abcam

Product datasheet

Human CD40L ELISA Kit (TNFSF5) ab196268

Human CD40L ELISA Kit (TNFSF5)

Recombinant

SimpleStep ELISA

8 Images

Overview

Product name

Colorimetric

Detection method

Precision

Recovery

Intra-assay

Sample	n	Mean	SD	CV%
supernatant	5			3.8%

Inter-assay

Sample	n	Mean	SD	CV%
supernatant	3			2.8%

Sample type Cell culture supernatant, Serum, Plasma

Assay type Sandwich (quantitative)

Sensitivity 2.9 pg/ml

Range 12.5 pg/ml - 800 pg/ml

._.o pg/....

Sample specific recovery

Sample type	Average %	Range
Cell culture supernatant	95	84% - 105%
Serum	96	93% - 100%
Hep Plasma	106	102% - 112%
EDTA Plasma	96	94% - 99%
Cit plasma	104	96% - 119%

Assay time 1h 30m

Assay duration One step assay

1

Species reactivity

Product overview

Reacts with: Human

Human CD40L ELISA Kit (TNFSF5) (ab196268) is a single-wash 90 min sandwich ELISA designed for the quantitative measurement of CD40L (TNFSF5) protein in cell culture supernatant, serum, and plasma. It uses our proprietary SimpleStep ELISA® technology. Quantitate Human CD40L (TNFSF5) with 2.9 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.

Notes

CD40L (CD40 Ligand) is a glycoprotein belonging to the TNF family. CD40 Ligand is expressed predominantly on activated CD4 T lymphocytes and is also found in other cells including NK cells, mast cells, basophils, and eosinophils. It is involved in B cell proliferation and IgG production. CD40 Ligand is also involved in immunoglobulin class switching.

Platform

Microplate

Properties

Storage instructions

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

Components	1 x 96 tests
10X Human CD40L Capture Antibody	1 x 600µl
10X Human CD40L Detector Antibody	1 x 600µl
10X Wash Buffer PT (ab206977)	1 x 20ml
Antibody Diluent 4BI	1 x 6ml
Human CD40L Lyophilized Recombinant Protein	2 vials
Plate Seals	1 unit
Sample Diluent 25BS	1 x 20ml
Sample Diluent NS (ab193972)	1 x 50ml
SimpleStep Pre-Coated 96-Well Microplate (ab206978)	1 unit

Components	1 x 96 tests
Stop Solution	1 x 12ml
TMB Development Solution	1 x 12ml

Function Mediates B-cell proliferation in the absence of co-stimulus as well as IgE production in the

presence of IL-4. Involved in immunoglobulin class switching.

 $\label{eq:condition} \textit{Release of soluble CD40L from platelets is partially regulated by \textit{GP IIb/IIIa}, actin polymerization, \\$

and an matrix metalloproteinases (MMP) inhibitor-sensitive pathway.

Tissue specificity Specifically expressed on activated CD4+ T-lymphocytes.

Involvement in disease Defects in CD40LG are the cause of X-linked immunodeficiency with hyper-lgM type 1 (HIGM1)

[MIM:308230]; also known as X-linked hyper IgM syndrome (XHIM). HIGM1 is an immunoglobulin isotype switch defect characterized by elevated concentrations of serum IgM and decreased amounts of all other isotypes. Affected males present at an early age (usually within the first year of life) recurrent bacterial and opportunistic infections, including Pneumocystis carinii pneumonia and intractable diarrhea due to cryptosporidium infection. Despite substitution treatment with intravenous immunoglobulin, the overall prognosis is rather poor, with a death rate of about 10%

before adolescence.

Sequence similarities Belongs to the tumor necrosis factor family.

Post-translational The soluble form derives from the membrane form by proteolytic processing.

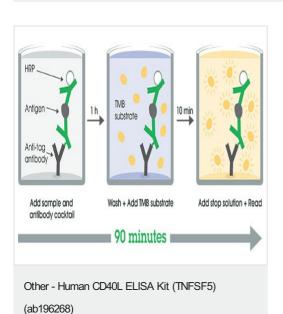
modifications N-linked glycan is a mixture of high mannose and complex type. Glycan structure does not

influence binding affinity to CD40.

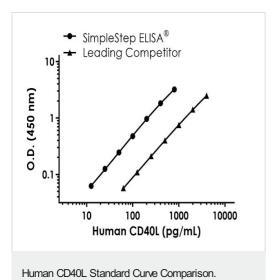
Not O-glycosylated.

Cellular localization Secreted and Cell membrane.

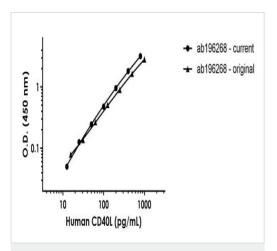
Images



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.

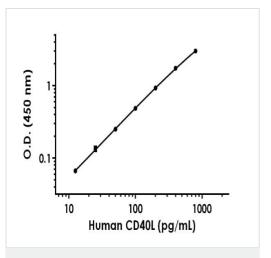


Standard Curve comparison between human CD40L SimpleStep ELISA kit and traditional ELISA kit from leading competitor. SimpleStep ELISA kit shows increased sensitivity.

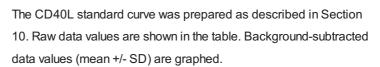


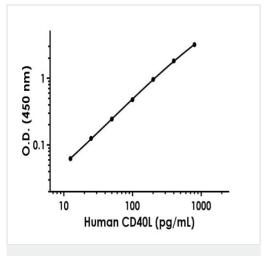
Human CD40L Standard Curve Comparison between current and original ELISA kit.

Standard Curve comparison between current human CD40L SimpleStep ELISA kit and original ELISA kit. The current SimpleStep ELISA kit shows increased sensitivity.

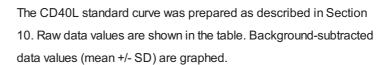


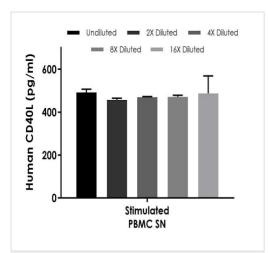
Example of human CD40L standard curve in Sample Diluent 25BS.





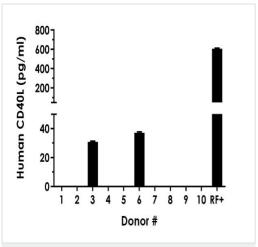
Example of human CD40L standard curve in Sample Diluent NS.





Interpolated concentrations of native CD40L in human PBMC cell culture supernatant samples stimulated for 46 hours with 1.5% PHA-M.

Unstimulated PBMC cell culture supernatant samples were also measured; all values were below the detectable range of the assay. The concentrations of CD40L were measured in duplicates, interpolated from the CD40L standard curves and corrected for sample dilution. Undiluted samples are as follows: stimulated PBMC supernatant 100%. The interpolated dilution factor corrected values are plotted (mean +/- SD, n=2). The mean CD40L concentration was determined to be 488 pg/ml in stimulated PBMC supernatant.



Serum from ten individual healthy human female donors and RF positive serum from one rheumatoid arthritis patient donor was measured in duplicate.

Interpolated dilution factor corrected values are plotted (mean +/-SD, n=2). The mean CD40L concentration in healthy serum was determined to be 34 pg/mL with a range of ND (non-detectable) – 37.6 pg/mL. The mean CD40L concentration in rheumatoid arthritis serum was determined to be 607 pg/mL.



(TNFSF5) (ab196268)

To learn more about the advantages of recombinant antibodies see **here**.

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