abcam

Product datasheet

Human IL-1 beta ELISA Kit ab214025

SimpleStep ELISA®

5 Images

Overview

Product name Human IL-1 beta ELISA Kit

Detection method Colorimetric

Precision

Intra-assay

Sample	n	Mean	SD	CV%
Overall	8			4.8%

Inter-assay

Sample	n	Mean	SD	CV%
Overall	3			5.6%

Sample type Cell culture supernatant, Serum, Heparin Plasma, EDTA Plasma, Citrate Plasma

Assay type Sandwich (quantitative)

Sensitivity 5.64 pg/ml

Range 14.06 pg/ml - 900 pg/ml

Recovery

Sample specific recovery

Sample type	Average %	Range
Cell culture supernatant	98	96% - 100%
Serum	103	101% - 105%
Heparin Plasma	100	99% - 101%
EDTA Plasma	93	90% - 96%
Citrate Plasma	86	84% - 88%

Assay time 1h 30m

Assay duration One step assay

Species reactivity **Reacts with:** Human
Does not react with: Mouse, Rat, Cow

Product overview Abcam's Human IL-1 beta SimpleStep ELISA® kit **(ab184861) has been re-developed** with new capture and detector antibodies. This new kit will have the same name but a different product number (ab214025). We have identified new recombinant monoclonal antibodies to use in the SimpleStep ELISA platform that provide a higher sensitivity when quantification of IL-1 beta in human serum, plasma, and cell culture supernatants.

IL-1beta *in vitro* SimpleStep ELISA® (Enzyme-Linked Immunosorbent Assay) kit is designed for the quantitative measurement of IL-1beta protein in human serum, plasma and cell culture supernatants.

The SimpleStep ELISA® employs an affinity tag labeled capture antibody and a reporter conjugated detector antibody which immunocapture the sample analyte in solution. This entire complex (capture antibody/analyte/detector antibody) is in turn immobilized via immunoaffinity of an anti-tag antibody coating the well. To perform the assay, samples or standards are added to the wells, followed by the antibody mix. After incubation, the wells are washed to remove unbound material. TMB substrate is added and during incubation is catalyzed by HRP, generating blue coloration. This reaction is then stopped by addition of Stop Solution completing any color change from blue to yellow. Signal is generated proportionally to the amount of bound analyte and the intensity is measured at 450 nm. Optionally, instead of the endpoint reading, development of TMB can be recorded kinetically at 600 nm.

Notes Interleukin 1 beta (IL-1 beta) is produced by activated macrophages and stimulates thymocyte proliferation by inducing IL-2 release, B-cell maturation and proliferation, and fibroblast growth factor activity. IL-1 proteins are involved in the inflammatory response, being identified as endogenous pyrogens, and are reported to stimulate the release of prostaglandin and collagenase from synovial cells.

Tested applications **Suitable for:** Sandwich ELISA

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 IL-1beta Capture Antibody	1 x 600µl
10X Human IL-1beta Detector Antibody	1 x 600µl
10X Wash Buffer PT (ab206977)	1 x 20ml
Antibody Diluent 4BI	1 x 6ml
Human IL-1beta Lyophilized Recombinant Protein	2 vials
Plate Seals	1 unit

Components	1 x 96 tests
SimpleStep Pre-Coated 96-Well Microplate (ab206978)	1 unit
Sample Diluent NS	1 x 50ml
Stop Solution	1 x 12ml
TMB Substrate	1 x 12ml

Function	Potent proinflammatory cytokine. Initially discovered as the major endogenous pyrogen, induces prostaglandin synthesis, neutrophil influx and activation, T-cell activation and cytokine production, B-cell activation and antibody production, and fibroblast proliferation and collagen production. Promotes Th17 differentiation of T-cells.
Tissue specificity	Expressed in activated monocytes/macrophages (at protein level).
Sequence similarities	Belongs to the IL-1 family.
Post-translational modifications	Activation of the IL1B precursor involves a CASP1-catalyzed proteolytic cleavage. Processing and secretion are temporarily associated.
Cellular localization	Cytoplasm, cytosol. Lysosome. Secreted, exosome. Cytoplasmic vesicle, autophagosome. Secreted. The precursor is cytosolic. In response to inflammasome-activating signals, such as ATP for NLRP3 inflammasome or bacterial flagellin for NLRC4 inflammasome, cleaved and secreted. IL1B lacks any known signal sequence and the pathway(s) of its secretion is(are) not yet fully understood (PubMed:24201029). On the basis of experimental results, several unconventional secretion mechanisms have been proposed. 1. Secretion via secretory lysosomes: a fraction of CASP1 and IL1B precursor may be incorporated, by a yet undefined mechanism, into secretory lysosomes that undergo Ca(2+)-dependent exocytosis with release of mature IL1B (PubMed:15192144). 2. Secretory autophagy: IL1B-containing autophagosomes may fuse with endosomes or multivesicular bodies (MVBs) and then merge with the plasma membrane releasing soluble IL1B or IL1B-containing exosomes (PubMed:24201029). However, autophagy impacts IL1B production at several levels and its role in secretion is still controversial. 3. Secretion via exosomes: ATP-activation of P2RX7 leads to the formation of MVBs containing exosomes with entrapped IL1B, CASP1 and other inflammasome components. These MVBs undergo exocytosis with the release of exosomes. The release of soluble IL1B occurs after the lysis of exosome membranes (By similarity). 4. Secretion by microvesicle shedding: activation of the ATP receptor P2RX7 may induce an immediate shedding of membrane-derived microvesicles containing IL1B and possibly inflammasome components. The cytokine is then released in the extracellular compartment after microvesicle lysis (PubMed:11728343). 5. Release by translocation through permeabilized plasma membrane. This may occur in cells undergoing pyroptosis due to sustained activation of the inflammasome (By similarity). These mechanisms may not be not mutually exclusive.

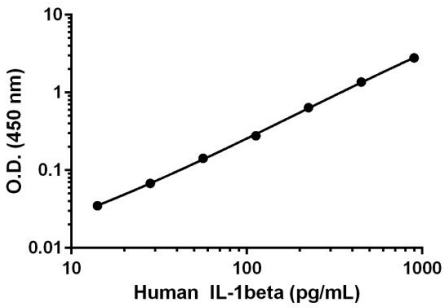
Applications

Our Abpromise guarantee covers the use of **ab214025** in the following tested applications.

The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

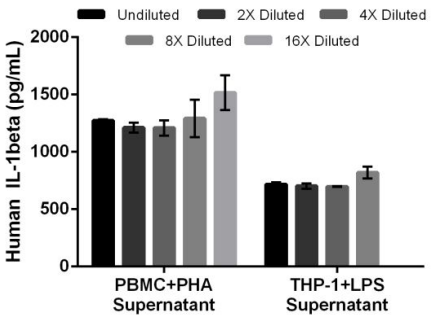
Application	Abreviews	Notes
Sandwich ELISA		Use at an assay dependent concentration.

Images



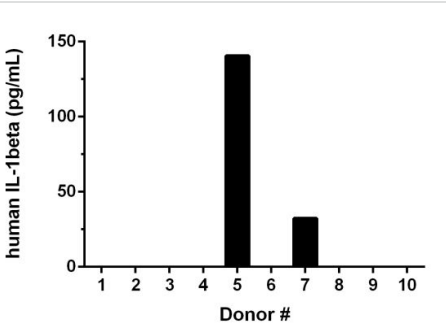
Example of human IL-1beta standard curve.

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



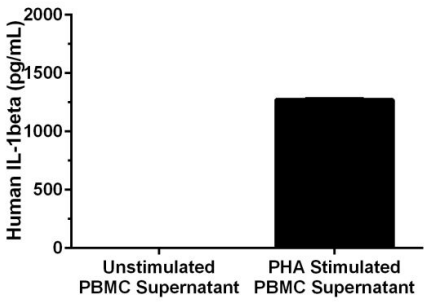
Interpolated concentrations of native IL-1beta in human PHA stimulated PBMC supernatant and LPS stimulated THP-1 supernatant samples.

The concentrations of IL-1beta were measured in duplicates, interpolated from the IL-1beta standard curves and corrected for sample dilution. Undiluted samples are as follows: PBMC supernatant 50% and 100% THP-1 supernatant. The interpolated dilution factor corrected values are plotted (mean +/- SD, n=2). The mean IL-1beta concentration was determined to be 1301 pg/mL in PBMC supernatant and 734 pg/mL in THP-1 supernatant.



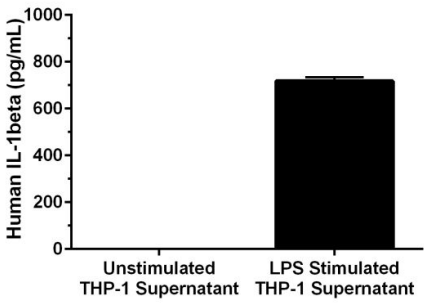
Serum from ten individual healthy human male donors was measured in duplicate.

Interpolated values are plotted (mean +/- SD, n=2). IL-1beta was measured in 2 donor serum samples (30 pg/mL and 140 pg/mL) and the remaining 8 samples measured less than the lowest point of the IL-1beta standard curve.



Human peripheral blood mononuclear cells were cultured unstimulated or stimulated with 10 µg/mL PHA.

Conditioned media was harvested after 48 hours. IL-1beta was measured in 50% unstimulated and PHA stimulated PBMC supernatant. The concentrations of IL-1beta were measured in duplicate, interpolated from the IL-1beta standard curves and corrected for sample dilution. The interpolated dilution factor corrected values are plotted (mean +/- SD, n=2). The mean IL-1beta concentration was determined to be 1273 pg/mL in PHA stimulated PBMC supernatant. There was no detectable signal in unstimulated supernatant.



THP-1 cells were cultured unstimulated or stimulated with 5 µg/mL Lipopolysaccharide (LPS).

Conditioned media was harvested after 48 hours. IL-1beta was measured in 100% unstimulated and LPS stimulated THP-1 supernatant. The concentrations of IL-1beta were measured in duplicate and interpolated from the IL-1beta standard curves. The interpolated values are plotted (mean +/- SD, n=2). The mean IL-1beta concentration was determined to be 718 pg/mL in LPS stimulated THP-1 supernatant. There was no detectable signal in unstimulated supernatant.

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