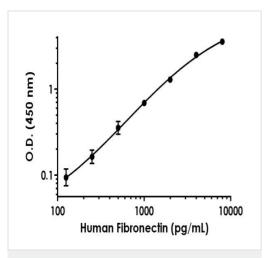
glutamine and the epsilon-amino group of a lysine residue, forming homopolymers and heteropolymers (e.g. fibrinogen-fibronectin, collagen-fibronectin heteropolymers).

Phosphorylated by FAM20C in the extracellular medium.

Proteolytic processing produces the C-terminal NC1 peptide, anastellin.

細胞内局在 Secreted, extracellular space, extracellular matrix.

画像

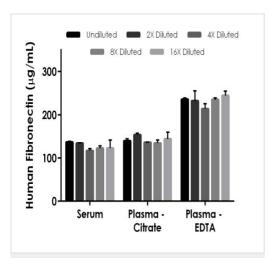


Example of human Fibronectin standard curve in Sample Diluent NS.

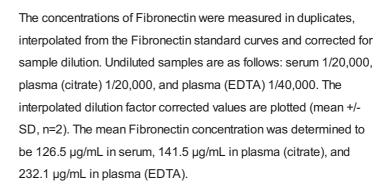
| Standard Curve Measurements | | | | | |
|-----------------------------|-------|-------|-------|--|--|
| Concentration | O.D 4 | Mean | | | |
| (pg/ml) | 1 | 2 | O.D | | |
| 0 | 0.148 | 0.161 | 0.154 | | |
| 125 | 0.234 | 0.264 | 0.249 | | |
| 250 | 0.298 | 0.340 | 0.319 | | |
| 500 | 0.468 | 0.555 | 0.511 | | |
| 1,000 | 0.816 | 0.874 | 0.845 | | |
| 2,000 | 1.453 | 1.443 | 1.448 | | |
| 4,000 | 2.622 | 2.704 | 2.663 | | |
| 8,000 | 3.687 | 3.760 | 3.724 | | |

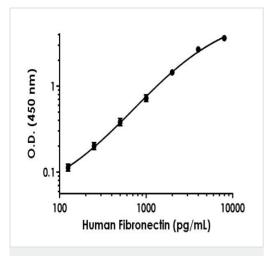
Example of human Fibronectin standard curve in Sample Diluent NS.

Example of human Fibronectin standard curve in Sample Diluent NS. The Fibronectin standard curve was prepared as described. Raw data values are shown in the table. Background-subtracted data values (mean +/- SD) are graphed.



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





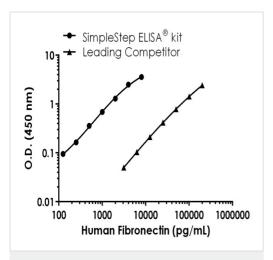
Example of human Fibronectin standard curve in 1X Cell Extraction Buffer PTR.

Background-subtracted data values (mean +/- SD) are graphed.

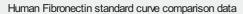
| Concentration | O.D 4 | Mean | |
|---------------|-------|-------|-------|
| (pg/ml) | 1 | 2 | O.D |
| 0 | 0.157 | 0.153 | 0.155 |
| 125 | 0.275 | 0.260 | 0.267 |
| 250 | 0.369 | 0.342 | 0.356 |
| 500 | 0.563 | 0.510 | 0.537 |
| 1,000 | 0.926 | 0.833 | 0.880 |
| 2,000 | 1.668 | 1.537 | 1.603 |
| 4,000 | 2.996 | 2.686 | 2.841 |
| 8,000 | 3.821 | 3.733 | 3.777 |

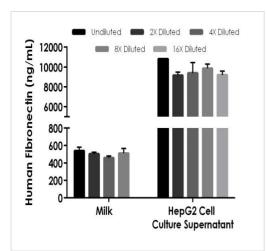
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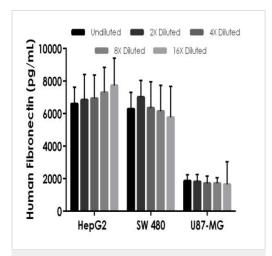


Standard curve comparison between human Fibronectin
SimpleStep ELISA® kit and traditional ELISA kit from leading
competitor. SimpleStep ELISA kit shows comparable sensitivity.

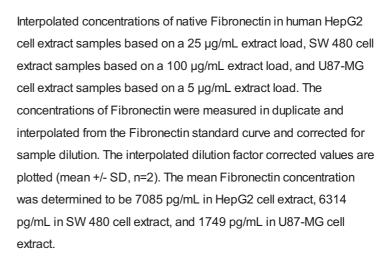


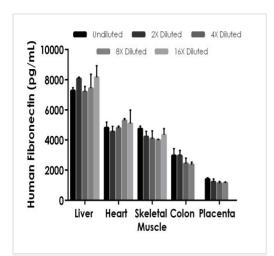


Interpolated concentrations of native Fibronectin in human breast milk and HepG2 cell culture supernatant (4 days) samples. The concentrations of Fibronectin were measured in duplicates, interpolated from the Fibronectin standard curves and corrected for sample dilution. Undiluted samples are as follows: milk 1/100, and HepG2 cell culture supernatant 1/2000. The interpolated dilution factor corrected values are plotted (mean +/- SD, n=2). The mean Fibronectin concentration was determined to be 502.9 ng/mL in milk, and 9683 ng/mL in HepG2 cell culture supernatant.



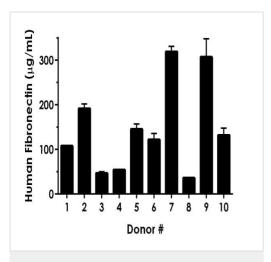
Interpolated concentrations of native Fibronectin in HepG2, SW 480 and U87-MG cell extracts.





Interpolated concentrations of native Fibronectin in various tissue extracts.

Interpolated concentrations of native Fibronectin in human liver tissue extract based on a 100 µg/mL extract load, heart tissue extract based on a 100 µg/mL extract load, skeletal muscle based on a 100 µg/mL extract load, colon tissue extract based on a 5 µg/mL extract load, and placenta tissue extract based on a 1.25 µg/mL extract load. The concentrations of Fibronectin were measured in duplicate and interpolated from the Fibronectin standard curve and corrected for sample dilution. The interpolated dilution factor corrected values are plotted (mean +/- SD, n=2). The mean Fibronectin concentration was determined to be 7634 pg/mL in liver tissue extract, 4910 pg/mL in heart tissue extract, 4278 pg/mL in skeletal muscle tissue extract, 2684 pg/mL in colon tissue extract, and 1224 pg/mL in placenta tissue extract.



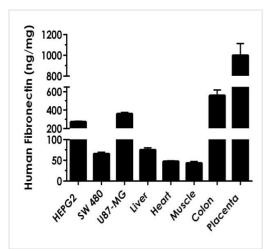
Serum from ten individual healthy male human donors was measured in duplicate.

Interpolated dilution factor corrected values are plotted (mean +/- SD, n=2). The mean Fibronectin concentration was determined to be 146.2 μ g/mL with a range of 35.99 – 319.2 μ g/mL.

| Sample Diluent Buffer | n= | Minimal Detectable Dose |
|-------------------------------|----|----------------------------|
| Sample Diluent NS | 16 | 20.6 pg/mL |
| 1X Cell Extraction Buffer PTR | 32 | 22.3 pg/mL |

Assay sensitivity.

The MDD was determined by calculating the mean of zero standard replicates and adding 2 standard deviations then extrapolating the corresponding concentration.



Interpolated concentrations of native Fibronectin in human cell and tissue extract samples.

The concentrations of Fibronectin were measured in three different dilutions in duplicate and interpolated from the Fibronectin standard curve and corrected for sample dilution. The interpolated dilution factor corrected values are plotted in ng of Fibronectin per mg of extract (mean +/- SD, n=3). Fibronectin concentration was determined to be 271.8 ng/mg in HepG2 cell extract, 65.52 ng/mg in SW 480 cell extract, 359.6 ng/mg in U87-MG cell extract, 75.22 ng/mg in liver tissue extract, 47.20 ng/mg in heart tissue extract, 43.52 ng/mg in skeletal muscle tissue extract, 557.9 ng/mg in colon tissue extract, and 998.8 ng/mg in placenta tissue extract samples.

| Dilution Factor | Interpolated value | 1/20,000 Human Serum | 1/20,000 Human Plasma (Citrate) | 1/40,000 Human Plasma (EDTA) | 1/100 De-fatted Breast Milk | 1/2,000 HepG2 SN (4 days) |
|--------------------|--------------------|----------------------------|--|---------------------------------------|--------------------------------------|------------------------------------|
| | pg/mL | 6832 | 6984 | 5890 | 5398 | 5408 |
| Undiluted | % Expected value | 100 | 100 | 100 | 100 | 100 |
| 2 | pg/mL | 3349 | 3836 | 2901 | 2520 | 2289 |
| | % Expected value | 98 | 110 | 99 | 93 | 85 |
| 4 | pg/mL | 1459 | 1700 | 1335 | 1144 | 1174 |
| | % Expected value | 85 | 97 | 91 | 85 | 87 |
| 8 | pg/mL | 765.2 | 838.8 | 733.9 | 638.1 | 616.5 |
| | % Expected value | 90 | 96 | 100 | 95 | 91% |
| 16 | pg/mL | 383.3 | 450.1 | 381.8 | NL | 287.1 |
| | % Expected value | 90 | 103 | 104 | NL | 85 |

NL - Non-Linear

Linearity of dilution.

Linearity of dilution is determined based on interpolated values from the standard curve. Linearity of dilution defines a sample concentration interval in which interpolated target concentrations are directly proportional to sample dilution.

Native Fibronectin was measured in serum, plasma, de-fatted breast milk, and cell culture supernatant (SN) samples in a 2-fold dilution series. Sample dilutions are made in Sample Diluent NS.

| Dilution Factor | Interpolated value | 25 µg/mL HepG2 Cell Extract | 100 µg/mL SW 480 Cell Extract | 5 µg/ml U87-MG Cell Extract |
|--------------------|--------------------|--------------------------------------|--|--------------------------------------|
| | pg/mL | 6602 | 6291 | 1860 |
| Undiluted | % Expected value | 100 | 100 | 100 |
| 2 | pg/mL | 3423 | 3508 | 909.3 |
| | % Expected value | 104 | 112 | 98 |
| - 12 | pg/mL | 1735 | 1587 | 428.9 |
| 4 | % Expected value | 105 | 101 | 92 |
| | pg/mL | 913.2 | 767.8 | 213.6 |
| 8 | % Expected value | 111 | 98 | 92 |
| 16 | pg/mL | 483.2 | 360.7 | 102.7 |
| | % Expected value | 117 | 92 | 88 |

Native Fibronectin was measured in the following biological samples in a 2-fold dilution series. Sample dilutions are made in 1X Cell Extraction Buffer PTR.

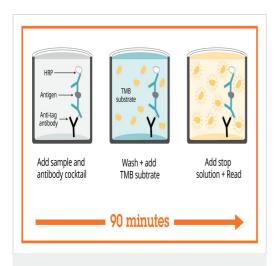
Linearity of dilution.

| Dilution Factor | Interpolated value | 100 µg/mL Liver Tissue Extract | 100 µg/mL Heart Tissue Extract | 100 µg/ml Skeletal Muscle Tissue Extract | 5 µg/mL Colon Tissue Extract | 1.25 µg/mL Placenta Tissue Extract |
|--------------------|--------------------|--|--|---|--|--|
| 11-20-1-2 | pg/mL | 7282 | 4817 | 4758 | 2973 | 1405 |
| Undiluted | % Expected value | 100 | 100 | 100 | 100 | 100 |
| 2 | pg/mL | 4040 | 2273 | 2109 | 1481 | 607.0 |
| | % Expected value | 111 | 94 | 89 | 100 | 86 |
| | pg/mL | 1801 | 1199 | 1020 | 608.6 | 281.5 |
| 4 | % Expected value | 99 | 100 | 86 | 82 | 80 |
| | pg/mL | 929.9 | 662.7 | 499.6 | 295.8 | 143.8 |
| 8 | % Expected value | 102 | 110 | 84 | 80 | 82 |
| 16 | pg/mL | 510.3 | 318.3 | 270.9 | NL | NL |
| | % Expected value | 112 | 106 | 91 | NL | NL |

NL - Non-Linear

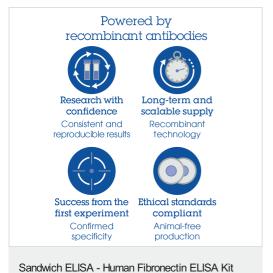
Linearity of dilution.

Native Fibronectin was measured in the following biological samples in a 2-fold dilution series. Sample dilutions are made in 1X Cell Extraction Buffer PTR.



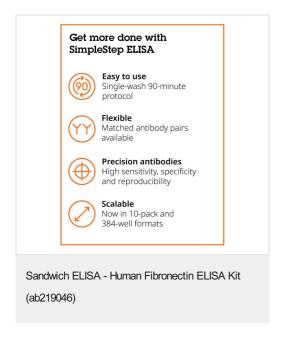
SimpleStep ELISA technology allows the formation of the antibodyantigen complex in one single step, reducing assay time to 90 minutes. Add samples or standards and antibody mix to wells all at once, incubate, wash, and add your final substrate. See protocol for a detailed step-by-step guide.

Sandwich ELISA - Human Fibronectin ELISA Kit (ab219046)



To learn more about the advantages of recombinant antibodies see **<u>here</u>**.

(ab219046)



To learn more about the advantages of SimpleStep ELISA[®] kits see **here**.

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

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