

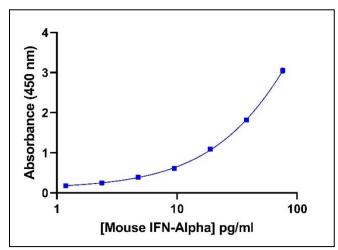
VeriKine-HS[™] Mouse Interferon Alpha All Subtype ELISA Kit Technical Data Sheet

Assay Range: 1.19 - 76 pg/ml, Sample Detection Range: 2.38 - 152 pg/ml Compatibility: Serum, Plasma, Tissue Culture Media Assay Length: 1 hr 54 min

INTRODUCTION

Interferons (IFNs) are a group of cytokines which exhibit pleitropic activities and play major roles in both innate and adaptive immunity. Type I IFNs consist of multiple IFN-Alpha genes and at least one IFN- β gene in most vertebrates, and a few other family members such as limitin in the mouse. The Verikine-HS Mouse IFN-Alpha All Subtype ELISA kit has been developed to quantitate levels of IFN-Alpha in serum, plasma, and tissue culture media

<u>Figure 1.</u> Representative Mouse IFN-Alpha Standard Curve in Sample Diluent



Specifications This kit quantitates mouse interferon alpha (IFN-Alpha) in serum, plasma, and tissue culture media (TCM) using a sandwich immunoassay. Interferon binds to plates coated with antibody and detection is accomplished using a biotinylated detection antibody followed by streptavidin conjugated to horseradish peroxidase (HRP). The substrate is tetramethyl-benzidine (TMB) and the standard provided is mammalian expressed mouse interferon alpha 4.

Specificity Recognizes all 14 mouse IFN-Alpha subtypes. Very weak cross-reactivity detected with human IFN-Alpha14. No cross-reactivity with mouse: IFN-Beta, IFN-Gamma; rat: IFN-Alpha1, IFN-Alpha14.

PRECISION

<u>Table 1.</u> Intra and Inter-Assay CVs To test precision within an assay (*intra*), and between assays (*inter*), independent assays testing endogenous IFN-Alpha concentrations in three lots of normal mouse serum were run by multiple operators.

Intra-Assay CV: 17 replicates of each concentration on a plate Inter-Assay CV: 6 independent assays run by same operator

Concentration (ng/ml)	Endogenous Level		
Concentration (pg/ml)	Low	Medium	High
Intra-Assay CV (%)	4.4	3.3	2.8
Inter-Assay CV (%)	7.8	5.8	8.7

SPIKE RECOVERY

<u>Table 2.</u> Average Linearity of Recovery of Serially Diluted Serum Containing Endogenous Mouse IFN-Alpha A single lot of normal mouse serum containing a high amount of endogenous mouse interferon alpha was serially diluted and analyzed across multiple runs. A total of 21 independent assays were run to obtain the data below.

.	Average Conce	% Linearity,	
Dilution	Not Adjusted for Dilution	Adjusted for Dilution	Adjusted for Dilution
2	39.5	79.1	
4	19.1	76.6	96.8
8	9.7	77.3	97.8
16	4.9	78.8	99.7
32	2.5	80.7	102.0
64	1.2	77.7	98.2
Avg		78.4	98.9

PERFORMANCE CHARACTERIZATION

<u>Figure 2.</u> Endogenous Mouse IFN-Alpha Quantified in Plasma or Serum From Three Mouse Strains Using PBL's High Sensitivity Mouse IFN-Alpha All Subtype ELISA Total endogenous levels of mouse IFN-Alpha were quantified in pooled plasma (blue, left) and individual sera (red and green, right) for three mouse strains: CD-1, BALB/c, and C57BL/6. Results from this experiment led to the recommendation to pre-screen serum, as a significant portion of samples contain quantifiable levels of endogenous interferon alpha.

