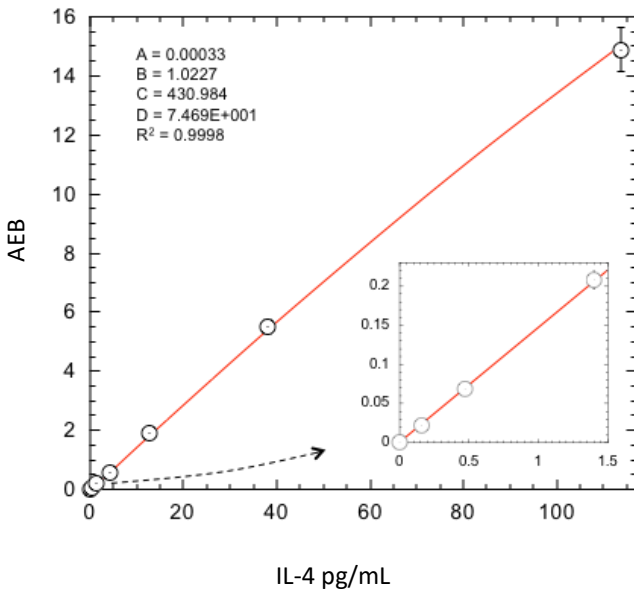


**Description**

Human Interleukin-4 (IL-4) is a monomeric cytokine, approximately 13-18 kDa, expressed by Th2-biased CD4+ T cells, mast cells, basophils, and eosinophils. IL-4 has a compact, globular fold (similar to other cytokines) stabilized by 3 disulphide bonds. Mature human IL-4 shares 55%, 39%, and 43% amino acid sequence identity with bovine, mouse, and rat IL-4, respectively. By binding to IL-4 receptor or receptor complex, IL-4 has many biological functions. It promotes cell proliferation, survival, and immunoglobulin class switch to IgG4 and IgE in human B cells, acquisition of the Th2 phenotype by naïve CD4+ T cells, priming and chemotaxis of mast cells, eosinophils, and basophils, and the proliferation and activation of epithelial cells. IL-4 plays an important role in the development of allergic inflammation and asthma.

**Calibration Curve:** Four-parameter curve fit parameters are depicted.



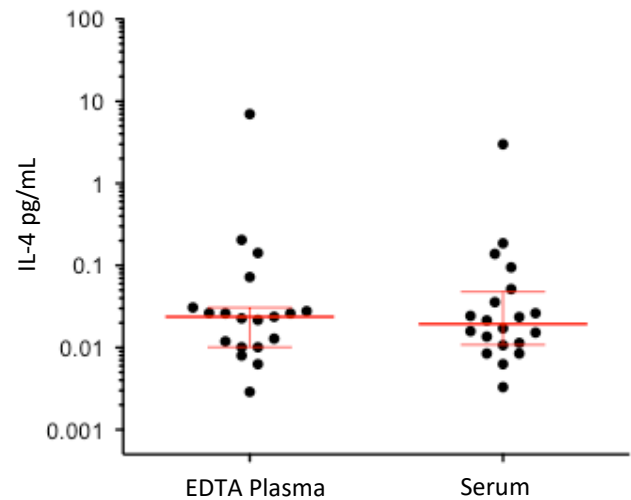
**Lower Limit of Quantification (LLOQ):** Triplicate measurements of serially diluted calibrator were read back on the calibration curve over 2 reagent lots across 3 instruments (12 runs total).

**Limit of Detection (LOD):** Calculated as 2.5 standard deviations from the mean of background signal read back on each calibration curve over 2 reagent lots across 3 instruments (12 runs total).

<b>LLOQ</b>	<b>0.039 pg/mL</b> pooled CV 18.1% mean recovery 115.2%
<b>LOD</b>	<b>0.0046 pg/mL</b> range 0.0008–0.0084 pg/mL
<b>Dynamic range (serum and plasma)</b>	0–200 pg/mL
<b>Diluted Sample volume*</b>	100 µL per measurement
<b>Tests per kit</b>	96

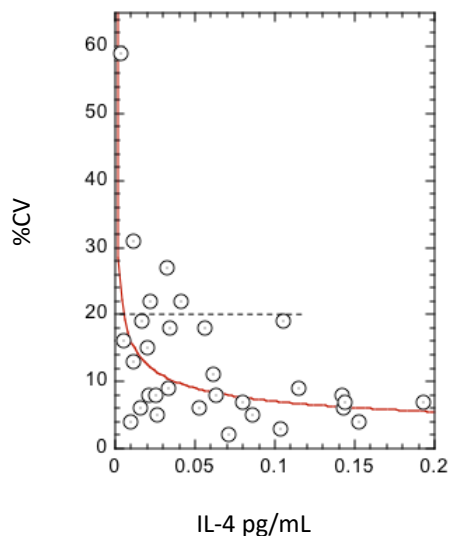
\*See Kit Instruction for details

**Endogenous Sample Reading:** Healthy donor matched EDTA plasma (n=19) and serum (n=20) were measured. Error bars depict median and interquartile ranges.



Sample Type	Median IL-4 pg/mL	% Above LOD
EDTA Plasma	0.024	80%
Serum	0.019	90%

**Sample Dose CV Profile:** Triplicate measurements of diluted serum samples assayed over multiple runs (30 measurements).



<b>Spike and Recovery (Serum)</b>	<b>Mean = 83.1%</b> Range: 72.7–93.4%
<b>Admixture Linearity</b>	<b>Mean = 93.7%</b>
<b>Dilution Linearity (512x)</b>	<b>Mean = 110%</b>

**Precision:** Four samples consisting of two serum-based panels and two IL-4 controls were assayed in replicates of three at two separate times per day for five days using a single lot of reagents and calibrators. Analysis of variance (fully nested ANOVA) results are summarized in the following table.

Sample	Mean (pg/mL)	Within run CV	Between run CV	Between day CV
Control 1	8.98	3.5%	3.5%	0.0%
Control 2	92.7	5.2%	4.5%	4.1%
Panel 1	6.26	3.9%	4.8%	0.0%
Panel 2	67.7	5.5%	10.9%	0.0%

**Spike and Recovery:** IL-4 spiked into 4 serum samples at 2 levels.

**Admixture Linearity:** High IL-4 serum sample admixed with low IL-4 sample, mean of 10 levels.

**Dilution Linearity:** Spiked plasma sample diluted 2x serially from MRD (2x) to 512x with Sample Diluent.