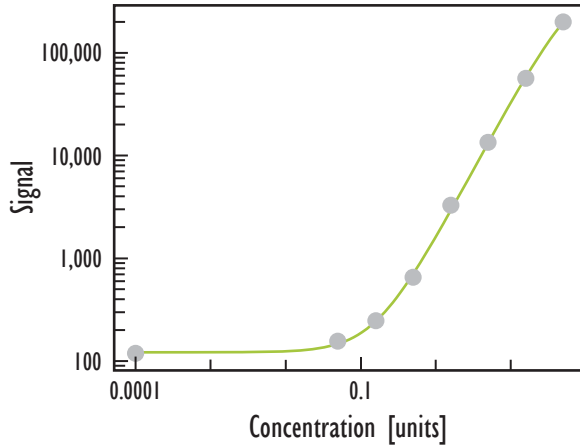
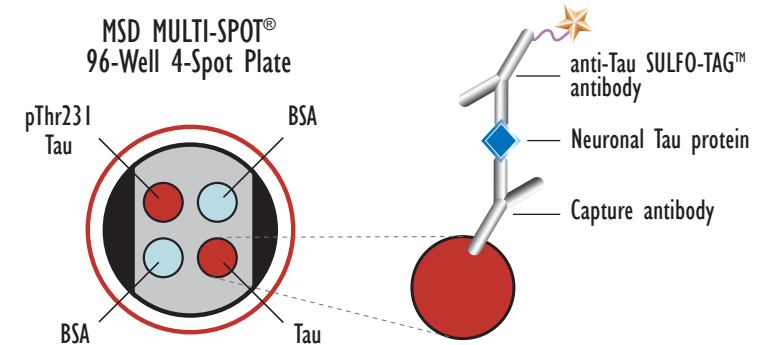
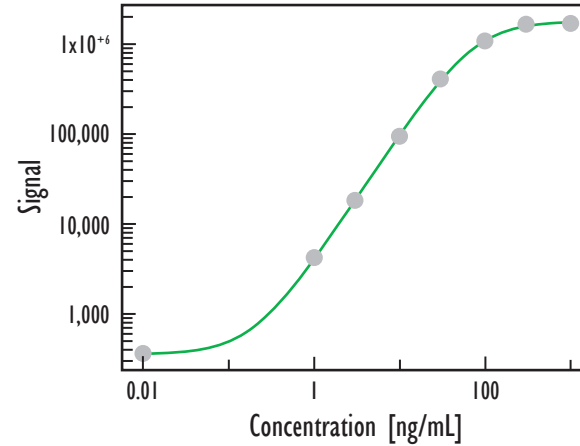


Quantification of a Neuronal Tau Standard

Phospho Tau (Thr231)



Total Tau



Purified neuronal Tau was added to MSD MULTI-SPOT 4 Spot 96-well plates coated with anti-total-Tau and anti-phospho-Tau antibodies on two of the four spatially distinct electrodes per well. Phosphorylated and total Tau were detected with anti-total-Tau antibody labeled with MSD SULFO-TAG reagent.

units pTau(231)	Phospho Tau (Thr231)			S/B
	Average	StdDev	%CV	
0	118	21	17	
0.05	156	22	14	1
0.16	247	10	4	2
0.5	654	48	7	6
1.6	3,272	135	4	28
5	13,396	576	4	113
16	56,554	2,512	4	478
50	199,993	4,688	2	1,690

Tau (ng/mL)	Total Tau			S/B
	Average	StdDev	%CV	
0	361	14	4	0
1	4,235	268	6	12
3	18,202	2,139	12	50
10	93,597	23,009	25	259
30	406,815	22,293	5	1,127
100	1,082,204	26,674	2	2,998
300	1,658,455	14,964	1	4,594
1,000	1,686,650	13,417	1	4,672

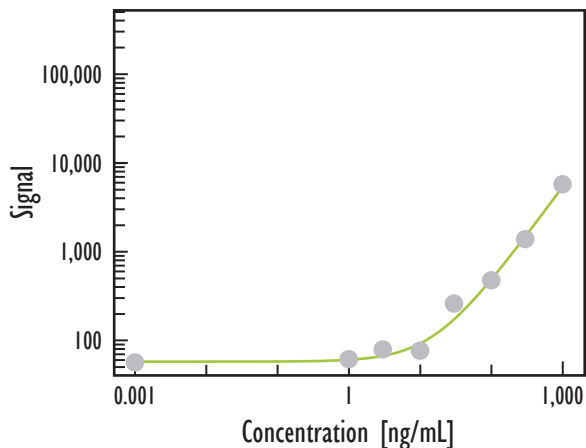
Detection Limits	
Phospho Tau (Thr231)	0.09 units*
Total Tau	46.1 pg/mL

* Phosphorylated Tau is quantified in units because the percentage of the total Tau that is phosphorylated will vary with different preparations of the standard.

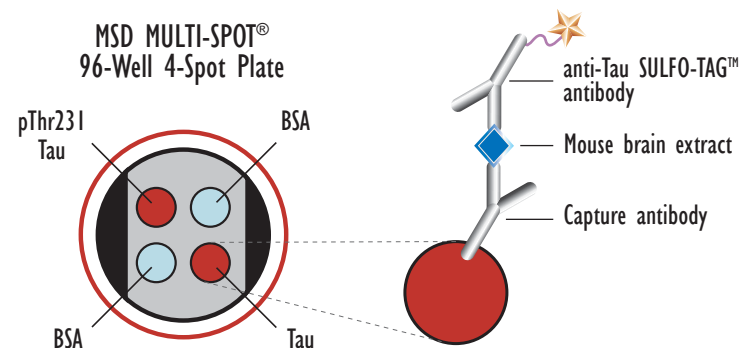
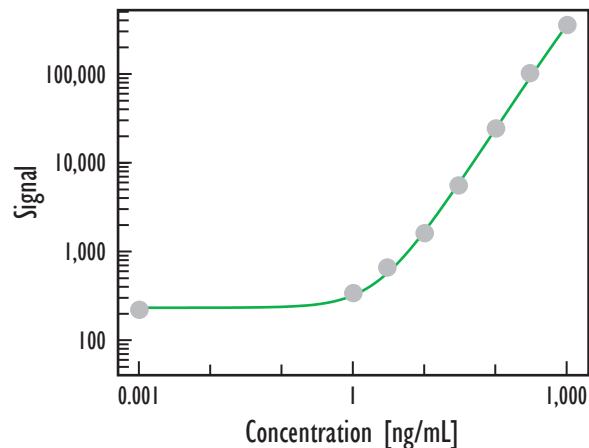
Kit Size	SI2400	SI6000
1 plate	K15121D-1	K11121D-1
5 plates	K15121D-2	K11121D-2
20 plates	K15121D-3	K11121D-3
20 plates (Base)	K15121A-3	K11121A-3

Detection of Phosphorylated (Thr231) and Total Tau in Mouse Brain Extract

Phospho Tau (Thr231)



Total Tau



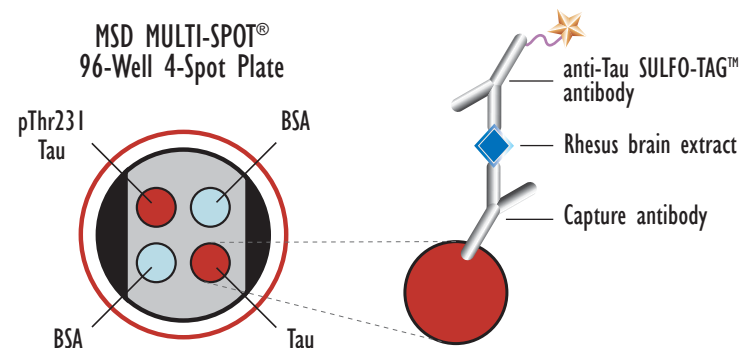
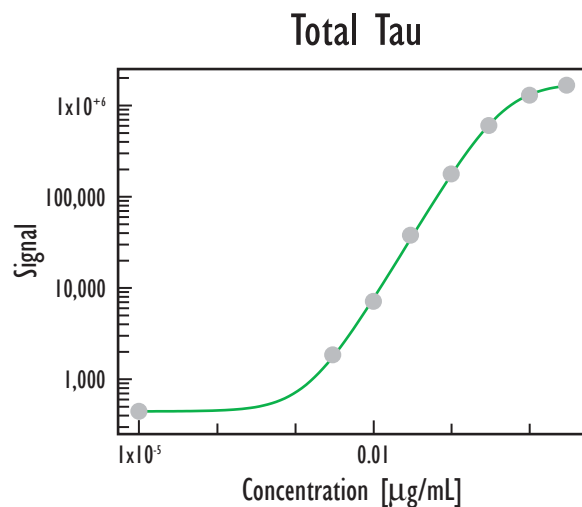
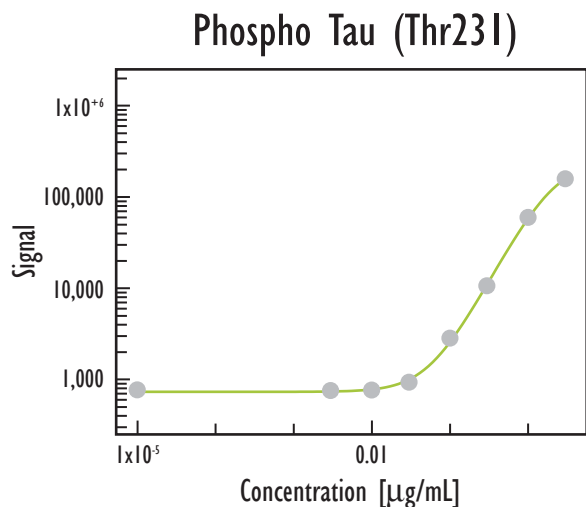
Extract (ng/well)	Phospho Tau (Thr231)			S/B
	Average	StdDev	%CV	
0	56	19	33	
1	61	31	50	1.1
3	79	17	21	1.4
10	76	26	33	1.4
30	259	40	15	4.6
100	475	59	12	8.4
300	1,380	166	12	24.5
1,000	5,726	680	12	101.6

Extract (ng/well)	Total Tau			S/B
	Average	StdDev	%CV	
0	218	37	17	
1	337	16	5	1.5
3	654	45	7	3.0
10	1,597	89	6	7.3
30	5,519	512	9	25.3
100	24,256	1,333	5	111.1
300	102,034	9,042	9	467.3
1,000	356,478	21,275	6	1,632.7

Mouse brain extract was prepared by homogenization of whole mouse brain in MSD Tris Lysis Buffer, followed by centrifugation to clear debris. Dilutions were prepared in MSD Tris Lysis Buffer. Samples were added to MSD MULTI-SPOT 4 Spot 96-well plates coated with anti-total-Tau and anti-phospho-Tau antibodies on two of the four spatially distinct electrodes per well. Phosphorylated and total Tau were detected with anti-total Tau antibody labeled with MSD SULFO-TAG reagent.

Detection Limit of Mouse Brain Extract [ng/well]	
Phospho Tau (Thr231)	15
Total Tau	1.1

Detection of Phosphorylated (Thr231) and Total Tau in Rhesus Brain Extract



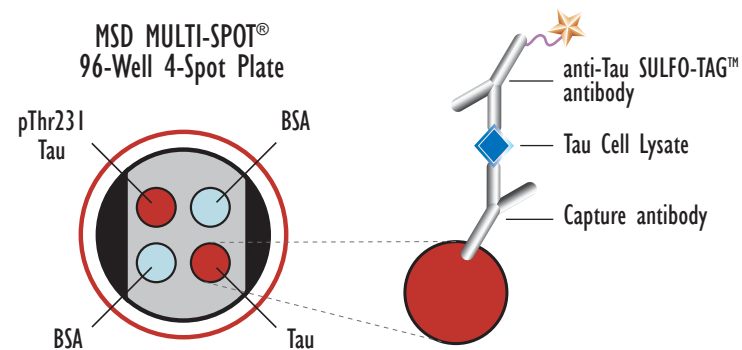
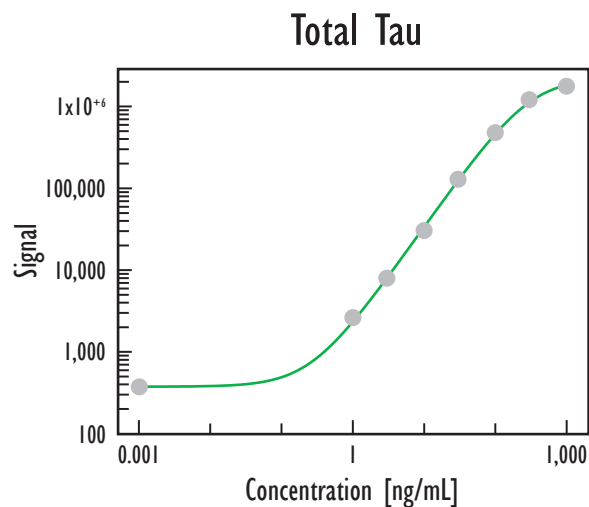
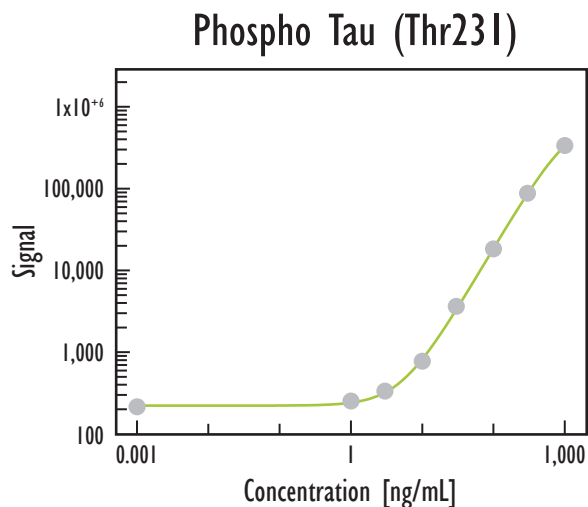
Extract (µg/well)	Phospho Tau (Thr231)			S/B
	Average	StdDev	%CV	
0	758	36	5	
0.003	750	34	5	1
0.01	767	42	5	1
0.03	929	75	8	1
0.1	2,832	537	19	4
0.3	10,530	1,086	10	14
1	59,382	2,367	4	78
3	157,825	6,848	4	208

Extract (µg/well)	Total Tau			S/B
	Average	StdDev	%CV	
0	442	42	9	
0.003	1,830	130	7	4
0.01	7,083	531	7	16
0.03	37,746	1,743	5	85
0.1	176,684	4,303	2	400
0.3	600,636	23,678	4	1,358
1	1,296,457	47,217	4	2,932
3	1,668,563	91,566	5	3,773

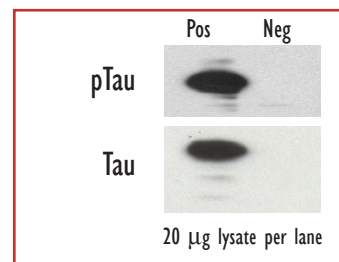
Detection Limit [ng/well]	
Phospho Tau (Thr231)	19.5
Total Tau	0.6

Rhesus brain extract was prepared by homogenization of rhesus brain tissue in MSD Tris Lysis Buffer, followed by centrifugation to clear debris. Dilutions were prepared in MSD Tris Lysis Buffer. Samples were added to MSD MULTI-SPOT 4 Spot 96-well plates coated with anti-total-Tau and anti-phospho-Tau antibodies on two of the four spatially distinct electrodes per well. Phosphorylated and total Tau were detected with anti-rhesus Tau antibody labeled with MSD SULFO-TAG reagent.

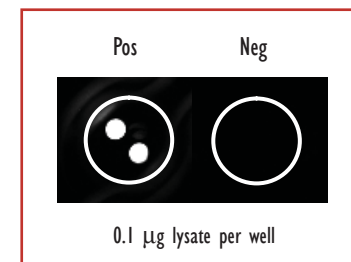
Detection of Phosphorylated (Thr231) and Total Tau in Human Tau-441 Overexpressing CHO Cells



Traditional Western



MSD Experimental



Extract (ng/well)	Phospho Tau (Thr231)			S/B
	Average	StdDev	%CV	
0	214	14	7	
1	252	27	11	1.2
3	333	8	2	1.6
10	774	75	10	3.6
30	3,621	344	9	16.9
100	18,233	812	4	85.1
300	87,296	5,858	7	407.3
1,000	335,282	26,140	8	1,564.3

Extract (ng/well)	Total Tau			S/B
	Average	StdDev	%CV	
0	369	43	12	
1	2,605	241	9	7.1
3	7,911	29	0	21.4
10	30,304	3,311	11	82.1
30	129,035	16,129	13	349.4
100	481,056	35,082	7	1,302.5
300	1,218,344	62,380	5	3,298.8
1,000	1,778,074	102,560	6	4,814.3

Detection Limit of Lysate [pg/well]	
Phospho Tau (Thr231)	1,480
Total Tau	108

CHO cells were transfected with a human Tau-441 expression plasmid and harvested after 48 hrs. Whole cell lysates were added to MSD MULTI-SPOT 4 Spot 96-well plates coated with anti-total-Tau and anti-phospho-Tau antibodies on two of the four spatially distinct electrodes per well. Phosphorylated and total Tau were detected with anti-total-Tau antibody labeled with MSD SULFO-TAG reagent.

