

Mouse CXCL2 Sandwich ELISA Kit Datasheet

For the quantitative detection of mouse CXCL2 concentrations in serum, plasma and cell culture supernatants.

General Information

Catalogue Number	KE10022
Product Name	Mouse CXCL2 Sandwich ELISA Kit
Species cross-reactivity	Mouse
Range (calibration Range)	31.25-500 pg/mL
Tested applications	Quantification ELISA

Database Links

Entrez Gene	20310
SwissProt	P10889

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 - 500 pg/bottle; lyophilized*	2 bottles	
Detection antibody , biotinylated (100X) - 120 µL/vial	1 vial	
Streptavidin-horseradish peroxidase (HRP) (100X) - 120 µL/vial	1 vial	
Sample Diluent PT 1-tf - 30 mL/bottle. For serum and plasma	1 bottle	
Sample Diluent PT 1-eg - 30 mL/bottle. For cell culture supernatants.	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	

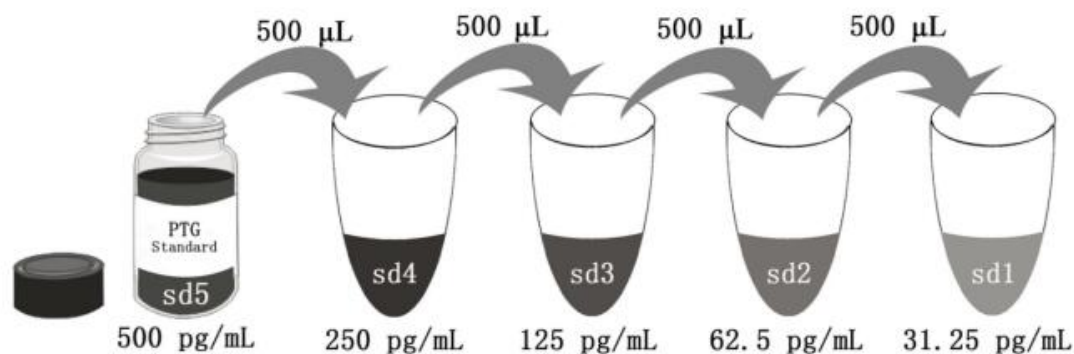
NB: Do not use the kit after the expiration date.

Sample Diluent PT 1-tf is for protein standard, serum and plasma samples.

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

Detection Diluent is for Detection antibody and Streptavidin-HRP.

*Add 1 mL Sample Diluent PT 1-tf or PT 1-eg in standard. This reconstitution gives a stock solution of 500 pg/mL.



Add # µL of Standard diluted in the previous step	—	500 µL	500 µL	500 µL	500 µL
# µL of Sample Diluent PT 1-tf or PT 1-eg	1000 µL	500 µL	500 µL	500 µL	500 µL
	"sd5"	"sd4"	"sd3"	"sd2"	"sd1"

Product Description

KE10022 is a solid phase sandwich Enzyme Linked-Immuno-Sorbent Assay (Sandwich ELISA). The CXCL2 ELISA kit is to be used to detect and quantify protein levels of endogenous CXCL2. The assay recognizes mouse CXCL2. An antibody specific for CXCL2 has been pre-coated onto the microwells. The CXCL2 protein in samples is captured by the coated antibody after incubation.

Following extensive washing, another antibody of biotinylated specific for CXCL2 is added to detect the captured CXCL2 protein. For signal development, Streptavidin-HRP 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

Cxcl2, also known as macrophage inflammatory protein 2-alpha (Mip2-alpha), is a member of CXC chemokine family that binds to CXC chemokine receptor 2 (Cxcr2). Cxcl2 is mainly produced by macrophages, endothelial cells, epithelial cells and tumor cells. Cxcl2 plays important roles in various biological progresses such as angiogenesis, inflammation, immune response and cancer biological behaviors. Under inflammatory conditions in CNS, microglia and astrocytes may secrete Cxcl2 as well as other chemokines, which induces chemotaxis and infiltration of circulatory neutrophils. Indeed, previous studies suggest that Cxcl2 is involved in Alzheimer disease, multiple sclerosis and brain ischemia.

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

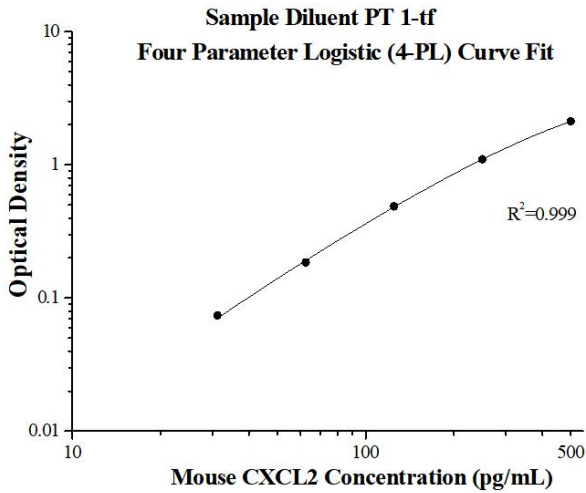
This product is sold for lab research and development use ONLY and not for use in humans or animals. Avoid any skin and eye contact with Stop Solution and TMB. In case of contact, wash thoroughly with water.

Assay Procedure Summary

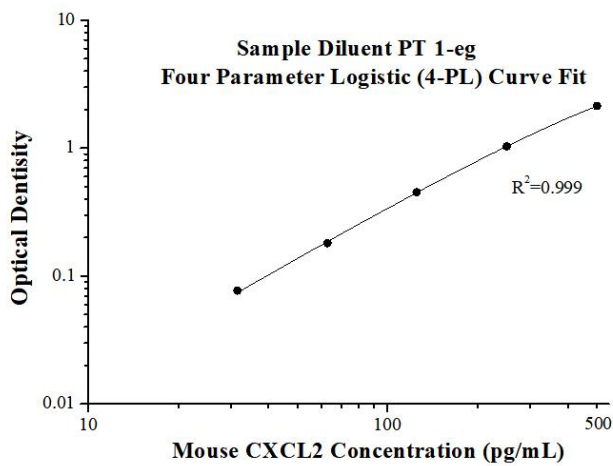
Step	Reagent	Volume	Incubation	Wash	Notes
1	Standard and Samples	100 µL	120 min	4 times	Cover Wells incubate at 4°C
2	Diluent Antibody Solution	100 µL	60 min	4 times	Cover Wells incubate at 37°C
3	Diluent HRP Solution	100 µL	40 min	4 times	Cover Wells incubate at 37°C
4	TMB Substrate	100 µL	15-20 min	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	0.084 0.082	0.083	-
31.25	0.159 0.155	0.157	0.074
62.5	0.265 0.271	0.268	0.185
125	0.547 0.595	0.571	0.488
250	1.133 1.236	1.184	1.102
500	2.171 2.249	2.21	2.127



(pg/mL)	O.D	Average	Corrected
0	0.05 0.045	0.048	-
31.25	0.13 0.119	0.125	0.077
62.5	0.227 0.23	0.229	0.181
125	0.503 0.498	0.501	0.453
250	1.092 1.07	1.081	1.034
500	2.155 2.212	2.184	2.136

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	438.8	13.6	3.1
2	20	137.6	4.4	3.2
3	20	30.7	1.6	5.1

Inter-assay Precision				
Sample	n	Mean (pg/mL)	SD	CV%
1	24	443.1	12.1	2.7
2	24	145.2	4.4	3.0
3	24	30.0	1.5	5.1

Recovery

The recovery of CXCL2 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 (%)
Mouse serum	1:8	86	72-133
Cell culture supernatants	1:2	104	85-121
	1:4	96	82-103

Sensitivity

The minimum detectable dose of mouse CXCL2 is 20.5 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 CXCL2 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 1:4)

		Mouse serum (Sample Diluent PT 1-tf)	Cell culture supernatants (Sample Diluent PT 1-eg)
1:2	Average% of Expected	76	116
	Range (%)	75-79	114-119
1:4	Average% of Expected	83	112
	Range (%)	80-90	110-114
1:8	Average% of Expected	89	104
	Range (%)	83-101	102-105
1:16	Average% of Expected	94	99
	Range (%)	86-106	96-102

References

1. Vansaun MN. et al. (2013). PLoS One. 8(9):e73054.
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4. Haraguchi K. et al. (2012). J. Neurosci.32:3931-3941.
5. L. Bozoyan. et al. (2015). J. Neuroinflammation. 12:173.
6. Hosking MP. et al. (2010). PLoS One. 5(6):e11340.