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Certificate of Analysis

Product Name: ELISA Kit for Brain Natriuretic Peptide (BNP)

Cat.No.: AAA20421 Organism: Mus musculus (Mouse)

Lot.No. L231009272

Introduction

Item	Standard	Test Result	
Description	This immunoassay kit allows for the specific measurer		
	concentration in mouse serum, plasma, tissue homo	Conform	
	lysates, cell culture supernates and other biological fluids.		
Identification	Colorimetric	Positive	
Composition	Pre-coated, ready to use 96-well strip plate	1	
	Standard	2	
	Standard Diluent	1 × 20ml	
	Detection Reagent A (green)	$1 \times 120 \mu l$	
	Detection Reagent B (red)	$1 \times 120 \mu l$	
	Assay Diluent A	1 × 12ml	Conform
	Assay Diluent B	1 × 12ml	Comoni
	TMB Substrate	1 × 9ml	
	Stop Solution	1 × 6ml	
	Wash Buffer(30 x concentrate)	1 × 20ml	
	Plate sealer for 96 wells	4	
	Instruction manual	1	
Assay Range	12.35-1,000pg/mL		Conform

Sensitivity

The minimum detectable dose of mouse BNP is typically less than 4.81pg/mL.

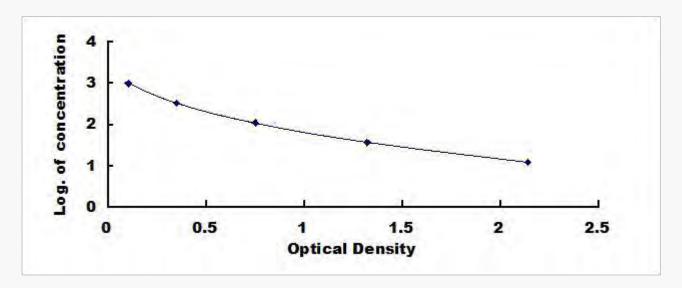
The sensitivity of this assay, or Lower Limit of Detection (LLD) was defined as the lowest protein concentration that could be differentiated from zero. It was determined the mean O.D. Value of 20 replicates of the zero standard added by their three standard deviations.

Standard curve

The standard curve is provided for demonstrated only. The client should perform the standard test in each independent experiment.

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pg/ml	Standard		Average	Log of concentration	
12.35	2.123	2.151	2.137	1.092	
37.04	1.318	1.325	1.322	1.569	
111.11	0.749	0.753	0.751	2.046	
333.33	0.335	0.368	0.352	2.523	
1000	0.107	0.105	0.106	3.000	



Recovery

Different matrices were spiked with certain level of mouse BNP and the recovery rates were evaluated.

Matrix	Recovery range (%)	Average(%)	
serum(n=5)	81-94	87	
EDTA plasma(n=5)	80-90	85	
heparin plasma(n=5)	84-96	91	

Linearity

The linearity of the kit was assayed by testing samples spiked with high concentration of mouse BNP and also several dilutions of these samples. The results were demonstrated by the percentage of calculated concentration to the expected.

Sample	1: 2	1: 4	1: 8	1: 16	
serum(n=5)	83-95%	87-102%	81-101%	96-106%	
EDTA plasma(n=5)	78-96%	90-99%	93-105%	80-91%	
heparin plasma(n=5)	87-103%	85-92%	79-89%	82-103%	

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Precision

Intra-assay Precision (Precision within an assay): 3 samples with low, middle and high level of mouse BNP were tested 20 times on one plate, respectively.

Inter-assay Precision (Precision between assays): 3 samples with low, middle and high level of mouse BNP were tested on 3 different plates, 8 replicates in each plate.

	Intra-assay Precision			Inter-assay Precision		
Sample	1	2	3	1	2	3
n	20	20	20	24	24	24
Mean (pg/ml)	36.14	112.87	333.58	38.36	115.47	335.32
SD	1.524	6.053	21.519	1.745	6.427	22.862
CV (%)	4.2	5.4	6.5	4.5	5.6	6.8

Date: <u>2023.10.16</u>