Quanterix™
The Science of Precision Health

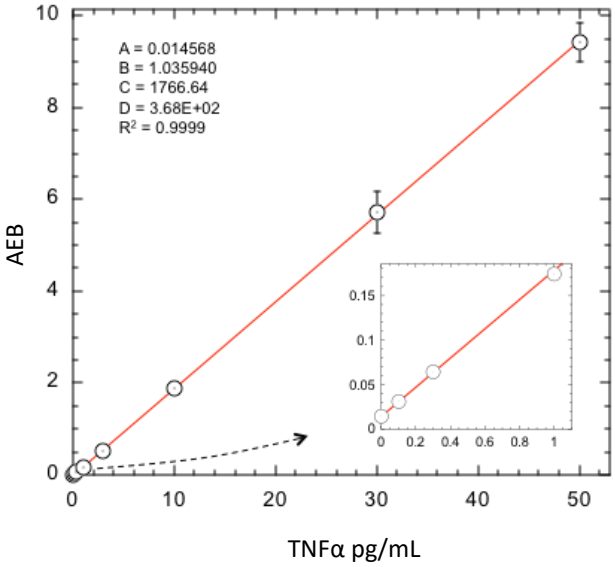
Simoa® TNFα Advantage Kit
HD-1/HD-X Data Sheet

Item 101580

Description

Human tumor necrosis factor alpha (TNFα) is a homotrimeric transmembrane protein that functions as a proinflammatory cytokine. It is produced mainly by macrophages but also by a variety of other cell types, including monocytes, neutrophils, and T-cells. The involvement of TNFα in several signal transduction pathways links the protein to such diverse functions as acute inflammation, apoptosis, septic shock, cellular proliferation, and differentiation. Human TNFα is a non-glycosylated protein of 157 amino acids, with a molecular weight of approximately 17,000 daltons. The clinical relevance of TNFα stems from its association with numerous disease states including rheumatoid arthritis, cancer, cachexia, and Crohn's disease.

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



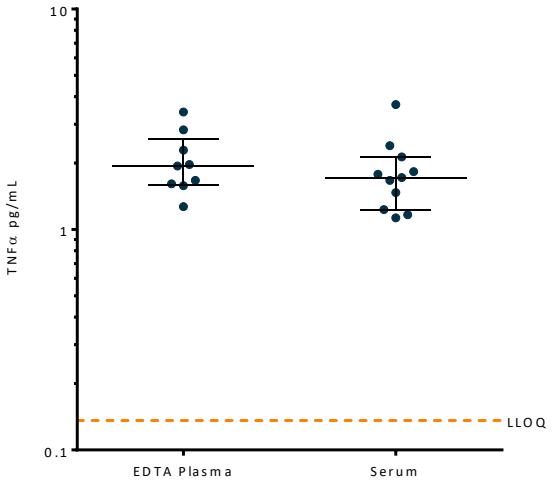
Lower Limit of Quantification (LLOQ): Triplicate measurements of serially diluted calibrator were read back on the calibration curve over 2 reagent lots across 3 instruments (12 runs total).

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).

LLOQ	**0.034 pg/mL**
LOD	**0.016 pg/mL** SD 0.0186 pg/mL
Dynamic range (serum and plasma)	0–200 pg/mL
Diluted Sample volume*	100 µL per measurement
Tests per kit	192

***See Kit Instruction for details**

Endogenous Sample Reading: Healthy donor matched EDTA plasma (n=9) and serum (n=11) were measured. Error bars depict mean and SEM.



Sample Type	Median TNFα pg/mL	% Above LOD
Serum	1.78	100%
EDTA Plasma	1.94	100%

Quanterix Corporation
900 Middlesex Turnpike, Billerica, MA 01821
techsupport@quanterix.com

Precision: Five samples consisting of three serum-based panels and two TNFα 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
Control 1	2.77	3.4%	4.1%	0.0%
Control 2	29.9	2.8%	4.8%	0.0%
Panel 1	1.71	4.9%	9.1%	0.0%
Panel 2	2.15	5.5%	9.2%	0.0%
Panel 3	43.4	3.5%	0.4%	1.0%

Spike and Recovery: TNFα spiked into 4 serum samples at 2 levels.

Admixture Linearity: High TNFα serum sample admixed with low TNFα sample, mean of 10 levels.

Dilution Linearity: 1 spiked serum sample was diluted 2x serially from MRD (4x) to 64x with Sample Diluent.

Lot Consistency: 5 samples tested with 2 reagent lots across 2 runs x 3 instruments.

Spike and Recovery (Serum)	**Mean = 77.9%** Range: 68.2–87.1%
Admixture Linearity	**Mean = 108%**
Dilution Linearity (64x)	**Mean = 121%** Range: 101–139%
Lot Consistency	**Mean Difference = -2.7%** Range: 1.34–41.1 pg/mL

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

Quanterix Corporation
900 Middlesex Turnpike, Billerica, MA 01821
techsupport@quanterix.com