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Certification Of Analysis

Product Name: Mouse High mobility group protein B1 (HMGB1) ELISA Kit

Catalog Number: AAA13297

Species Reactivity: Mouse (Mus musculus)

Lot No: L11UHM29 **Exp:** May 28, 2022

Introduction

Item	Standard	Test Result		
Description	For the quantitative detection of Mouse High mobility	Conform		
	B1 (HMGB1) concentration in serum, plasma and oth			
	fluids.			
Identification	Sandwich	Positive		
Composition	Assay plate (96 Wells)	1		
	Standard (lyophilized)	2		
	Sample Diluent	$1 \times 20 \text{ mL}$		
	Biotin-Conjugate (concentrate 100 x)	$1 \times 120 \mu$ L		
	Biotin-Conjugate Diluent	$1 \times 20 \text{ mL}$		
	Streptavidin-HRP (concentrate 100 x)	$1 \times 120 \mu$ L	Conform	
	Streptavidin-HRP Diluent	$1 \times 20 \text{ mL}$		
	Substrate Solution	$1 \times 12 \text{ mL}$		
	Stop Solution	$1 \times 10 \text{ mL}$		
	Wash Buffer (concentrate 25 x)	$1 \times 20 \text{ mL}$		
	Adhesive Films	4		
	Instruction manual	1		
Assay Range	say Range 7.8-500 pg/mL			

Sensitivity

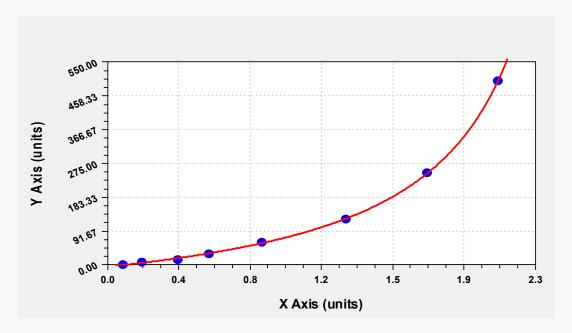
The limit of detection of Mouse HMGB1 defined as the analyte concentration resulting in an absorbance significantly higher than that of the dilution medium (mean plus 2 standard deviations) was determined to be 5.2 pg/mL (mean of 6 independent assays).

Typical data

For convenience in result calculation, absorbance as abscissa and standard concentrations can be used as ordinate. The standard curve provided in the manual is only for reference, experimenters should draw the standard curve based on data of themselves.

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pg/mL	Standard		Average	
500	2.113	2.071	2.092	
250	1.719	1.715	1.717	
125	1.276	1.290	1.283	
62.5	0.841	0.829	0.835	
31.25	0.535	0.567	0.551	
15.6	0.377	0.389	0.383	
7.8	0.205	0.185	0.195	
0	0.083	0.107	0.095	



Rational Function: $y=(a+bx)/(1+cx+dx^2)$

Coefficient Data:

a = -5.27306000843E+000

b = 5.54409383854E+001

c = -3.74892646533E-001

d = 1.29838850476E-003

Recovery

The recovery of Mouse HMGB1 spiked to levels throughout the range of the assay was evaluated.

Sample Type	Number	Recovery range (%)	Average(%)	
Mouse serum	10	90-95	93	
Mouse plasma	10	91-97	94	

Linearity

To assess the linearity of the assay, samples containing high concentrations of Mouse HMGB1 were serially diluted with Sample Diluent to produce samples with values within the dynamic range of the assay.

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Sample Type	1: 2	1: 4	1: 8	1: 16
Mouse serum	91-97%	90-95%	89-95%	93-99%
Mouse plasma	92-96%	94-100%	90-96%	91-98%

Precision

Intra-assay Precision (Precision within an assay)

Three samples of known concentration were tested twenty times on one plate to assess intra-assay precision. Inter-assay Precision (Precision between assays)

Three samples of known concentration were tested in forty separate assays to assess inter-assay precision. CV(%) = SD/meanX100

	Intra-assay Precision			Inter-assay Precision		
Sample	1	2	3	1	2	3
n	20	20	20	20	20	20
Mean (pg/mL)	0.375	0.657	0.927	0.409	0.734	1.114
SD	0.025	0.047	0.070	0.029	0.057	0.097
CV (%)	6.7	7.2	7.6	7.1	7.8	8.7

Date: 2021.11.29