

Human IL-29 Sandwich ELISA Kit Datasheet

For the quantitative detection of Human IL-29 in in serum, plasma, cell culture supernatants.

General Information

Catalogue Number	KE00241
Product Name	AuthentiKine™ Human IL-29 Sandwich ELISA Kit
Species cross-reactivity	Human
Range (calibration Range)	62.5 -4000 pg/mL
Tested applications	Quantification ELISA

Database Links

Entrez Gene	282618
SwissProt	Q8IU54

Kit Components & Storage

Microplate - antibody coated 96-well microplate (8 well × 12 strips)	1 plate	Unopened Kit: Store at 2-8°C for 6 months or -20°C for 12 months. Opened Kit: All reagents stored at 2-8°C for 7 days. Please use a new standard for each assay.
Protein standard - 8000 pg/bottle; lyophilized*	2 bottles	
Detection antibody, HRP-conjugated (100×) - 120 µL/vial	1 vial	
Sample Diluent PT 4B1 - 30 mL/bottle. For Human serum and plasma	1 bottle	
Sample Diluent PT 1-ef - 30 mL/bottle. For cell culture supernatants.	1 bottle	
Detection Diluent - 30 mL/bottle	1 bottle	
Wash Buffer Concentrate (20×) - 30 mL/bottle	1 bottle	
Tetramethylbenzidine Substrate (TMB) - 12 mL/bottle	1 bottle	
Stop Solution - 12 mL/bottle	1 bottle	
Plate Cover Seals	3 pieces	

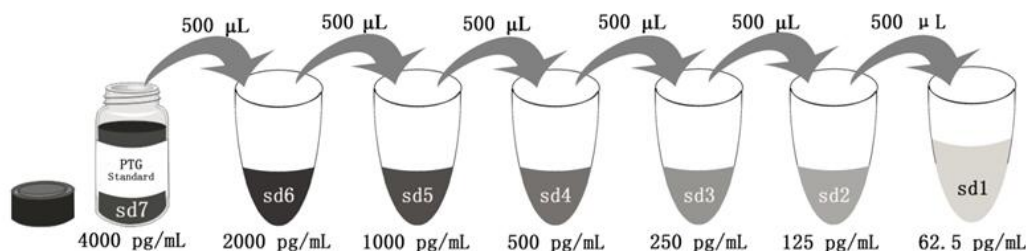
NB: Do not use the kit after the expiration date.

Sample Diluent PT 4B1 is for protein standard, serum and plasma samples. .

Sample Diluent PT 1-ef is for protein standard and cell culture supernatants .

Detection Diluent is for Detection antibody .

*Add 2 mL Sample Diluent PT 4B1 or PT 1-ef in protein standard. This reconstitution gives a stock solution of 4000 pg/mL.



Add # µL of Standard diluted in the previous step	—	500 µL	500 µL	500 µL	500 µL	500 µL	500 µL
# µL of Sample Diluent PT 4B1 or PT 1-ef	2000 µL	500 µL	500 µL	500 µL	500 µL	500 µL	500 µL
	"sd7"	"sd6"	"sd5"	"sd4"	"sd3"	"sd2"	"sd1"

Product Description

KE00241 is a solid phase sandwich Enzyme Linked-Immuno-Sorbent Assay (Sandwich ELISA). The IL-29 ELISA kit is to be used to detect and quantify protein levels of endogenous IL-29. The assay recognizes human IL-29. An antibody specific for IL-29 has been pre-coated onto the microwells. The IL-29 protein in samples is captured by the coated antibody after incubation. Following extensive washing, another horseradish peroxidase (HRP)-conjugated antibody specific for IL-29 is added to detect the captured IL-29 protein. For signal development, followed by Tetramethyl-benzidine (TMB) reagent. Solution containing sulfuric acid is used to stop color development and the color intensity which is proportional to the quantity of bound protein is measurable at 450 nm with the correction wavelength set at 630 nm.

Background

Interleukin-29 (IL-29) is a cytokine belonging to the Type III interferon family, which has another two subfamilies IL-28A and IL-28B. They are also known as IFN- 1, IFN- 2 and IFN- 3, respectively. IL-29 is produced predominantly by maturing dendritic cells and macrophages. IL-29 plays an important role in the immune response against pathogens and especially against viruses by mechanisms similar to type I interferons. IL-29 receptor signals through JAK-STAT pathways leading to activated expression of interferon-stimulated genes and production of antiviral proteins.

Sample Preparation

Samples may require proper dilution to fall within the range of the assay. 1:2 dilution is recommended for serum or plasma. 1:2 or 1:4 dilution is recommended for cell culture supernatants.

Safety Notes

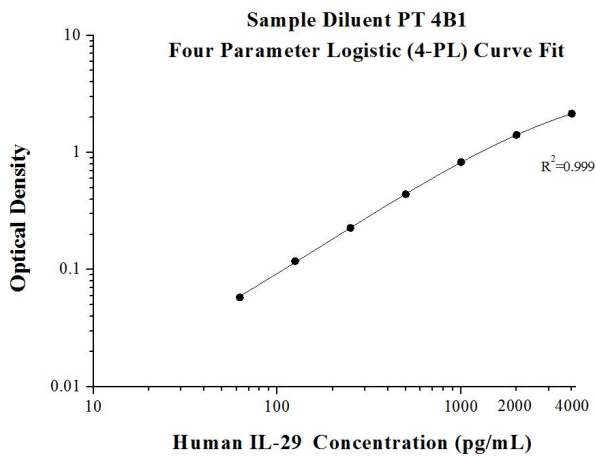
This product is sold for lab research and development use ONLY and not for use in humans or animals. Avoid any skin and eye contact with Stop Solution and TMB. In case of contact, wash thoroughly with water.

Assay Procedure Summary

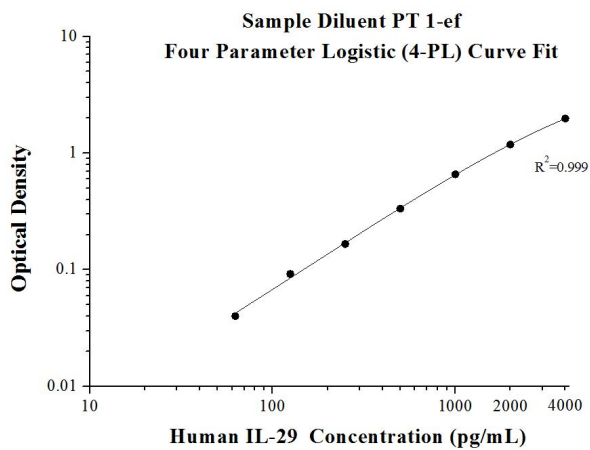
Step	Reagent	Volume	Incubation	Wash	Notes
1	Standard and Samples	100 µL	120 min	4 times	Cover Wells incubate at 37°C
2	Diluent Detection antibody, HRP-conjugated Solution	100 µL	60 min	4 times	Cover Wells incubate at 37°C
3	TMB Substrate	100 µL	15-20 min	Do not wash	Incubate in the dark at 37°C
4	Stop Solution	100 µL	0 min	Do not wash	-
5	Read plate at 450 nm and 630 nm immediately after adding Stop solution. DO NOT exceed 5 minutes.				

Example data

These standard curves are provided for demonstration only. A standard curve should be generated for each set of samples assayed.



(pg/mL)	O.D	Average	Corrected
0	0.106 0.09	0.098	0
62.5	0.159 0.152	0.156	0.058
125	0.216 0.216	0.216	0.118
250	0.337 0.312	0.325	0.227
500	0.554 0.523	0.539	0.441
1000	0.941 0.911	0.926	0.828
2000	1.527 1.493	1.51	1.412
4000	2.278 2.213	2.246	2.148



(pg/mL)	O.D	Average	Corrected
0	0.085 0.097	0.091	0
62.5	0.128 0.134	0.131	0.04
125	0.178 0.187	0.183	0.092
250	0.239 0.274	0.257	0.166
500	0.417 0.433	0.425	0.334
1000	0.745 0.752	0.749	0.658
2000	1.249 1.302	1.276	1.185
4000	2.045 2.092	2.069	1.978

Precision

Intra-assay Precision (Precision within an assay) Three samples of known concentration were tested 20 times on one plate to assess intra-assay precision.

Inter-assay Precision (Precision between assays) Three samples of known concentration were tested in 24 separate assays to assess inter-assay precision.

Intra-assay Precision				
Sample	n	Mean (pg/mL)	SD	CV%
1	20	229.0	9.73	4.2
2	20	878.2	24.62	2.8
3	20	3674.4	139.35	3.8

Inter-assay Precision				
Sample	n	Mean (pg/mL)	SD	CV%
1	24	212.1	8.67	4.1
2	24	869.2	25.66	3.0
3	24	3569.6	145.19	4.1

Recovery

The recovery of IL-29 spiked to three different levels in four samples throughout the range of the assay in various matrices was evaluated.

Sample Type		Average% of Expected	Range (%)
Human plasma	1:2	91	80-105
	1:4	93	72-108
Cell culture supernatants	1:2	104	98-109
	1:4	101	90-121

Sample Values

Serum -Sixteen serum samples from healthy volunteers were evaluated for IL-29 in this assay. No medical histories were available for the donors used in this study.

Sample Type	Mean of Detectable (pg/mL)	%Detectable	Range (pg/mL)
Human serum (n=16)	185.8	62.5	27-744

HUVEC cells were stimulated by adding human IFN-alpha at 100 ng/ml for 20 hours, followed by the addition of 30 µg/mL Poly(I:C) for 3 hours. Aliquots of the cell culture supernates were removed and assayed for levels of human IL-29.

Condition	(pg/mL)
Unstimulated	46.8
Stimulated	378.1

Sensitivity

The minimum detectable dose of IL-29 is 3.3 pg/mL. This was determined by adding two standard deviations to the concentration corresponding to the mean O.D. of 20 zero standard replicates.

Linearity

To assess the linearity of the assay, three samples were spiked with high concentrations of IL-29 in various matrices and diluted with the appropriate Sample Diluent to produce samples with values within the dynamic range of the assay.

		Human plasma (PT 4B1)	Cell culture supernatants (PT 1-ef)
1:2	Average% of Expected	104	78
	Range (%)71-86	84-124	71-86
1:4	Average% of Expected	100	82
	Range (%)	76-124	74-90
1:8	Average% of Expected	88	83
	Range (%)	73-103	76-90
1:16	Average% of Expected	77	98
	Range (%)	69-86	80-115

References

1. Yoshitaka Hosokawa. et al. (2017) Immunol Invest. 46(6):615-624.
2. Katrin Witte et al. (2010) Cytokine Growth Factor Rev. 21(4):237-51.
3. Noah E Kelm. et al. (2016) Crit Rev Oncol Hematol. 106:91-8.
4. Jia-Min Wang. et al. (2019) J Cell Mol Med. 23(12):7926-7932.