



IL-1 β (Human) ELISA Kit

(Catalog # E4818-100; 96 assays, Storage at 4°C)

04/20

I. Introduction:

IL-1 β is a potent proinflammatory cytokine. It promotes Th17 differentiation of T-cells and synergizes with IL12/interleukin-12 to induce IFNG synthesis from T-helper 1 (Th1) cells. BioVision's IL-1 β (Human) ELISA kit is based on Sandwich-ELISA principle. The micro ELISA plate provided in this kit has been pre-coated with an antibody specific to Human IL-1 β . Standards or samples are added to the micro ELISA plate wells that bind to the specific antibody. Then a biotinylated detection antibody specific for Human IL-1 β and Avidin-Horseradish Peroxidase (HRP) conjugate are added successively to each micro plate well and incubated. The wells are washed, a TMB substrate solution is added to the wells and blue color develops in proportion to the amount of IL-1 β bound. The enzyme-substrate reaction is terminated by the addition of stop solution and the color turns yellow. The optical density (OD) is measured spectrophotometrically at a wavelength of 450 nm. The OD value is proportional to the concentration of Human IL-1 β . The concentration of Human IL-1 β in the samples can be calculated by comparing the OD of the samples to the standard curve.

II. Applications:

In vitro quantitative determination of IL-1 β concentrations in human serum, plasma and other biological fluids

Sensitivity: 4.69 pg/mL

Detection Range: 7.81-500 pg/mL

Specificity: No Significant cross-reactivity or interference between Human IL-1 β and analogues was observed.

Precision: Coefficient of variation is < 10%.

III. Sample Type:

Serum, plasma and other biological fluids

IV. Kit Contents:

Components	E4818-100	Part Number	Storage
Micro ELISA Plate	8 wells x12 strips	E4818-100-1	-20°C
Reference Standard	2 vials	E4818-100-2	-20°C
Biotinylated Detection Ab (100x)	120 μ l	E4818-100-3	-20°C
HRP Conjugate (100x)	120 μ l	E4818-100-4	-20°C (protect from light)
Reference Standard & Sample Diluent	20 ml	E4818-100-5	4°C
Biotinylated Detection Antibody Diluent	14 ml	E4818-100-6	4°C
HRP Conjugate Diluent	14 ml	E4818-100-7	4°C
Wash Buffer (25X)	30 ml	E4818-100-8	4°C
Substrate Reagent	10 ml	E4818-100-9	4°C (protect from light)
Stop Solution	10 ml	E4818-100-10	4°C
Plate Sealer	5	E4818-100-11	4°C

V. User Supplied Reagents and Equipment:

- Microplate reader capable of measuring absorbance at 450 nm
- Clean Eppendorf tubes for preparing standards or sample dilutions

VI. Storage and Handling:

Store at 4°C.

VII. Reagent and Sample Preparation:

Bring all reagents to room temperature before use. Before using the kit, spin tubes and bring down all components to the bottom of tubes.

- **Wash Buffer (25X):** Dilute 30 ml of Concentrated Wash Buffer with 720 ml of deionized or distilled water to prepare 750 ml of Wash Buffer. (Note: if crystals have formed in the concentrate, warm it in a 40°C water bath and mix it gently until the crystals have completely dissolved)
- **Biotinylated Detection Antibody working solution:** Calculate the required amount (100 μ L/well). Centrifuge the stock tube before use; dilute the 100x Concentrated Biotinylated Detection Antibody to 1x working solution with Biotinylated Detection Antibody Diluent.
- **HRP Conjugate working solution:** Calculate the required amount before the experiment (100 μ L/well). In preparation, slightly more than calculated should be prepared. Dilute the 100x Concentrated HRP Conjugate to 1x working solution with Concentrated HRP Conjugate Diluent.
- **Standard:** Centrifuge the standard at 10,000xg for 1 min. Add 1.0 ml of Standard and Sample Diluent, let it stand for 10 min and invert it gently several times. After it dissolves fully, mix it thoroughly with a pipette. This reconstitution produces a working solution of 500 pg/ml. Then make serial dilutions as needed. The recommended dilution gradient is as follows: 500, 250, 125, 62.5, 31.25, 15.63, 7.81, 0 pg/mL. Prepare 7 tubes, add 500 μ l of Standard and Sample Diluent to each tube. Pipette 500 μ l of the 500 pg/ml stock solution to the



XI. Precision:

Intra-assay Precision (Precision within an assay): 3 samples with low, mid-range and high level Human IL-1 β were tested 20 times on one plate, respectively.

Inter-assay Precision (Precision between assays): 3 samples with low, mid-range and high level Human IL-1 β were tested on 3 different plates, 20 replicates in each plate.

Sample	Intra-assay Precision			Inter-assay Precision		
	1	2	3	1	2	3
n	20	20	20	20	20	20
Mean (pg/mL)	27.70	46.20	197.60	29.10	47.80	192.60
Standard deviation	1.40	2.60	9.50	1.70	2.80	6.00
C V (%)	5.05	5.63	4.81	5.84	5.86	3.12

XII. Recovery:

The recovery of Human IL-1 β spiked at three different levels in samples throughout the range of the assay was evaluated in various matrices.

Sample Type	Range (%)	Average Recovery (%)
Serum (n=5)	94-110	100
EDTA plasma (n=5)	89-102	95
Cell culture media (n=5)	84-98	91

XIII. Linearity:

Samples were spiked with high concentrations of Human IL-1 β and diluted with Reference Standard & Sample Diluent to produce samples with values within the range of the assay.

		Serum (n=5)	EDTA plasma (n=5)	Cell culture media (n=5)
1:2	Range (%)	95-109	88-100	94-108
	Average (%)	102	94	102
1:4	Range (%)	95-109	82-94	93-105
	Average (%)	100	88	99
1:8	Range (%)	98-110	82-95	97-109
	Average (%)	105	88	103
1:16	Range (%)	96-108	80-91	97-113
	Average (%)	102	86	103

XIV. RELATED PRODUCTS:

- IL-1 beta, rat recombinant (4130)
- IL-1 beta, human recombinant (4128)
- IL-1 beta, murine recombinant (4129)
- IL-1 β (rat) ELISA Kit (K4796)

FOR RESEARCH USE ONLY! Not to be used on humans.