Simoa® p-Tau 205 Advantage PLUS Kit
HD-X Data Sheet Item 105634

Description: This data sheet summarizes data from the analytical validation performed at Quanterix to characterize performance of the p-Tau 205 Advantage PLUS kit on the Simoa HD-X Analyzer®.

Matrix Types: Assay performance and a minimum required dilution (MRD) were evaluated in multiple matrices (Table 1). See the Kit Instructions for sample and dead volume requirements.

Table 1. Minimum required dilutions

Matrix Types	Serum, EDTA Plasma, CSF
Diluted Sample Volume	100 uL per measurement
Serum MRD	2x
EDTA Plasma MRD	2x
CSF MRD	8x

Calibration Curve: The reconstitution volume, assigned concentrations, Limit of Detection (LOD), analytical Upper Limit of Quantification (ULOQ), analytical Lower Limit of Quantification (LLOQ) described here are representative and may vary from kit lot to kit lot (Figure 1). Refer to the Certificate of Analysis (CoA) for lot-specific calibrator concentrations and reconstitution volumes.

Limit of Quantification (LOQ): The analytical LLOQ was determined as the lowest concentration of the analyte in Sample Diluent with a recovery between 80 – 120% and a CV < ±20%. The analytical ULOQ (ULOQ) is the concentration of the highest calibrator. The analytical LLOQ and the analytical ULOQ multiplied by the MRD yields the functional LLOQ (fLLOQ) and the functional ULOQ (fLLOQ). The LLOQ was experimentally verified for each kit lot. Minor variations in ULOQ between kit lots may be observed where lot matching was performed (Table 2).

Limit of Detection (LOD): The LOD was calculated as 2.5 standard deviations above the mean of the background (Cal A) (Table 2).

Table 2. LLOQ and LOD

	Analytical	Functional
LLOQ	0.2250 pg/mL	Serum (2x): 0.4500 pg/mL EDTA Plasma (2x): 0.4500 pg/mL CSF (8x): 1.8000 pg/mL
ULOQ	164.0000 pg/mL	Serum (2x): 328.0000 pg/mL EDTA Plasma (2x): 328.0000 pg/mL CSF (8x): 1312.0000 pg/mL
LOD	0.0470 pg/mL	N/A

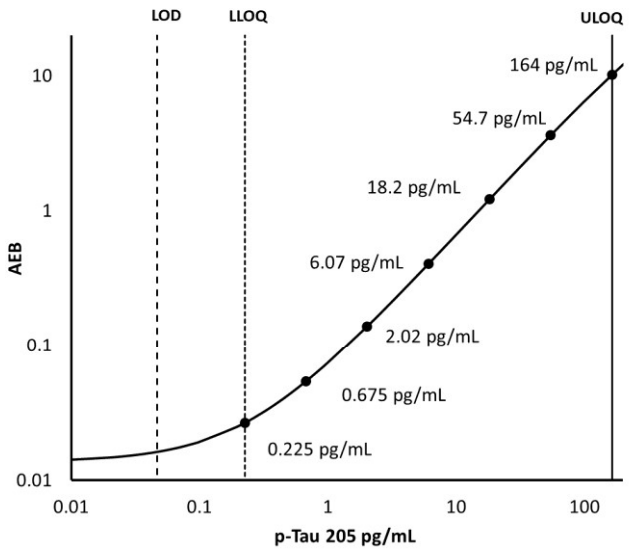

Figure 1. Example calibrator curve

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

Reference Ranges: Range was established using normal matched human serum and EDTA plasma (n=20) and normal human CSF (n=20). The mean and median analyte concentrations and the percent of samples above fLLOQ and LOD are reported below (Figure 2 and Table 3).

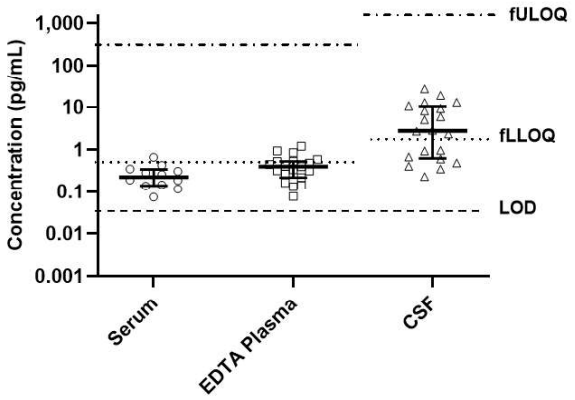

Figure 2. Normal sample readings. Bars depict the median with the interquartile range.

Table 3. Normal sample readings

Sample Type	Mean pg/mL	Median pg/mL	% Above LOD	% Above fLLoQ
Serum *	0.6691 †	N/A †	55.0%	5.0%
EDTA Plasma *	0.7320	0.5900	90.0%	35.0%
CSF *	9.9682	8.8411	90.0%	60.0%

* Samples below fLLoQ are not included in the median and mean
† Only one sample is above fLLOQ

Precision: Precision was calculated using serum, EDTA plasma, CSF, and assay kit controls diluted to the appropriate MRD (Table 4). Triplicate measurements were made across 12 runs. 2 reagent lots were each tested 3 times on 2 instruments, providing a mean of 36 individual measurements. Four distinct precision values were calculated from the 36 measurements:

1. **Within Run %CV** describes the variability of the %CV within a run, inclusive of 2 reagent lots and 2 instruments.
2. **Run to Run Conc %CV** describes the variability of the concentration value across all 12 runs, inclusive of 2 reagent lots and 2 instruments.
3. **Instrument %CV** describes instrument-to-instrument variability.
4. **Lot %CV** describes lot-to-lot variability.

Table 4. Precision

Sample	Mean (pg/mL)	Within Run %CV	Run to Run Conc %CV	Instrument %CV	Lot %CV
Control 1	6.1288	8.1%	11.2%	1.5%	2.5%
Control 2	92.0900	6.5%	10.6%	0.5%	1.2%
EDTA Plasma 1	1.6995	11.3%	15.9%	5.5%	2.6%
EDTA Plasma 2	14.4509	8.7%	17.0%	0.6%	0.4%
Serum 1	1.9288	7.2%	17.0%	3.2%	1.2%
Serum 2	15.7415	4.5%	15.7%	4.0%	11.7%
CSF 1	16.4800	3.3%	15.5%	8.7%	5.2%
CSF 2	43.4283	2.6%	12.0%	4.2%	2.0%

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

Spike Recovery: Percent recovery was calculated as the difference between the spiked (with antigen) sample and the un-spiked sample, relative to the spiked (with antigen) Sample Diluent (Table 5).

Linearity: 12 normal human samples (4 serum, 4 EDTA plasma, 4 CSF) spiked with calibrator were diluted 2x serially with sample diluent from 2x to 128x for serum and EDTA plasma, and 8x to 512x for CSF. Linearity refers to the assay's ability to produce proportional and accurate results across a defined dilution range. Linearity was assessed by performing serial dilutions of samples diluted with Sample Diluent (Table 5).

Table 5. Spike Recovery and Linearity

Spike Recovery Serum	**Mean 89.4%** range 68.0 – 109.6%
Spike Recovery EDTA Plasma	**Mean 76.3%** range 55.1 – 93.5%
Spike Recovery CSF	**Mean 107.3%** range 98.0 – 121.1%
Linearity **(Serum, 2x – 128x)**	**Mean 110.0%** range 91.0 – 133.1%
Linearity **(EDTA Plasma, 2x – 128x)**	**Mean 130.0%** range 102.3– 173.6%
Linearity **(CSF, 8x – 512x)**	**Mean 100.0%** range 68.5 – 108.9%

Other Performance Characteristics: Cross-reactivity with p-Tau 217, p-Tau 231, p-Tau 181 and p-Tau 212 was interrogated. No significant cross-reactivity was observed with any of those tested antigens. Cross-reactivity with the p-Tau 202 site was not examined, thereby cross-reactivity with the p-Tau 202 site cannot be ruled out.

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