Human HDAC1 Sandwich ELISA Kit Datasheet

For the quantitative detection of Human HDAC1 concentrations in serum, plasma.

General Information

Catalogue Number	KE00033
Product Name	Human HDAC1 Sandwich ELISA Kit
Species cross-reactivity	Human
Range (calibration Range)	62.5-4000 pg/mL
Tested applications	Quantification ELISA

Database Links

Entrez Gene	3065
SwissProt	Q13547

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 (100X) - 120 μL/vial	1 vial	
HRP-conjugated antibody (HRP) (100X) - 120 μL/vial	1 vial	
Sample Diluent PT 1-ag - 30 mL/bottle	1 bottle	
Detection Diluent - 30 mL/bottle	1 bottle	
Wash Buffer Concentrate (20X) - 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	

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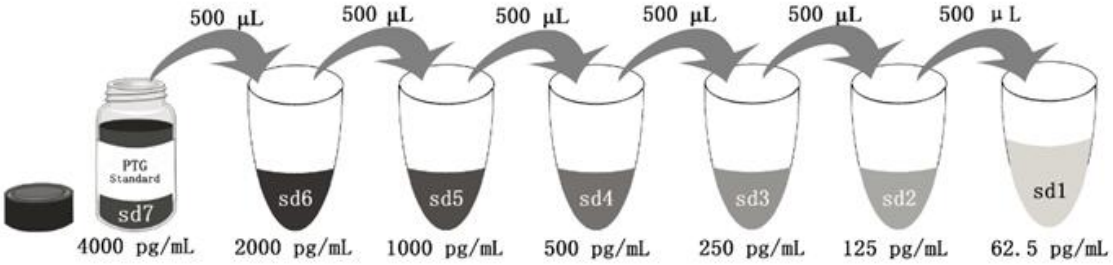
NB: Do not use the kit after the expiration date.
Sample Diluent PT 1-ag is for protein standard and samples.
Detection Diluent is for Detection antibody and HRP-conjugated antibody.
*Add **2 mL** Sample Diluent PT 1-ag in protein standard. This reconstitution gives a stock solution of 4000 pg/mL.



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 1-ag	**2000 µL**	500 µL	500 µL	500 µL	500 µL	500 µL	500 µL
	"sd7"	"sd6"	"sd5"	"sd4"	"sd3"	"sd2"	"sd1"

Product Description

KE00033 is a solid phase sandwich Enzyme Linked-Immuno-Sorbent Assay (Sandwich ELISA). The HDAC1 ELISA kit is to be used to detect and quantify protein levels of endogenous HDAC1. The assay recognizes human HDAC1. An antibody specific for HDAC1 has been pre-coated onto the microwells. The HDAC1 protein in samples is captured by the coated antibody after incubation. Following extensive washing, another antibody specific for HDAC1 is added to detect the captured HDAC1 protein. For signal development, horseradish peroxidase (HRP)-conjugated 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 450 nm with the correction wavelength set at 630 nm.

Background

Histone deacetylases (HDAC) are a class of enzymes that remove the acetyl groups from the lysine residues leading to the formation of a condensed and transcriptionally silenced chromatin. The protein encoded by this gene belongs to the histone deacetylase/ acuc/ apha family and is a component of the histone deacetylase complex, which is responsible for gene expression silencing. It also plays an important role in the control of cell proliferation and differentiation by interacting with RB, p53 and other transcription factors. HADC inhibition causes apoptosis in tumor cells and HDAC inhibitors may be developed as anti-cancer agents. This kit is used to quantify HDAC1 level in vivo.

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.

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Safety Notes

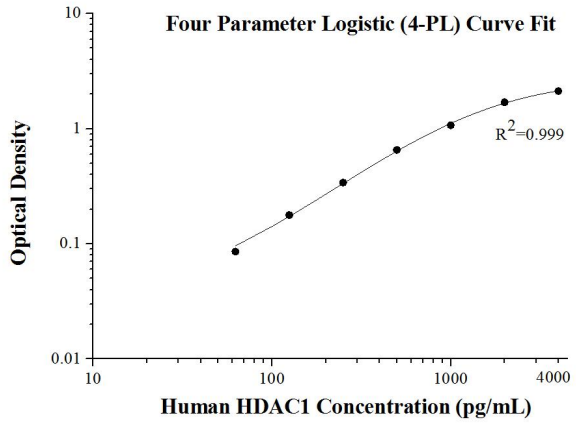
This product is sold for lab research and development use ONLY and not for use in humans or animals. Avoid any skin and eye contact with Stop Solution and TMB. In case of contact, wash thoroughly with water.

Assay Procedure Summary

Step	Reagent	Volume	Incubation	Wash	Notes
1	Standard and Samples	100 µL	60 min	4 times	Cover Wells incubate at 37°C
2	Diluent Antibody Solution	100 µL	60 min	4 times	Cover Wells incubate at 37°C
3	Diluent HRP Solution	100 µL	40 min	4 times	Cover Wells incubate at 37°C
4	TMB Substrate	100 µL	15-20 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.				

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	0.042 0.04	0.041	-
62.5	0.131 0.122	0.1265	0.0855
125	0.215 0.222	0.2185	0.1775
250	0.383 0.38	0.3815	0.3405
500	0.695 0.696	0.6955	0.6545
1000	1.103 1.123	1.113	1.072
2000	1.692 1.78	1.736	1.695
4000	2.163 2.158	2.1605	2.1195

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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	2,406.1	197.3	8.2
2	20	499.5	19.9	4.0
3	20	163.5	6.3	3.9

Inter-assay Precision				
Sample	n	Mean (pg/mL)	SD	CV%
1	24	2,282.9	158.9	7.0
2	24	484.8	24.5	5.1
3	24	157.3	6.9	4.4

Recovery

The recovery of HDAC1 spiked to three different levels in four samples throughout the range of the assay in human plasma averaged 100%, ranging from 86% - 127%.

Sample Values

Twenty-four serum and plasma samples from healthy volunteers were evaluated for human HDAC1 in this assay. All samples measured less than the lowest standard, 62.5 pg/mL. No medical histories were available for the donors used in this study.

Sensitivity

The minimum detectable dose of human HDAC1 is 11.0 pg/mL. This was determined by adding two standard deviations to the concentration corresponding to the mean O.D. of 20 zero standard replicates.

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Linearity

To assess the linearity of the assay, three samples were spiked with high concentrations of HDAC1 in human plasma and diluted with the appropriate **Sample Diluent PT 1-ag** to produce samples with values within the dynamic range of the assay. (The samples were initially diluted 1:1)

		Human plasma
1:2	Average% of Expected	82
	Range (%)	80-84
1:4	Average% of Expected	89
	Range (%)	87-91
1:8	Average% of Expected	102
	Range (%)	88-116
1:16	Average% of Expected	103
	Range (%)	91-118

References

1. McLaughlin F, et al. Histone deacetylase inhibitors open new doors in cancer therapy. Biochem Pharmacol. 68:1139-44 (2004).

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