MSD® Human Total Tau Kit

For quantitative determination in human cerebrospinal fluid
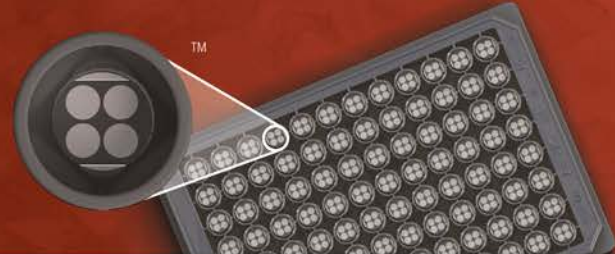
Catalog Numbers
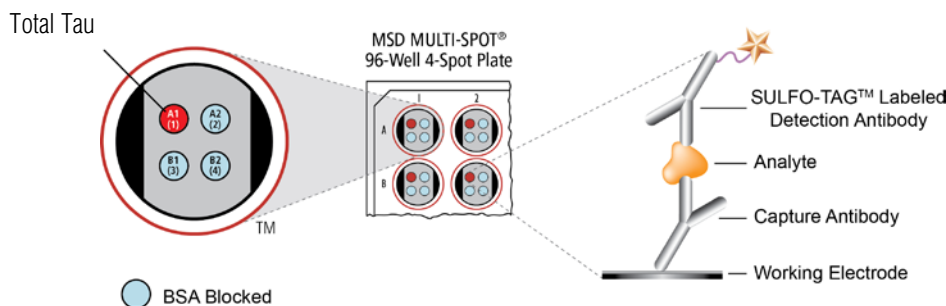
Human Total Tau Kit	
Kit size	
1 plate	K151LAE-1
5 plates	K151LAE-2
25 plates	K151LAE-4

Ordering Information

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Total Tau

The human tau protein is expressed primarily in neuronal cells where it localizes to the axon and impacts both cytoskeletal structure and cell signaling. Tau has emerged as a putative therapeutic target for many neurodegenerative disorders, including Alzheimer's disease, frontotemporal dementia, Pick's disease, progressive supranuclear palsy, and corticobasal degeneration.[1] A key characteristic of these tauopathies is the presence of intracellular neurofibrillary tangles made up predominantly of hyperphosphorylated forms of the protein. Tau and Aβ42 have been identified as core biomarkers of Alzheimer's disease. Their levels in cerebrospinal fluid (CSF) reproducibly distinguished normal and Alzheimer's patients, and the combination may be useful in identifying patients with mild cognitive impairment (MCI).[2-4] Studies aimed at evaluating the association between Alzheimer-type pathologic changes in the brain and antemortem CSF levels of Aβ42 and tau protein indicated that levels of both proteins correlated with the presence of neurofibrillary tangles and Aβ in the brain.[5] CSF total tau and Aβ42 levels are effective markers for discriminating incipient Alzheimer's from age-related memory impairment, depression, and some secondary dementias.[6]

The MSD Human Total Tau Assay has been validated for the detection of total tau protein in CSF. The performance of this kit is consistent with the principles outlined in "Fit-for-Purpose Method Development and Validation for Successful Biomarker Measurement" by J.W. Lee, et al.[7] Representative data from the assay validation are presented below. Lot-specific standard curve can be found in the certificate of analysis (C of A) supplied with the kit. A copy of the lot-specific C of A can be found at www.mesoscale.com by entering the kit model number into the search box.

This datasheet outlines the performance of the assay. The assay is available on 96-well 4-spot plates.

Assay Sensitivity

	Tau (pg/mL)
LLOD Range	1.07–23.7
LLOQ	30.0
ULOQ	8000

Testing of the kit involved a minimum of 12 runs conducted by three analysts across at least 3 days (N=54 runs across three kit lots). The lower limit of detection (LLOD) is a calculated concentration based on a signal of 2.5 standard deviations above the background. The lower limit of quantification (LLOQ) is the lowest concentration where the %CV of the calculated concentration is less than 20% and the percent recovery of the standard is between 80% and 120%. The upper limit of quantification (ULOQ) is the highest concentration where the %CV of the calculated concentration is less than 20% and the percent recovery of the standard is between 80% and 120%.

MSD Advantage

- ➢ **Multiplexing:** Multiple analytes can be measured in one well using typical sample volumes of 50 μL or less without compromising speed or performance
- ➢ **Large dynamic range**: Linear range of up to five logs enables the measurement of native levels of biomarkers in normal and diseased samples without multiple dilutions
- ➢ **Minimal background:** The stimulation mechanism (electricity) is decoupled from the signal (light)
- ➢ **Simple protocols:** Only labels near the electrode surface are detected, enabling assays with fewer washes
- ➢ **Flexibility:** Labels are stable, non-radioactive, and conveniently conjugated to biological molecules
- ➢ **High sensitivity and precision:** Multiple excitation cycles of each label enhance light levels and improve sensitivity
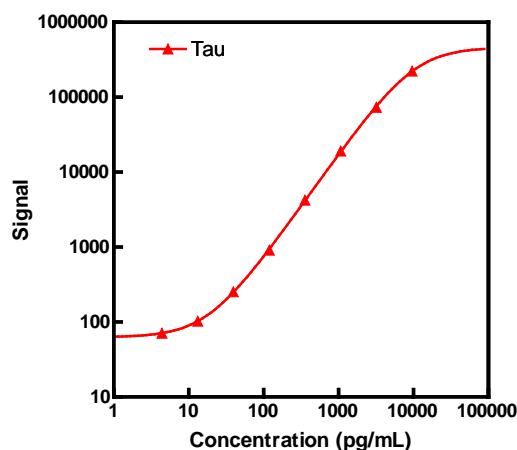
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Typical Standard Curve

The following standard curve is an example of the dynamic range of the Human Total Tau Assay.

| Tau		
Conc. (pg/mL)	Average Signal	%CV
0	57	17.1
4.39	71	8.7
13.2	103	6.8
39.5	253	6.0
119	913	6.5
356	4230	7.3
1067	19 164	6.2
3200	73 507	7.3
9600	223 102	6.2

Linearity

To assess linearity, CSF samples from normal and Alzheimer's disease (AD) individuals were diluted 2-fold, 4-fold, 8-fold, and 16-fold with Diluent 35 prior to testing. Measured concentrations were corrected for dilution factor to determine the actual total tau levels in the sample. Recovery at each dilution was calculated relative to the optimal sample dilution, 1:4.

Average percent recovery and range of recovery for normal and AD samples at each dilution are presented in the graph and table below. The graph of percent recovery versus dilution factor shows that a 2-fold dilution may be used for higher sensitivity with minimal effect on recovery. A minimum sample dilution of 2-fold is recommended.

% Recovery=(measured∗dilution factor)/(measured at 1:4 dilution∗4)∗100
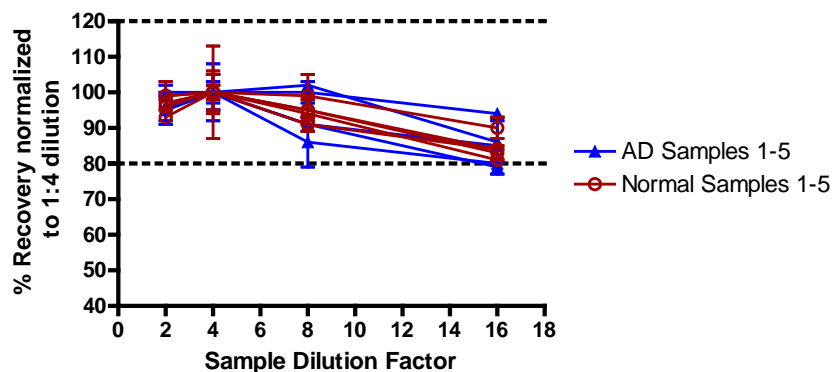
		Tau	
Sample	Fold Dilution	Average %Recovery	%Recovery Range
Normal CSF (N=5)	2	97	92–99
	4	100	N/A
	8	94	87–96
	16	89	83–93
AD CSF (N=5)	2	96	92–98
	4	100	N/A
	8	94	89–99
	16	87	83–93

Spike Recovery

CSF from normal and Alzheimer's disease (AD) individuals was spiked with calibrator at multiple levels throughout the range of the assay. The samples were then diluted 4-fold and tested for recovery.

% Recovery=measured/expected∗100

Sample	Tau		
	Spike Conc. (pg/mL)	Average %Recovery	%Recovery Range
Normal CSF (N=5)	4000	101	93–107
	1000	110	106–113
	250	105	99–109
AD CSF (N=5)	4000	101	97–104
	1000	112	106–116
	250	106	104–107

Samples

Normal and Alzheimer's disease (AD) individual patient CSF samples and pooled human CSF samples were purchased from commercial vendors. Sample collection methods and pre-analytical variables may cause variability in the measured range of normal and diseased samples. The individual patient samples were well-curated; handling was consistent with accepted protocols. Samples were diluted 4-fold prior to measuring with the Human Total Tau Kit. Aβ42 concentration in the samples was measured using MSD Human Aβ42 Kit. The table below displays median and range of concentrations for each sample set. Concentrations are corrected for sample dilution. A graphical representation is also provided for the individual normal and AD patient samples.

			Tau (pg/mL)
Well-curated, Individual, Human CSF Samples	Normal	Median (pg/mL)	214
		Range (pg/mL)	89–314
		# of Samples	10
		% of Samples in Quantitative Range	80%
	AD	Median (pg/mL)	590
		Range (pg/mL)	359–1096
		# of Samples	10
		% of Samples in Quantitative Range	100%
Pooled Human CSF		Median (pg/mL)	1560
		Range (pg/mL)	105–10 756
		# of Samples	10
		% of Samples in Quantitative Range	90%
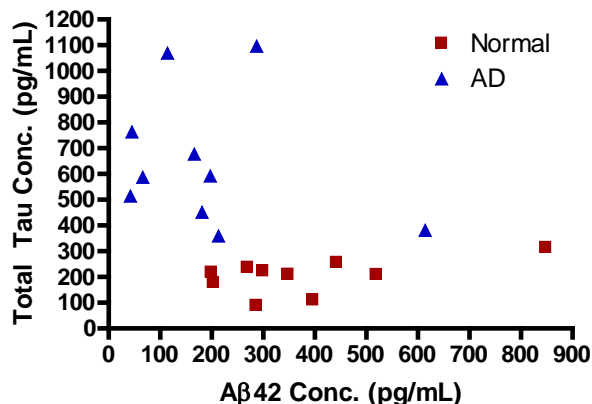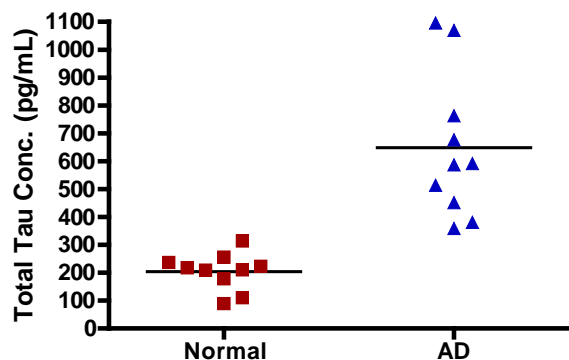
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MSD Neurodegenerative Disease Assays

Precision

Control samples were built using pooled human CSF with or without spiked tau calibrator. Two sets of control samples were prepared independently, using 4-fold dilutions. Each set contained three controls with total tau levels spanning the expected range of total tau in human CSF samples. Concentrations were measured for all controls using three independent Human Total Tau Kit lots. Representative data from one set of controls is presented in the tables below. For this study, four analysts ran tests over 10 days (N=26 runs across three kit lots). The control data for each kit lot and an inter-kit lot summary are presented in the upper table. Concentrations are dilution-adjusted.

Avg. Intra-plate Calc. Conc. %CV is the average concentration %CV of the control replicates on an individual plate. Inter-plate Calc. Conc. %CV is the variability of measured control concentration across plates, with replicate information as indicated in the table. Total error was calculated as the (Inter-plate Calc. Conc. %CV)+(absolute value of % Conc. Recovery Relative to Final Expected Concentration-100%). The %CV of each control should be <20%. Total error for controls should be <30%.

The concentrations presented in the inter-lot summary represent the assigned concentrations for each control. The lower table displays measured concentrations for each kit relative to the final expected concentration. Control concentrations measured by each kit should be within 20% of the expected concentration.

	Sample ID	Calc. Conc. (pg/mL)	Inter-plate Calc. Conc. %CV	Avg. Intra-plate Calc. Conc. %CV	% Total Error
Kit Lot 1 N=3	Control 1	4928	5.0	3.3	12
	Control 2	1167	3.0	2.5	3
	Control 3	304	6.3	3.2	9
Kit Lot 2 N=5	Control 1	4734	7.2	3.1	10
	Control 2	1118	2.8	3.3	7
	Control 3	260	4.8	5.4	17
Kit Lot 3 N=18	Control 1	4504	9.5	2.6	12
	Control 2	1184	7.2	5.3	8
	Control 3	305	18.6	5.0	21
Inter-Lot Summary N=26	Control 1	4598	9.0	2.4	
	Control 2	1170	6.5	4.3	
	Control 3	296	17.1	4.5	

	% Conc. Recovery Relative to Final Expected Concentration		
	Kit Lot 1	Kit Lot 2	Kit Lot 3
Control 1	107	103	98
Control 2	100	96	101
Control 3	103	88	103

References

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3. Hampel H, Frank R, Broich K, Teipel SJ, Katz RG, Hardy J, Herholz K, Bokde AL, Jessen F, Hoessler YC, Sanhai WR, Zetterberg H, Woodcock J, Blennow K. Biomarkers for Alzheimer's Disease: academic, industry, and regulatory perspectives. Nat Rev Drug Disc. 2010 July(9):560-74.
4. Blennow K, Hampel H, Weiner M, Zetterberg H. Cerebrospinal fluid and plasma biomarkers in Alzheimer disease. Nat Rev Neurol. 2010 Mar;6(3):131-44.
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