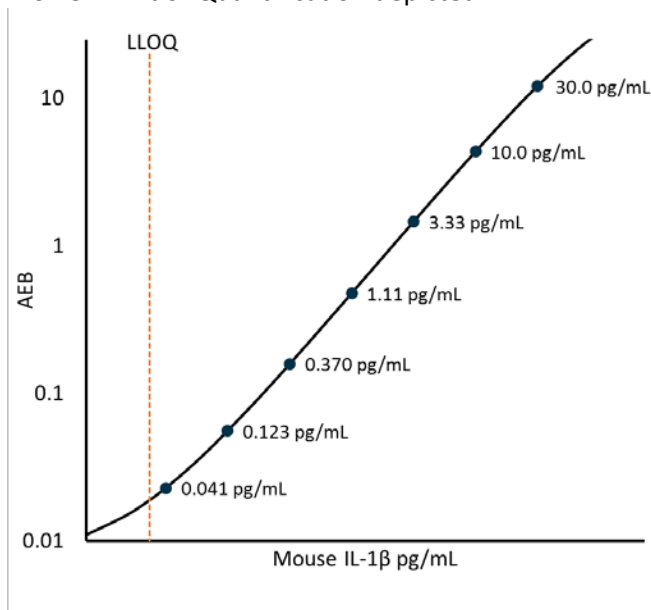


Description

The IL-1 family consists primarily of three proteins: IL-1α, IL-1β (agonists) and IL-1ra (antagonist) which interact with the IL-1 receptor. IL-1β shares 33% homology with IL-1α. IL-1β exists as a 33 kDa precursor which is cleaved by caspase-1 into its 17 kDa active form. It is unknown how IL-1β is actively secreted but it is suggested exocytosis, transport by multi-drug resistance transporters, and cell death may all play a role. Knockout models of IL-1β show no gross physiological detriment, though its role is suspected to function in disease states rather than healthy tissue. Evidence shows potential involvement in Long Term Potentiation demonstrating increases following induction, and the prevention of induction with a competitive antagonist. IL-1β is believed to be part of an inflammatory response thought to be protective but often goes awry. There is a distinguishable link between oxidative stress, glutamate excitotoxicity and IL-1β.

Calibration Curve: Calibrator concentrations and Lower Limit of Quantification depicted.



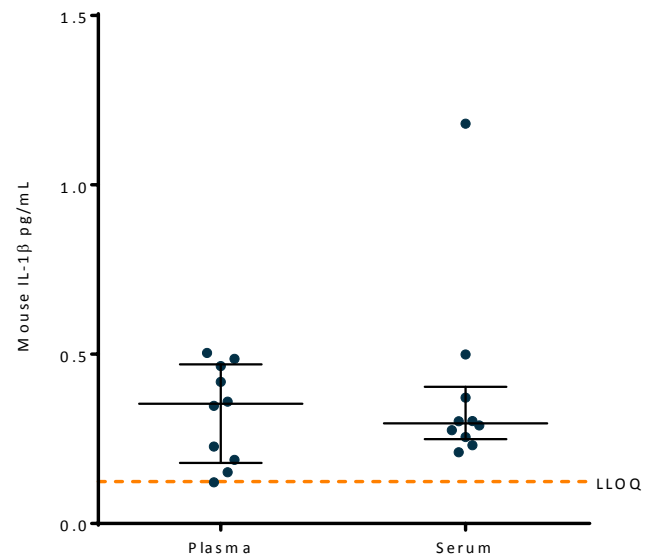
Lower Limit of Quantification (LLOQ): Triplicate measurements of serially diluted calibrator were read back on the calibration curve over 6 runs each for 1 reagent lot across 3 instruments (6 runs total).

Limit of Detection (LOD): Calculated as 2.5 standard deviations from the mean of background signal read back on each calibration curve over 6 runs each for 1 reagent lot across 2 instruments (6 runs total).

LLOQ	0.0309 pg/mL pooled CV 16% mean recovery 94%
LOD	0.0081 pg/mL range 0.0030-0.0132 pg/mL
Dynamic range	Serum and plasma: 0 – 120 pg/mL
Diluted Sample volume*	100 μL per measurement
Tests per kit	192

*See Kit Instruction for details

Endogenous Sample Reading: Healthy donor plasma (n=10) and serum (n=10) were measured. Bars depict median with interquartile range. Orange line represents functional LLOQ.



Sample Type	Mean mouse IL-1β pg/mL	Median mouse IL-1β pg/mL	% Above LLOQ	% Above LOD
Serum	0.392	0.296	100%	100%
Plasma	0.327*	0.354	90%	100%

*Values below LLOQ are not included in the mean

Precision: Measurements of 3 serum- and plasma-based panels and 2 calibrator-based controls. Triplicate measurements were made for 6 runs each for 1 reagent lot across 2 instruments (6 runs total, 18 measurements).

Sample	Mean (pg/mL)	Within run CV	Between run CV	Between inst CV
Control 1	0.916	5.6%	5.5%	3.4%
Control 2	23.4	7.5%	5.7%	5.0%
Panel 1	0.309	8.3%	8.7%	8.4%
Panel 2	0.162	15.4%	8.4%	6.4%
Panel 3	30.7	4.7%	2.1%	1.0%

Spike and Recovery: 2 serum and 2 plasma samples were spiked at high and low concentrations within the range of the assay and analyzed on SR-X.

Dilution Linearity: 1 spiked plasma and 1 spiked serum sample were diluted 2X serially from MRD (4x) to 512x with Sample Diluent.

Spike and Recovery (Serum/Plasma)	Mean = 91% Range: 65–124%
Dilution Linearity (512x)	Mean = 98% Range: 80–121%

The Simoa mouse IL-1β Discovery assay kit is formulated for use on either the SR-X or HD-1 platform. Data in this document was obtained from runs on the SR-X platform unless otherwise noted. Some differences in performance claims between the HD-1 and SR-X may be observed when comparing datasheets for the two platforms. This may be due to experiments run at different time-points with different reagent lots and different samples, or may be due to minor differences in antibody and analyte behavior in the different assay formats.