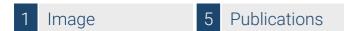


# Datasheet for ABIN625012

# **IL-15 ELISA Kit**





## Overview

Quantity: Target: Reactivity:	96 tests  IL-15 (IL15)  Human  Sandwich ELISA
	Human
Reactivity:	
	Sandwich ELISA
Method Type:	
Detection Range:	3-800 pg/mL
Minimum Detection Limit:	3 pg/mL
Application:	ELISA
Product Details	
Purpose:	Human IL-15 ELISA Kit for cell culture supernatants, Heparin and/or EDTA treated plasma, and serum samples. Citrate is not recommended.
Sample Type:	Plasma, Cell Culture Supernatant, Serum
Analytical Method:	Quantitative
Detection Method:	Colorimetric
Specificity:	This ELISA kit shows no cross-reactivity with any of the cytokines tested: Human Angiogenin, BDNF, BLC, ENA-78, FGF-4, IL-1 alpha, IL-1 beta, IL-2, IL-3, IL-4, IL-5, IL-7, IL-8, IL-9, IL-10, IL-11, IL-12 p70, IL-12 p40, IL-13, I-309, IP-10, G-CSF, GM-CSF, IFN-gamma, Leptin, MCP-1, MCP-2, MCP-3, MDC, MIP-1 alpha, MIP-1 beta, MIP-1 delta, PARC, PDGF, RANTES, SCF, TARC, TGF-beta, TIMP-1, TIMP-2, TNF-alpha, TNF-beta, TPO, VEGF.
Sensitivity:	3 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 μL 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-15 (IL15)
Alternative Name:	IL-15 (IL15 Products)
Background:	IL-15 (Interleukin-15) is found in conditioned medium of a monkey kidney epithelial cell line, CV-1/EBNA. It is a glycoprotein of 14-15 kDa . Human IL-15 has been cloned from a human stromal cell line, IMTLH. Stromal cell line IL-15 has been shown to be produced by human fetal

1/EBNA. It is a glycoprotein of 14-15 kDa . Human IL-15 has been cloned from a human stromal cell line, IMTLH. Stromal cell line IL-15 has been shown to be produced by human fetal astrocytes and microglia in response to IL-1beta or IFN-gamma and may thus play a role in T-cell mediated immune responses in the human central nervous system. IL-15 is a recently identified novel cytokine that shares many biological properties with IL-2, including T-Cell, B-Cell and NK-cell-stimulatory activities. The Human IL-15 ELISA (Enzyme-Linked Immunosorbent Assay) kit is an in vitro enzyme-linked immunosorbent assay for the quantitative measurement of human IL-15 in serum, plasma (collect plasma using EDTA or heparin as an anticoagulant. Citrate plasma is not recommended for use in this assay), cell culture supernatants and urine. This assay employs an antibody specific for human IL-15 present in a sample is bound to the wells by

the immobilized antibody. The wells are washed and biotinylated anti-human IL-15 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-15 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: 3600

UniProt: P40933

Pathways: JAK-STAT Signaling, Glycosaminoglycan Metabolic Process

# **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  $\mu L$  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  $\mu L$  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) is used for dilution of serum/plasma samples, and Assay Diluent B (Item E) is used for dilution of culture supernatants and urine. 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 and urine) into Item C vial to prepare a 50 ng/ml standard. Dissolve the powder thoroughly by a gentle mix. Add 12  $\mu$ l IL-15 standard from the vial of Item C, into a tube with 738  $\mu$ l Assay Diluent A or 1x Assay Diluent B to prepare a 800 pg/ml stock standard solution. Pipette 300  $\mu$ l Assay Diluent A or 1x

Assay Diluent B into each tube. Use the stock standard solution to produce a dilution series (shown below). Mix each tube thoroughly before the next transfer. Assay Diluent A or 1x Assay Diluent B serves as the zero standard (0 pg/ml). 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 µl 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 80-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 10,000-fold with 1x Assay Diluent B. For example: Briefly spin the vial (Item G) and pipette up and down to mix gently. Add 2 µl of HRP-Streptavidin concentrate into a tube with 198.0 µl 1x Assay Diluent B to prepare a 100-fold diluted HRP-Streptavidin solution (do not store the diluted solution for next day use). Mix through and then pipette 120 µl of prepared 100-fold diluted solution into a tube with 12 ml 1x Assay Diluent B to prepare a final 10,000 fold diluted HRP-Streptavidin solution.

Assay Procedure:

1. Bring all reagents and samples to room temperature ( $18 - 25^{\circ}$ 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 ( $300 \,\mu$ l) 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 3. 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 3. 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  $50 \,\mu$ l of Stop Solution (Item I) to each well. Read at 450 nm immediately.

Calculation of Results:

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.

### **Application Details**

Restrictions:	For Research Use only
Handling	
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:

Shoman, Nabil, Tabl, Ghanem, Kafrawy: "Assessment of immunological changes in Epstein-Barr virus co-infection in Egyptian chronic HCV patients." in: **Memórias do Instituto Oswaldo Cruz**, Vol. 109, Issue 6, pp. 722-7, (2014) (PubMed).

Pfeiffer, Hoyer, Gerer, Voll, Knippertz, Gückel, Schuler, Schaft, Dörrie: "Triggering of NF-?B in cytokine-matured human DCs generates superior DCs for T-cell priming in cancer immunotherapy." in: **European journal of immunology**, Vol. 44, Issue 11, pp. 3413-28, (2014) (PubMed).

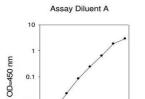
Gong, Liu, Chen, Xu, Lu, Jin: "Insights into the paracrine effects of uterine natural killer cells." in: **Molecular medicine reports**, Vol. 10, Issue 6, pp. 2851-60, (2014) (PubMed).

Lu, Bocca, Anderson, Wang, Manhua, Beydoun, Oehninger: "Modulation of the expression of the transcription factors T-bet and GATA-3 in immortalized human endometrial stromal cells (HESCs) by sex steroid hormones and cAMP." in: **Reproductive sciences (Thousand Oaks, Calif.)**, Vol. 20, Issue 6, pp. 699-709, (2013) (PubMed).

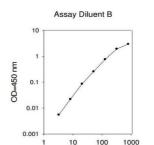
Cuchacovich, Hagan, Khan, Richert, Espinoza: "Tumor necrosis factor-alpha (TNF- $\alpha$ )-blockade-induced hepatic sarcoidosis in psoriatic arthritis (PsA): case report and review of the literature." in: **Clinical rheumatology**, Vol. 30, Issue 1, pp. 133-7, (2011) (PubMed).

0.01

0.001







Human IL-15 concentration (pg/ml)

## **ELISA**

#### Image 1.