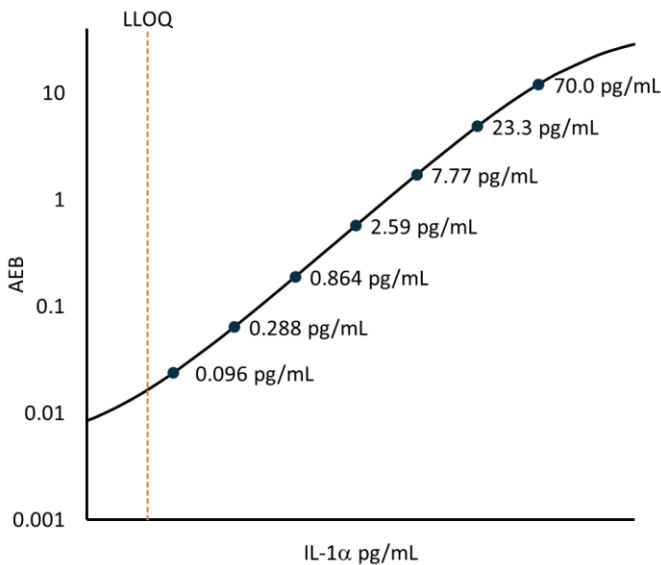


Description

The IL-1 family is integral to innate inflammation and reducing inflammation, helping facilitate specific immunological responses. The IL-1 family is initially translated as precursors, lacking signal peptides for secretion. These precursors exist in the cytosol and following cell death by necrosis, are released and activated by extracellular processing (giving rise to their being termed alarmins.) IL-1 receptor antagonist (IL-1Ra) is produced to dampen IL-1 responses, outcompeting the IL-1 family at its receptors. The IL-1 family contains a common Toll-IL-1-receptor (T1R) domain, being the key functional domain. Produced by activated macrophages, IL-1α stimulates thymocyte proliferation by inducing IL-2 release, B-cell maturation and proliferation, and fibroblast growth factor activity. IL-1 proteins are involved in the inflammatory response, being identified as endogenous pyrogens and are reported to stimulate the release of prostaglandin and collagenase from synovial cells.

Calibration Curve: Four parameters from the curve fit are depicted.



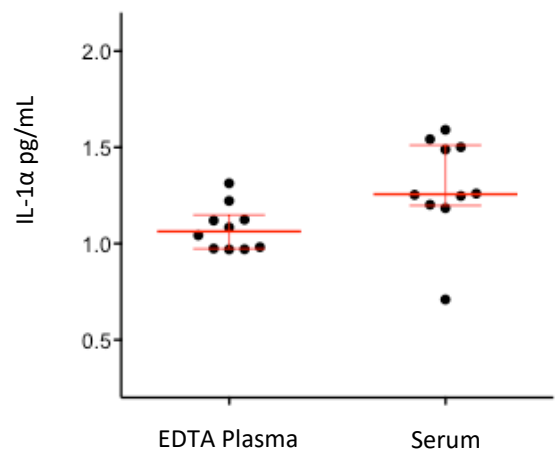
Lower Limit of Quantification (LLOQ): Triplicate measurements of serially diluted calibrator were read back on the calibration curve over 1 reagent lot on 1 instrument (5 runs total).

Limit of Detection (LOD): Calculated as 2.5 standard deviations from the mean of background signal read back on each calibration curve over 1 reagent lot across 4 instruments (6 runs total).

LLOQ	0.060 pg/mL pooled CV 9.6% mean recovery 112%
LOD	0.0085 pg/mL range 0.0044–0.0126 pg/mL
Dynamic range (serum and plasma)	0–700 pg/mL
Diluted Sample volume*	100 µL per measurement
Tests per kit	192

*See Kit Instruction for details

Endogenous Sample Reading: IL-1α in EDTA plasma (n=10) and serum (n=10) from non-medicated, non-immunized mice. Error bars depict median and interquartile ranges.



Sample Type	Median IL-1α pg/mL	% Above LOD
EDTA Plasma	1.063	100%
Serum	1.256	100%

Precision: Representative precision was estimated with repeated assay of serum and plasma samples using one instrument and one reagent lot. Within-run and between-run CVs are depicted in the following table. Within-run CVs reflect average CVs across 5 experiments of 3 replicates each.

Sample	Mean (pg/mL)	Within run CV	Between run CV
Serum Sample 1	71.6	4.31%	5.62%
Plasma Sample 2	6.15	4.36%	4.38%
Plasma Sample 3	0.942	7.80%	6.90%

Spike and Recovery: IL-1α spiked into 2 serum and 2 plasma samples at 2 levels.

Dilution Linearity (Plasma): Spiked plasma pools diluted serially from MRD (10x) to 320x with Sample Diluent.

Dilution Linearity (Serum): Spiked serum pools diluted serially from MRD (10x) to 320x with Sample Diluent.

Spike and Recovery (Serum/Plasma)	Mean = 92.3% Range: 73–115%
Dilution Linearity (Plasma, 320x)	Mean = 109% Range: 96.8–122%
Dilution Linearity (Serum, 320x)	Mean = 138% Range: 118–155%

Specificity (Competition): 2 serum spiked serum samples competitively inhibited with unlabeled IL-1a antibody.

Specificity (Cross Reactivity): mouse IL-1b spiked into buffer at 70 pg/mL.

Specificity (Competition)	100%
Specificity (Cross Reactivity)	0.061%