

# Datasheet for ABIN625141

# IL-1 beta ELISA Kit





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Quantity:	96 tests
Target:	IL-1 beta (IL1B)
Reactivity:	Mouse
Method Type:	Sandwich ELISA
Detection Range:	5-2000 pg/mL
Minimum Detection Limit:	5 pg/mL
Application:	ELISA

## **Product Details**

Purpose:	Mouse IL-1 beta (IL-1 F2) ELISA Kit for cell culture supernatants, plasma, and serum samples.	
Sample Type:	Serum, Plasma, Cell Culture Supernatant	
Analytical Method:	Quantitative	
Detection Method:	Colorimetric	
Specificity:	This ELISA kit shows no cross-reactivity with any of the cytokines tested: Mouse CD30, L CD30, T CD40, CRG-2, CTACK, CXCL16, Eotaxin, Eotaxin-2, Fas Ligand, Fractalkine, GCSF, GM-CFS, IFN- gamma, IGFBP-3, IGFBP-5, IGFBP-6, IL-1 alpha, IL-2, IL-3, IL-3 Rb, IL-4, IL-5, IL-6, IL-9, IL-10, IL-12 p40/p70, IL-12 p70, IL-13, IL-17, KC, Leptin R, LEPTIN(OB), LIX, L-Selectin, Lymphotactin, MCP-1, MCP-5, M-CSF, MIG, MIP-1 alpha, MIP-1 gamma, MIP-2, MIP-3 beta, MIP-3 alpha, PF-4, P-Selectin, RANTES, SCF, SDF-1 alpha, TARC, TCA-3, TECK, TIMP-1, TNF-alpha, TNF RI, TNF RII, TPO, VCAM-1, VEGF.	
Sensitivity:	< 5 pg/mL	

## **Product Details**

#### Characteristics:

- · Strip plates and additional reagents allow for use in multiple experiments
- · Quantitative protein detection
- · Establishes normal range
- · The best products for confirmation of antibody array data

#### Components:

- · Pre-Coated 96-well Strip Microplate
- · Wash Buffer
- · Stop Solution
- Assay Diluent(s)
- · Lyophilized Standard
- · Biotinylated Detection Antibody
- · Streptavidin-Conjugated HRP
- · TMB One-Step Substrate

#### Material not included:

- · Distilled or deionized water
- Precision pipettes to deliver 2 µL to 1 µL volumes
- Adjustable 1-25 µL pipettes for reagent preparation
- 100  $\mu$ L and 1 liter graduated cylinders
- Tubes to prepare standard and sample dilutions
- · Absorbent paper
- · Microplate reader capable of measuring absorbance at 450nm
- · Log-log graph paper or computer and software for ELISA data analysis

## **Target Details**

Target:	IL-1 beta (IL1B)
Alternative Name:	IL-1 beta (IL1B Products)
Background:	The Mouse IL-1beta ELISA (Enzyme-Linked Immunosorbent Assay) kit is an in vitro enzyme-linked immunosorbent assay for the quantitative measurement of mouse IL-1beta in serum, plasma and cell culture supernatants. This assay employs an antibody specific for mouse IL-1beta coated on a 96-well plate. Standards and samples are pipetted into the wells and IL-1beta present in a sample is bound to the wells by the immobilized antibody. The wells are washed and biotinylated anti-mouse IL-1beta antibody is added. After washing away unbound biotinylated antibody, HRP-conjugated streptavidin is pipetted to the wells. The wells are again washed, a TMB substrate solution is added to the wells and color develops in proportion to the amount of IL-1beta bound. The Stop Solution changes the color from blue to yellow, and the intensity of the color is measured at 450 nm. Reproducibility: Intra-Assay: CV<10% Inter-Assay: CV<12%.
Gene ID:	16176

Target Details	
UniProt:	P10749
Pathways:	NF-kappaB Signaling, Interferon-gamma Pathway, TLR Signaling, Negative Regulation of
	Hormone Secretion, Cellular Response to Molecule of Bacterial Origin, Carbohydrate
	Homeostasis, Glycosaminoglycan Metabolic Process, Myometrial Relaxation and Contraction,
	Regulation of Leukocyte Mediated Immunity, Positive Regulation of Immune Effector Process,
	Autophagy, Cancer Immune Checkpoints, Inflammasome
Application Details	
Application Notes:	Recommended Dilution for serum and plasma samples2 fold
Sample Volume:	100 μL
Plate:	Pre-coated
Protocol:	1. Prepare all reagents, samples and standards as instructed in the manual.
	2. Add 100 μL of standard or sample to each well.
	3. Incubate 2.5 h at RT or O/N at 4 °C.
	4. Add 100 $\mu L$ of prepared biotin antibody to each well.
	5. Incubate 1 h at RT.
	6. Add 100 μL of prepared Streptavidin solution to each well.
	7. Incubate 45 min at RT.
	8. Add 100 μL of TMB One-Step Substrate Reagent to each well.
	9. Incubate 30 min at RT.
	10. Add 50 μL of Stop Solution to each well.
	11. Read at 450 nm immediately.

### Reagent Preparation:

- 1. Bring all reagents and samples to room temperature (18 25 °C) before use.
- 2. Sample dilution: If your samples need to be diluted, Assay Diluent A (Item D) should be used for dilution of serum/plasma samples. 1x Assay Diluent B (Item E) should be used for dilution of culture supernatants. Suggested dilution for normal serum/plasma: 2 fold\*. \* Please note that levels of the target protein may vary between different specimens. Optimal dilution factors for each sample must be determined by the investigator.
- 3. Assay Diluent B should be diluted 5-fold with deionized or distilled water.
- 4. Preparation of standard: Briefly spin the vial of Item C. Add 400  $\mu$ L Assay Diluent A (for serum/plasma samples) or 1x Assay Diluent B (for cell culture medium) into Item C vial to prepare a 50 ng/mL standard. Dissolve the powder thoroughly by a gentle mix. Add 40  $\mu$ L IL-1beta standard from the vial of Item C, into a tube with 960.0  $\mu$ L Assay Diluent A or 1x Assay Diluent B to prepare a 2000 pg/mL stock standard solution. Pipette 400  $\mu$ L Assay Diluent A or 1x Assay Diluent B into each tube. Use the stock standard solution to produce a dilution series .

Mix each tube thoroughly before the next transfer. Assay Diluent A or 1x Assay Diluent B serves as the zero standard (0 pg/mL). 200  $\mu$ L 40  $\mu$ L standard +960.0  $\mu$ L 200myl 200  $\mu$ L 200  $\mu$ L 200  $\mu$ L 200  $\mu$ L 2000  $\mu$ L 2000

- 5. If the Wash Concentrate (20x) (Item B) contains visible crystals, warm to room temperature and mix gently until dissolved. Dilute 20 ml of Wash Buffer Concentrate into deionized or distilled water to yield 400 ml of 1x Wash Buffer.
- 6. Briefly spin the Detection Antibody vial (Item F) before use. Add 100  $\mu$ L of 1x Assay Diluent B into the vial to prepare a detection antibody concentrate. Pipette up and down to mix gently (the concentrate can be stored at 4 °C for 5 days). The detection antibody concentrate should be diluted 65-fold with 1x Assay Diluent B and used in step 4 of Part VI Assay Procedure.
- 7. Briefly spin the HRP-Streptavidin concentrate vial (Item G) and pipette up and down to mix gently before use. HRP-Streptavidin concentrate should be diluted 200-fold with 1x Assay Diluent B. For example: Briefly spin the vial (Item G) and pipette up and down to mix gently . Add 75  $\mu$ L of HRP-Streptavidin concentrate into a tube with 15 ml 1x Assay Diluent B to prepare a 200-fold diluted HRP-Streptavidin solution (don't store the diluted solution for next day use). Mix well.

#### Assay Procedure:

- 1. Bring all reagents and samples to room temperature (18 25 °C) before use. It is recommended that all standards and samples be run at least in duplicate.
- 2. Add 100  $\mu$ L of each standard (see Reagent Preparation step 2) and sample into appropriate wells. Cover well and incubate for 2.5 hours at room temperature or over night at 4 °C with gentle shaking.
- 3. Discard the solution and wash 4 times with 1x Wash Solution. Wash by filling each well with Wash Buffer (300 myl) using a multi-channel Pipette or autowasher. Complete removal of liquid at each step is essential to good performance. After the last wash, remove any remaining Wash Buffer by aspirating or decanting. Invert the plate and blot it against clean paper towels.
- 4. Add 100  $\mu$ L of 1x prepared biotinylated antibody (Reagent Preparation step 6) to each well. Incubate for 1 hour at room temperature with gentle shaking.
- 5. Discard the solution. Repeat the wash as in step
- 6. Add 100  $\mu$ L of prepared Streptavidin solution (see Reagent Preparation step 7) to each well. Incubate for 45 minutes at room temperature with gentle shaking.
- 7. Discard the solution. Repeat the wash as in step
- 8. Add 100  $\mu$ L of TMB One-Step Substrate Reagent (Item H) to each well. Incubate for 30 minutes at room temperature in the dark with gentle shaking.
- 9. Add 50 µL of Stop Solution (Item I) to each well. Read at 450 nm immediately.

### **Application Details**

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Calculate the mean absorbance for each set of duplicate standards, controls and samples, and subtract the average zero standard optical density. Plot the standard curve on log-log graph paper or using Sigma plot software, with standard concentration on the x-axis and absorbance on the y-axis. Draw the best-fit straight line through the standard points.

<u>Typical Data:</u> These standard curves are for demonstration only. A standard curve must be run with each assay. Assay Diluent A Mouse IL-1 beta concentration (pg/mL) 1 10 100 1000 10000 O D =4 50 n m 0.01 0.1 1 10 Assay Diluent B Mouse IL-1 beta concentration (pg/mL) 1 10 100 1000 10000 O D =4 50 n m 0.01 0.1 1 10

Sensitivity: The minimum detectable dose of IL-1beta is typically less than 5 pg/mL.

Recovery: Recovery was determined by spiking various levels of mouse IL-1beta into mouse serum, plasma and cell culture media. Mean recoveries are as follows: Sample Type Average % Recovery Range (%) Serum 99.36 89-108 Plasma 99.24 90-109 Cell culture media 98.64 89-110

<u>Linearity:</u> Sample Type Serum Plasma Cell Culture Media 1:2 Average % of Expected 98 97 98 Range (%) 90-106 89-107 88-106 1:4 Average % of Expected 98 96 96 Range (%) 90-108 89-106 91-108

Reproducibility: Intra-Assay: CV<10 % Inter-Assay: CV<12 %

Assay Precision:

Intra-Assay: CV< 10 % Inter-Assay: CV< 12 %

Restrictions:

For Research Use only

### Handling

Handling Advice:	Avoid repeated freeze-thaw cycles.	
Storage:	-20 °C	
Storage Comment:	The entire kit may be stored at -20°C for up to 1 year from the date of shipment. Avoid repeated freeze-thaw cycles. The kit may be stored at 4°C for up to 6 months. For extended storage, it is recommended to store at -80°C.	
Expiry Date:	6 months	

#### **Publications**

Product cited in:

Wang, Ji, Zhang, Zhao, Yan, Zhang, Shen, Yang, Fang, Tian, Zhu, Gong, Zhang, Wei, Wang, Li, Wan, Xie, She, Wang, Huang, Li: "Targeting CASP8 and FADD-like apoptosis regulator ameliorates nonalcoholic steatohepatitis in mice and nonhuman primates." in: **Nature medicine**, Vol. 23, Issue 4, pp. 439-449, (2017) (PubMed).

Weisser, Demel, Stein, Chen-Wichmann, Touzot, Santilli, Sujer, Brendel, Siler, Cavazzana, Thrasher, Reichenbach, Essers, Schwäble, Grez: "Hyperinflammation in patients with chronic granulomatous disease leads to impairment of hematopoietic stem cell functions." in: **The Journal of allergy and clinical immunology**, Vol. 138, Issue 1, pp. 219-228.e9, (2017) (PubMed ).

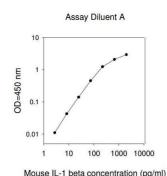
Fang, Yao, Li, Wang, Wang, Chen, Zhou, Liao: "The blockage of the Nogo/NgR signal pathway in microglia alleviates the formation of Aβ plaques and tau phosphorylation in APP/PS1 transgenic mice." in: **Journal of neuroinflammation**, Vol. 13, Issue 1, pp. 56, (2017) (PubMed).

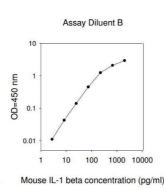
Thomas, Khanam, Vohora: "Augmentation of effect of venlafaxine by folic acid in behavioral paradigms of depression in mice: Evidence of serotonergic and pro-inflammatory cytokine pathways." in: **Pharmacological reports: PR**, Vol. 68, Issue 2, pp. 396-403, (2016) (PubMed).

Nouri, Karkhah, Mohammadzadeh, Sankian: "Elevated caspase-1 activity and IL-1β expression are associated with the IPAF inflammasome in an experimental model of allergy." in: **Molecular medicine reports**, Vol. 13, Issue 4, pp. 3356-62, (2016) (PubMed).

There are more publications referencing this product on: Product page

#### **Images**





### **ELISA**

Image 1.