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colorimetric sandwich ELISA kit datasheet

For the quantitative detection of human CD13 in serum, plasma, cell culture supernatants.

general information

Catalogue Number	KE00083
Product Name	CD13 ELISA Kit
Species cross-reactivity	Human CD13
Range (calibration Range)	0.313 - 20 ng/mL
Tested applications	Quantification ELISA

database links

Entrez Gene	**290** (Human)
SwissProt	**P15144** (Human)

kit components & storage

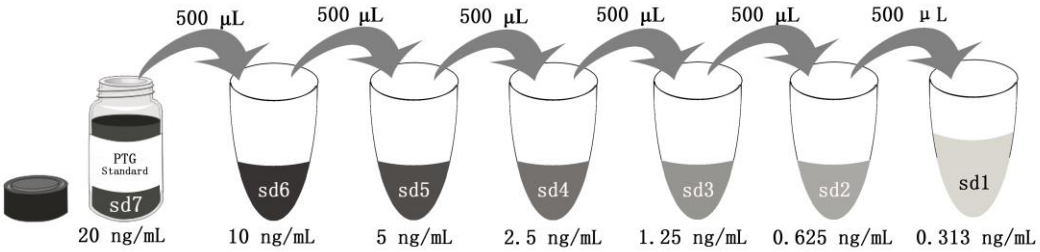
Microplate - antibody coated 96-well Microplate (8 wells ×12 strips)	1 plate	Store at -20°C for six months
Standard - 40 ng/bottle; lyophilized*	2 bottles	Store at -20°C for six months
Detection Antibody (100X) - 150 µL/vial	1 vial	Store at 2-8°C for six months
HRP-conjugated antibody (100X) - 150 µL /vial	1 vial	Store at 2-8°C for six months
Sample Diluent PT 3-ec- 30 mL/bottle	1 bottle	Store at 2-8°C for six months
Detection Diluent - 30 mL/bottle	1 bottle	Store at 2-8°C for six months
Wash Buffer Concentrate (20X) - 30 mL/bottle	1 bottle	Store at 2-8°C for six months
Tetramethylbenzidine Substrate (TMB) - 12 mL/bottle	1 bottle	Store at 2-8°C for six months
Stop Solution - 12 mL/bottle	1 bottle	Store at 2-8°C for six months
Plate Cover Seals	3 pieces	

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

Sample Diluent PT 3-ec is for Standard and serum, plasma, cell culture supernatants samples.

Detection Diluent is for Detection antibody and HRP-conjugated antibody.

*Add 2 mL Sample Diluent PT 3-ec in Standard, This reconstitution gives a stock solution of 20 ng/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 3-ec	2000 µL	500 µL	500 µL	500 µL	500 µL	500 µL	500 µL
	"sd7"	"sd6"	"sd5"	"sd4"	"sd3"	"sd2"	"sd1"

product description

KE00083 is a solid phase sandwich Enzyme Linked-Immuno-Sorbent Assay (Sandwich ELISA). The CD13 ELISA kit is to be used to detect and quantify protein levels of endogenous CD13. The assay recognizes human CD13. A polyclonal antibody specific for CD13 has been pre-coated onto the microwells. The CD13 protein in samples is captured by the coated antibody after incubation. Following extensive washing, a monoclonal antibody specific for CD13 is added to detect the captured CD13 protein. For signal development, horseradish peroxidase (HRP)-conjugated Anti-mouse antibody is added, 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 450nm.

background

CD13 is also named as APN, ANPEP(aminopeptidase N), PEPN and belongs to the peptidase M1 family. It is a 150 kDa cell surface glycoprotein originally identified on subsets of normal and malignant human myeloid cells CD13 reduced ROS-induced DNA damage after genotoxic chemo/radiation stress and protected cells from apoptosis. CD13 is identified as a protein marker correlating with an enrichment of dormant, slow-growing malignant stem cells, with most CD13+ cells resting in the G1/G0 phase of the cell cycle.

sample preparation

The serum or plasma samples may require proper dilution to fall within the range of the assay. A range of dilutions like 1:2, 1:4 is suggested according to the individual samples.

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.

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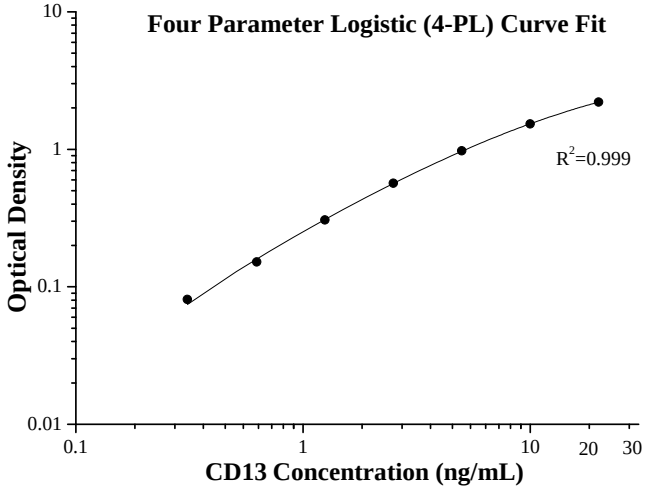
assay procedure summary

Step	Reagent	Volume	Incubation	Wash	Notes
1	Standard and Samples	100 µL	60 min	4 times	Cover Wells
2	Diluent Antibody Solution	100 µL	60 min	4 times	Cover Wells
3	Diluent HRP Solution	100 µL	40 min	4 times	Cover Wells
4	TMB Substrate	100 µL	15-30 min	Do not wash	Incubate in the dark at 37°C
5	Stop Solution	100 µL	0 min	Do not wash	-
6	Read plate at 450 nm and 630 nm immediately after adding Stop solution. DO NOT exceed 5 minutes.				

typical data

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



(ng/mL)	O.D	Average	Corrected
0	0.016 0.022	0.019	—
0.313	0.105 0.095	0.1	0.081
0.625	0.173 0.168	0.1705	0.1515
1.25	0.341 0.31	0.3255	0.3065
2.5	0.594 0.578	0.586	0.567
5	1.022 0.965	0.9935	0.9745
10	1.589 1.508	1.5485	1.5295
20	2.287 2.167	2.227	2.208

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			Inter-assay Precision		
Sample	1	2	3	1	2	3
n	20	20	20	24	24	24
Mean (ng/ml)	5.69	1.53	0.17	5.09	1.53	0.18
SD	0.46	0.10	0.01	0.32	0.08	0.01
CV%	8.2	6.3	8.4	6.3	5.0	7.3

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recovery

The recovery of CD13 spiked to three different levels in four samples throughout the range of the assay in vrious matrices was evaluated.

Sample Type		Average % of Expected	Range(%)
Citrate plasma	1:2	94	79-105
	1:4	88	84-90
Cell culture supernatants	1:2	110	97-127
	1:4	102	83-119

sensitivity

The minimum detectable dose of human CD13 is 0.058ng/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 CD13 in various matrices and diluted with the appropriate Sample Diluent to produce samples with values within the dynamic range of the assay.
(The samples were initially diluted 1:1)

		Citrate plasma	Cell culture supernatants
1:2	Average% of Expected	103	108
	Range(%)	101-105	105-110
1:4	Average% of Expected	108	111
	Range(%)	104-120	101-121
1:8	Average% of Expected	107	110
	Range(%)	93-117	102-117
1:16	Average% of Expected	106	89
	Range(%)	94-119	89

references

1. Satish P. R, et al. Proteomic Analysis of Urine Exosomes Reveals Renal Tubule Response to Leptospiral Colonization in Experimentally Infected Rats. PLoS Negl Trop Dis. 2015 Mar 20;9(3):e0003640.

2. Haraguchi N, Ishii H, et al. CD13 is a therapeutic target in human liver cancer stem cells. J Clin Invest 2010;120: 3326-3339.

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