

colorimetric sandwich ELISA kit datasheet

For the quantitative detection of human FKBPL in serum and plasma.

general information

Catalogue Number	KE00023
Product Name	FKBPL ELISA Kit
Species cross-reactivity	Human FKBPL
Range (calibration Range)	31.25 - 2000 pg/mL
Tested applications	Quantification ELISA

database links

Entrez Gene	63943 (Human)
SwissProt	Q9UIM3 (Human)

kit components & storage

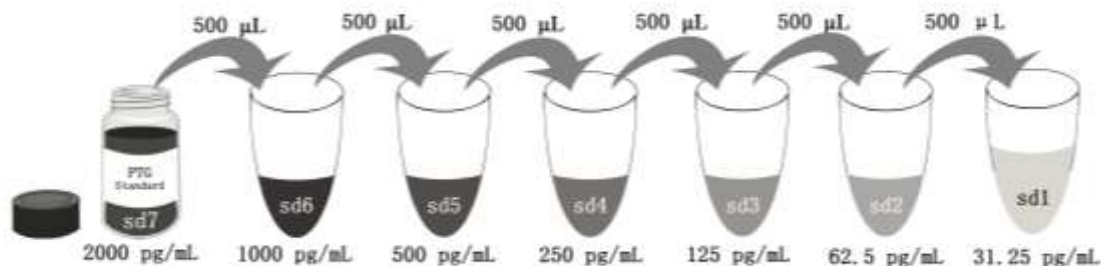
Microplate - antibody coated 96-well Microplate (8 wells × 12 strips)	1 plate	Store at 2-8°C for six months
Standard - 4000 pg/bottle; lyophilized*	2 bottles	Store at 2-8°C for six months
Detection antibody (100X) - 120 µL/vial	1 vial	Store at 2-8°C for six months
HRP-conjugated antibody (100X) - 120 µL/vial	1 vial	Store at 2-8°C for six months
Sample Diluent PT 1-a - 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 1-a is for standard and samples.

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

*Add **2 mL** Sample Diluent PT 1-a in standard. This reconstitution gives a stock solution of 2000 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-a	2000 µL	500 µL	500 µL	500 µL	500 µL	500 µL	500 µL
	"sd7"	"sd6"	"sd5"	"sd4"	"sd3"	"sd2"	"sd1"

product description

KE00023 is a solid phase sandwich Enzyme Linked-Immuno-Sorbent Assay (Sandwich ELISA). The FKBPL ELISA kit is to be used to detect and quantify protein levels of endogenous FKBPL. The assay recognizes human FKBPL. An antibody specific for FKBPL has been pre-coated onto the microwells. The FKBPL protein in samples is captured by the coated antibody after incubation. Following extensive washing, another antibody specific for FKBPL is added to detect the captured FKBPL 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 450nm with the correction wavelength set at 630 nm.

background

FKBPL, also named as DIR1, NG7 and WISp39, has similarity to the immunophilin protein family, which plays a role in immunoregulation and basic cellular processes involving protein folding and trafficking. It is thought to have a potential role in the induced radioresistance, probably by increasing the rate of DNA repair in cells exposed to X rays. Breast cancer cells stably overexpressing FKBPL became dependent on estrogen for their growth and were dramatically more sensitive to the antiestrogens Tamoxifen and Fulvestrant, whereas FKBPL knockdown reverses this phenotype. FKBPL is an estrogen-inducible gene that acts as a cochaperone in ERα/Hsp90 molecular complexes; furthermore, FKBPL levels may be both a prognostic indicator and determinant of response to endocrine therapy. This kit is used to quantify FKBPL 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.

safety notes

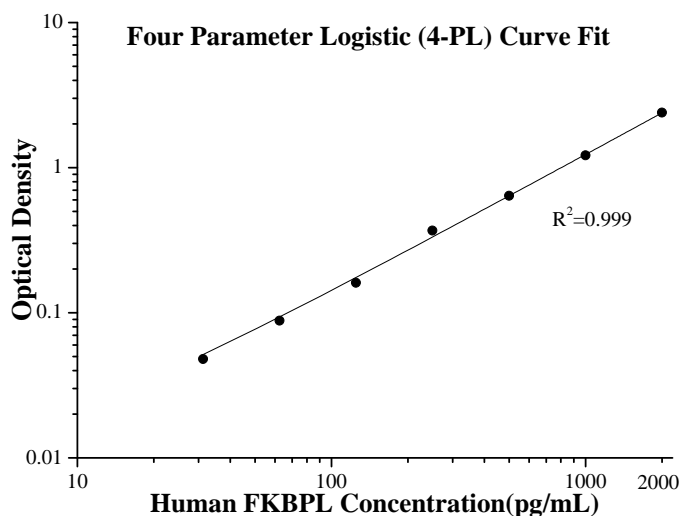
This product is sold for lab research and development use ONLY and not for use in humans or animals. Avoid any skin and eye contact with Stop Solution and TMB. In case of contact, wash thoroughly with water.

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.				

typical 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	0.046	0.0465	—
	0.047		
31.25	0.095	0.0945	0.048
	0.094		
62.5	0.127	0.1345	0.088
	0.142		
125	0.205	0.2075	0.161
	0.21		
250	0.412	0.415	0.3685
	0.418		
500	0.703	0.6855	0.639
	0.668		
1000	1.219	1.262	1.2155
	1.305		
2000	2.446	2.4375	2.391
	2.429		

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.

Sample	Intra-assay Precision			Inter-assay Precision		
	1	2	3	1	2	3
n	20	20	20	24	24	24
Mean (pg/mL)	1,084.9	266.4	80.8	1,112.7	275.0	85.8
SD	63.2	15.5	7.8	55.6	26.7	4.7
CV%	5.8	5.8	9.6	5.0	9.7	5.4

recovery

The recovery of FKBPL spiked to three different levels in four samples throughout the range of the assay in human plasma averaged 99%, ranging from 87%-115%.

sensitivity

The minimum detectable dose of human FKBPL is 7.0 pg/mL. This was determined by adding two standard deviations to the concentration corresponding to the mean O.D. of 20 zero standard replicates.

linearity

To assess the linearity of the assay, three samples were spiked with high concentrations of FKBPL in human plasma and diluted with the appropriate **Sample Diluent PT 1-a** 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	90
	Range (%)	83-102
1:4	Average% of Expected	95
	Range (%)	87-111
1:8	Average% of Expected	93
	Range (%)	91-95
1:16	Average% of Expected	101
	Range (%)	93-108

references

1. McKeen HD , et al. FKBPL regulates estrogen receptor signaling and determines response to endocrine therapy. Cancer Res. 70(3):1090-100(2010).
2. Jascur T, et al. Regulation of p21(WAF1/CIP1) stability by WISp39, a Hsp90 binding TPR protein. Mol Cell. 17(2):237-49,(2005)