Quanterix
Discovery Fueled by Ultra-Sensitivity

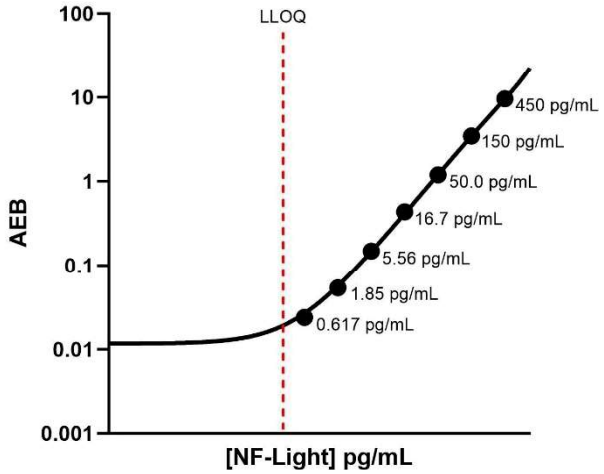
Simoa® NF-Light Advantage PLUS Kit
HD-X Data Sheet

Item 104364

Description

Neurofilament light (NF-L) is a 68 kDa cytoskeletal intermediate filament protein that is expressed in neurons. It associates with the 125 kDa Neurofilament medium (NF-M) and the 200 kDa Neurofilament heavy (NF-H) to form neurofilaments. They are major components of the neuronal cytoskeleton and are believed to function primarily to provide structural support for the axon and to regulate axon diameter. Neurofilaments can be released in significant quantity following axonal damage or neuronal degeneration. NF-L has been shown to associate with traumatic brain injury, multiple sclerosis, frontotemporal dementia, and other neurodegenerative diseases. The Simoa NF-Light Advantage PLUS assay is a digital immunoassay for the quantitative determination of NF-L in serum, plasma, and CSF.

Calibration Curve: Representative calibrator concentrations and Lower Limit of Quantification (LLOQ) depicted. For NF-Light Advantage PLUS, the reconstitution volume for calibrator concentrates may vary between kit lots, while keeping the target calibrator concentrations each level as consistent as possible: The minimum allowable concentration for Cal H is 450 pg/mL and the maximum allowable concentration for Cal B is 0.617 pg/mL.



MRD (Minimum Required Dilution)	
Diluted Sample Volume	100 µL per measurement
Human Serum and EDTA Plasma Dilution	1:4
Human CSF Dilution	1:100
Tests per kit	96

See Kit Instruction for details.

Lower Limit of Quantification (LLOQ): Triplicate measurements of serially diluted calibrator were read back on the calibration curve over 12 runs each for 2 reagent lots across 2 instruments (3 runs per lot, per instrument). The analytical LLOQ was set at the lowest concentration that read back within 80 – 120% of the expected value with a CV < 20%. The functional LLOQ (fLLOQ) values for each matrix represent the analytical LLOQ multiplied by the dilution factor used for the respective matrix (Table).

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

Assay Range: The upper end of the dynamic range, or functional Upper Limit of Quantification (ULOQ) is equal to the minimum top calibrator concentration multiplied by MRD.

Analytical LLOQ	**0.309 pg/mL** pooled CV 13.2% mean recovery 104%
Functional LLOQ (serum and plasma)	**1.24 pg/mL**
Functional ULOQ (serum and plasma)	**1800 pg/mL**
Functional LLOQ (CSF)	**30.9 pg/mL**
Functional ULOQ (CSF)	**45000 pg/mL**
LOD	**0.062 pg/mL** range 0.009–0.145pg/mL

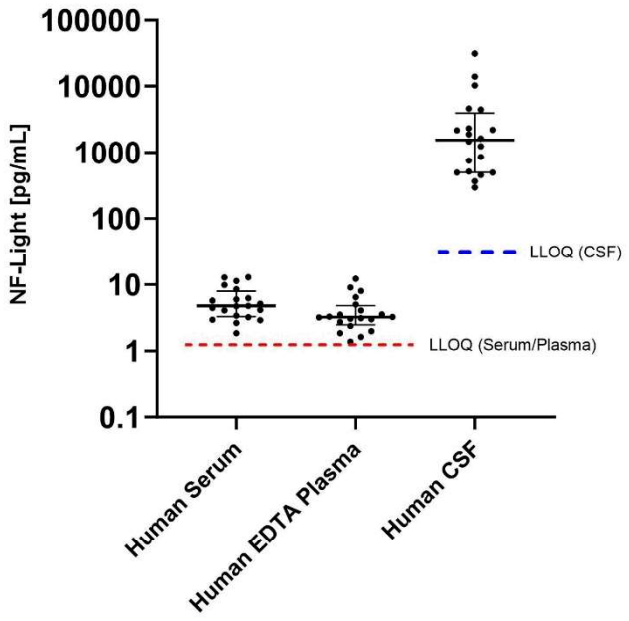
Quanterix Corporation
900 Middlesex Turnpike, Billerica, MA 01821
techsupport@quanterix.com

DOC TEMPLATE-0061 05

Simoa® NF-Light Advantage PLUS Kit
HD-X Data Sheet

Item 104364

Endogenous Sample Reading: Concentrations (pg/mL) were determined for matched serum (n=20) and EDTA plasma (n=20) from normal human donors and unmatched normal human CSF (n=20) using the NF-Light Advantage PLUS kit on HD-X. Bars depict median with interquartile range. The red and blue lines represent functional LLOQ.



Sample Type	Mean NF-Light pg/mL	Median NF-Light pg/mL	% Above LOD	% Above LLOQ
Human Serum	5.89	4.75	100%	100%
Human EDTA Plasma	4.15	3.23	100%	100%
Human CSF	4134	1552	100%	100%

Precision: Measurements of 2 human serum-based panels, 2 human EDTA plasma-based panels, and 2 calibrator-based controls were measured for precision. Triplicate measurements were made for 6 runs each for 2 reagent lot across 2 instruments (12 runs total, 36 measurements). All samples were diluted at the appropriate MRD for the sample matrix.

Sample	Mean (pg/mL)	Within run CV	Between run CV	Between inst CV	Between Lot CV
Control 1*	4.72	5.8%	14.6%	19.3%	6.1%
Control 2	380	3.5%	9.4%	1.0%	4.3%
Panel 1	10.1	3.9%	11.0%	3.7%	5.7%
Panel 2	7.36	7.0%	14.7%	6.9%	6.8%
Panel 3	73.1	2.6%	12.9%	0.1%	6.1%
Panel 4	594	2.6%	10.9%	5.1%	4.2%

*One replicate removed from CV calculations, determined to be an outlier.

Spike and Recovery: Two normal human samples for each matrix (EDTA plasma and CSF) were spiked at high (66 pg/mL) and low (13 pg/mL) concentrations within the range of the assay and analyzed on HD-X. Percent recovery is defined as the difference between the measured concentration of NF-Light in the spiked sample and the measured concentration in unspiked sample relative to the concentration of NF-Light in spiked calibrator diluent.

Dilution Linearity: Two human samples for each matrix (serum, plasma and CSF) were serially diluted with sample diluent through 7 levels of 2X dilutions. Each dilution series was run on the HD-X with two different lots of NF-Light Advantage PLUS assay kits with the MRD (4X) dilution applied. The total dilution of each human serum or human EDTA plasma sample ranged from 4X to 512X. Each human CSF dilution series was run on the HD-X with the MRD (100X) dilution applied. Total dilution of each CSF sample ranged from 100X to 12800X.

Spike and Recovery (Human Serum)	**Mean 93%** Range 87–100%
Spike and Recovery (Human EDTA Plasma)	**Mean 103%** Range 97–114%
Spike and Recovery (Human CSF)	**Mean 107%** Range 98–113%
Dilution Linearity (Human Serum, 4X – 512X)	**Mean 110%** Range 86–150%
Dilution Linearity (Human EDTA Plasma, 4X – 512X)	**Mean 108%** Range 98–121%
Dilution Linearity (Human CSF, 100X – 12800X)	**Mean 89%** range 71–105%

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