

Datasheet for ABIN625023

IL28A ELISA Kit





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Quantity:	96 tests
Target:	IL28A
Reactivity:	Human
Method Type:	Sandwich ELISA
Detection Range:	15-10000 pg/mL
Minimum Detection Limit:	15 pg/mL
Application:	ELISA
Product Details	
Purpose:	Human IL-28A (IFN-lambda 2) ELISA Kit for cell culture supernatants, plasma, and serum samples.
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-6, IL-7, IL-8, IL-9, IL-10, IL-11, IL-12 p70, IL-12 p40, IL-13, IL-15, IL-16, IL-17, IL-18, IL-29, I-309, IP-10, GCSF, GM-CSF, IFN-gamma, Leptin (OB), MCP-1, MCP-3, MDC, MIP-1 alpha, MIP-1 beta, MIP-1 delta, MMP-1, -2, -3, -10, PARC, RANTES, SCF, TARC, TGF-beta, TIMP-1, TIMP-2, TNF-alpha, TNF-beta, TPO, VEGF.
Sensitivity:	< 15 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:	IL28A	
Alternative Name:	IL-28A / IFN-lambda (IL28A Products)	
Background:	The Human IL-28A/IFN-lambda-2 (Interleukin 28A/ Interferon-lambda-2) ELISA (Enzyme-Linke Immunosorbent Assay) kit is an in vitro enzyme-linked immunosorbent assay for the quantitative measurement of human IL-28A in serum, plasma, cell culture supernatants and urine. This assay employs an antibody specific for human IL-28A coated on a 96-well plate. Standards and samples are pipetted into the wells and IL-28A present in a sample is bound to the wells by the immobilized antibody. The wells are washed and biotinylated anti-human IL-28A 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-28A 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:	282616	

UniProt:

Q8IZJ0

Application Details

Application Notes:		
Sample Volume:		
Plate:		
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 μL 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 C (Item L) should be used for dilution of serum/plasma/culture supernatants/urine. 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 (Item E) should be diluted 5-fold with deionized or distilled water before use.
- 4. Preparation of standard: Briefly spin the vial of Item C. Add 400 μ L Assay Diluent C (Item L) into Item C vial to prepare a 100 ng/mL standard solution. Dissolve the powder thoroughly by a gentle mix. Add 50 μ L IL-28A standard from the vial of Item C, into a tube with 450 μ L Assay Diluent C to prepare a 10,000 pg/mL standard solution. Pipette 400 μ L Assay Diluent C into each tube. Use the stock standard solution to produce a dilution series . Mix each tube thoroughly before the next transfer. Assay Diluent C serves as the zero standard (0 pg/mL). 50 μ L standard + 450 μ L 200 μ L pg/mL p
- 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

(Item E) 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 300-fold with 1x Assay Diluent B (Item E). For example: Briefly spin the vial (Item G) and pipette up and down to mix gently . Add 40 μ L of HRP-Streptavidin concentrate into a tube with 12 ml 1x Assay Diluent B to prepare a final 300 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 μ 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 μ 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 μ 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 μ 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.

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.

Sensitivity: The minimum detectable dose of IL-28A is typically less than 15 pg/mL.
Recovery: Recovery was determined by spiking various levels of IL-28A into normal human
serum, plasma and cell culture media. Mean recoveries are as follows: Sample Type Average %
Recovery Range (%) Serum 107.0 84-111 Plasma 110.4 84-120 Cell culture media 105.2 95-
114
<u>Linearity:</u> Sample Type Serum Plasma Cell Culture Media 1:2 Average % of Expected 99.72
101.5 97.31 Range (%) 89-107 90-108 87-106 1:4 Average % of Expected 85.88 93.51 88.27
Range (%) 78-99 85-105 80-100
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

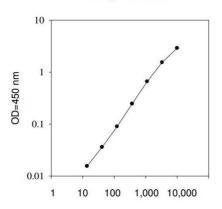
Handling Advice:	Avoid repeated freeze-thaw cycles.
Storage:	-20 °C
Storage Comment:	The entire kit may be stored at -20°C for up to 1 year from the date of shipment. Avoid repeated freeze-thaw cycles. The kit may be stored at 4°C for up to 6 months. For extended storage, it is recommended to store at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

Publications

Product cited in:

Stone, Giugliano, Schnell, Cheng, Leahy, Golden-Mason, Gale, Rosen: "Hepatitis C virus pathogen associated molecular pattern (PAMP) triggers production of lambda-interferons by human plasmacytoid dendritic cells." in: **PLoS pathogens**, Vol. 9, Issue 4, pp. e1003316, (2013) (PubMed).

Assay Diluent C



Human IL-28A concentration (pg/ml)

ELISA

Image 1.