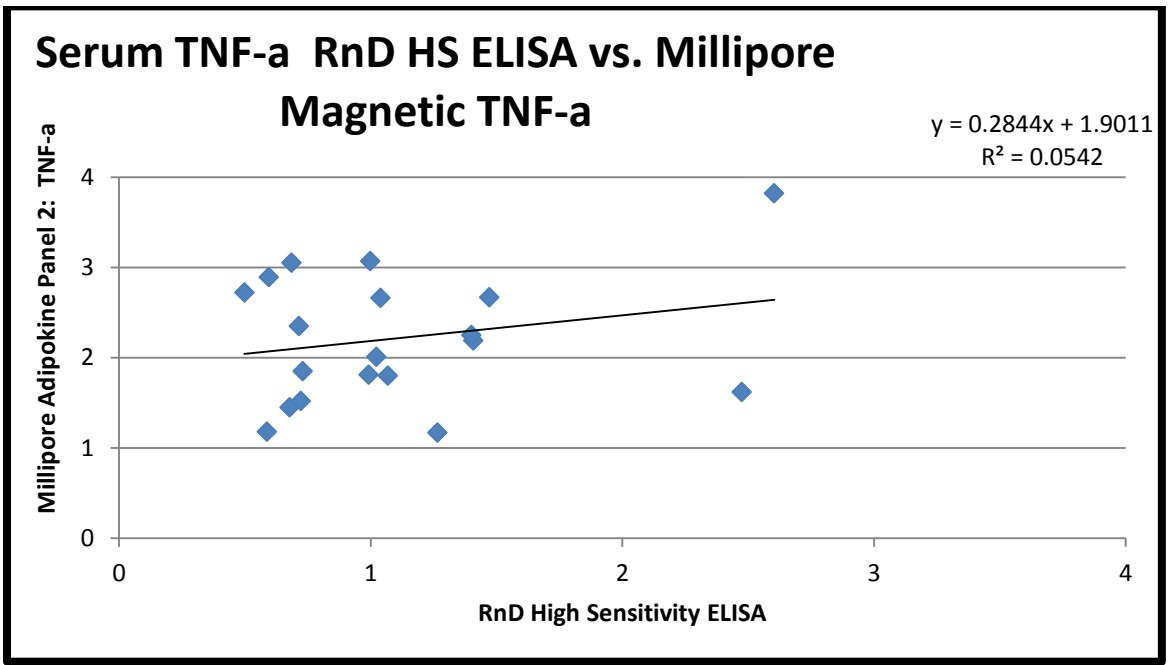


Units:	pg/mL	pg/mL
Assay Name	RnD High Sensitivity ELISA	Millipore Adipokine Panel 2: TNF-a
Catalog Number	HSTA00D	HADK2MAG-61K
Other Analytes On Multiplex	N/A	NGF, IL-6, Insulin, Leptin, IL-8, HGF, MCP-1, IL-1b
Volume Required Per Replicate	200uL	25uL
Typical Dilution	none	none
Manufacturer Defined Minimum Detectible Concentration	0.106 pg/mL	0.30 pg/mL
UVM Observed Minimum Detectible Concentration	0.45 pg/mL	0.30 pg/mL
UVM Run Date	11/7/2008	5/21/2014
SCS080504: 1	0.595	2.890
SCS080504: 2	1.408	2.190
SCS080504: 3	1.038	2.660
SCS080504: 4	2.603	3.820
SCS080504: 5	1.472	2.670
SCS080504: 6	0.715	2.350
SCS080504: 7	0.992	1.810
SCS080504: 8	0.498	2.720
SCS080504: 9	0.730	1.850
SCS080504: 10	0.677	1.450
SCS080504: 11	2.475	1.620
SCS080504: 12	0.685	3.050
SCS080504: 13	0.999	3.070
SCS080504: 14	0.587	1.180
SCS080504: 15	1.400	2.250
SCS080504: 16	1.022	2.010
SCS080504: 17	0.723	1.520
SCS080504: 18	1.069	1.800
SCS080504: 19	1.265	1.170
SCS080504: 20	1.552	2.560
<b>Average</b>	<b>1.125</b>	<b>2.232</b>



**Notes:**

Antibody sources may be different

Correlations may be different at higher levels of TNF-a

Only data for RnD HS ELISA (11-7-2008), accounting for different lot n=20 serum set than other correlation

Units:	pg/mL	pg/mL	pg/mL	pg/mL	pg/mL	pg/mL	pg/mL
Assay Name	Millipore Adipokine Panel 2: TNF-a	Millipore Magnetic Metabolic Hormone 13-Plex: TNF-a	Millipore High Sensitivity T-Cell Panel: TNF-a	Millipore Magnetic Cytokine Panel: TNF-a	Millipore Magnetic High Sensitivity Cytokine Panel: TNF-a	RnD High Sensitivity Cytokine Panel B	MESO Scale Ultra High Sensitive Cytokine Panel
Catalog Number	HADK2MAG-61K	HMHEMAG-34K	HSTCMAG-28SK	HCYTOMAG-60K	HSCYTMAG-60SK	LHSCM210	K15007C
Other Analytes On Multiplex	NGF, IL-6, Insulin, Leptin, IL-8, HGF, MCP-1, IL-1b	Amylin (Total), C-Peptide, Ghrelin, GIP, GLP Active, GLP Total, Glucagon, IL-6, Insulin, Leptin, MCP-1, PP, PYY, Amylin (active)	ITAC, GM-CSF, Fractalkine, IFNg, IL-10, MIP-3a, IL-12p70, IL-13, IL-17A, IL-1b, IL-2, IL-21, IL-4, IL-23, IL-5, IL-6, IL-7, IL-8, MIP-1a, MIP-1b	EGF, FGF-2, Eotaxin, TGF-a, G-CSF, Flt-3L, GM-CSF, Fractalkine, IFNa2, GRO, IL-10, MCP-3, IL-12p40, MDC, IL-12p70, PDGF-AA, IL-13, PDGF-BB, IL-15, sCD40L, IL-17, IL-1RA, IL-2Ra, IL-1a, IL-9, IL-1b, IL-2, IL-3, IL-4, IL-5, IL-6, IL-7, IL-8, IP-10, MCP-1, MIP-a, MIP-b, RANTES, TNF-a, TNF-b, VEGF	IL-1b, IL-2, IL-4, IL-5, IL-6, IL-7, IL-8, IL-10, IL-12p70, IL-13, IFNg, GM-CSF	GM-CSF, IFNg, IL-1b, IL-2, IL-5, IL-6, IL-7, IL-13, IL-15, IL-17A, IL-17F, IL-22, IL-23, IL-31, IL-33, IL-36B	IL-2, IL-8, IL-12p70, IL-1b, GM-CSF, IFNg, IL-6, IL-10
Volume Required Per Replicate	25uL	25uL	25uL	25uL	50uL	150uL	25uL
Typical Dilution	none	none	none	none	none	1:2	None
Manufacturer Defined Minimum Detectible Concentration	0.30 pg/mL	0.10 pg/mL	0.16 pg/mL	0.70 pg/mL	0.07 pg/mL	0.29 pg/mL	0.48 pg/mL
UVM Observed Minimum Detectible Concentration	0.30 pg/mL	0.23 pg/mL	0.16 pg/mL	0.70 pg/mL	0.07 pg/mL	0.551 pg/mL	
UVM Run Date	5/21/2014	10/9/2015	7/14/2015	3/7/2013	3/7/2013	5/2/2014	1/17/2013
SCS081611: 1	3.350	5.220	2.540	8.120	6.560	4.690	1.684
SCS081611: 2	5.010	4.420	3.180	9.440	8.850	5.320	2.065
SCS081611: 3	1.470	1.270	1.000	4.890	4.380	4.060	1.452
SCS081611: 4	2.260	2.150	0.940	4.050	4.600	1.780	0.950
SCS081611: 5			0.580	0.760	0.320	3.130	1.189
SCS081611: 6	2.250	2.290	1.650	4.570	4.520	2.270	1.111
SCS081611: 7	0.820	0.490	1.370	2.550	2.040	2.940	1.028
SCS081611: 8	5.040	3.850	3.070	7.960	8.740	3.510	1.787
SCS081611: 9	3.320	2.920	1.300	6.310	6.800	2.750	1.516
SCS081611: 10	1.730	1.450	1.490	3.910	3.700	2.270	1.070
SCS081611: 11	3.410	2.330	1.690	7.110	8.150	1.370	1.028
SCS081611:12	0.960	0.940	1.250	3.120	2.260	3.130	0.949
SCS081611: 13	4.360	3.300	2.780	7.820	7.200	4.240	1.844
SCS081611: 14	2.440	2.100	1.420	4.890	4.980	3.510	1.272
SCS081611: 15	2.470	2.430	1.850	5.580	5.580	3.880	1.691
SCS081611: 16	2.420	1.910	1.420	5.220	4.340	2.170	1.335
SCS081611: 17	4.700	4.520	6.540	8.500	8.760	1.580	1.093
SCS081611: 18	1.920	1.960	2.330	6.480	6.000	2.370	2.009
SCS081611: 19	3.130	2.430	2.100	6.010	5.710	3.130	1.379
SCS081611: 20	2.420	1.940	1.450	4.950	4.300	2.370	1.210
Average	2.815	2.522	1.998	5.612	5.390	3.024	1.383

# TNF-a Method Comparison - Quick Glance R2 Correlation Values

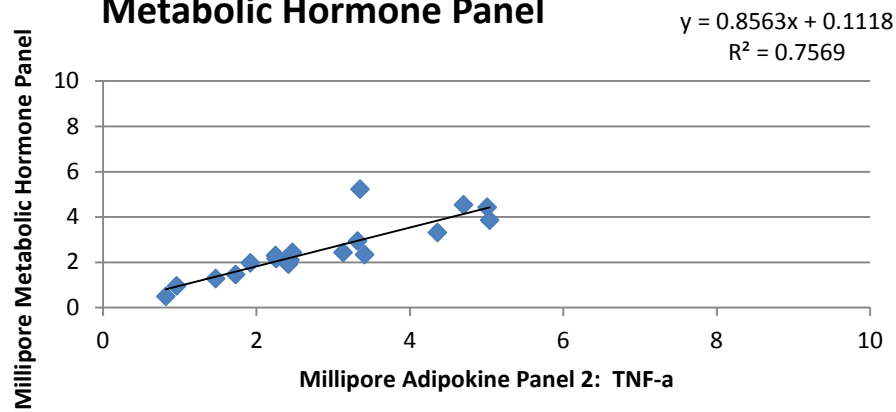
R <sup>2</sup> =	Millipore Adipokine Panel 2	Millipore Metabolic Hormone Panel	Millipore High Sensitivity T-Cell Panel	Millipore Magnetic Cytokine Panel	Millipore High Sensitivity Cytokine Panel	RnD High Sensitivity Cytokine Panel B	MESO Scale Ultra High Sensitive Cytokine Panel
Millipore Adipokine Panel 2	1	0.7569	0.4847	0.8413	0.8781	0.0648	0.2554
Millipore Metabolic Hormone Panel	0.7569	1	0.4898	0.8102	0.6937	0.1337	0.267
Millipore High Sensitivity T-Cell Panel	0.4847	0.4898	1	0.5083	0.4812	0.00007	0.069
Millipore Magnetic Cytokine Panel	0.8413	0.8102	0.5083	1	0.9286	0.083	0.4018
Millipore High Sensitivity Cytokine Panel	0.8781	0.6937	0.4812	0.9286	1	0.0187	0.2843
RnD High Sensitivity Cytokine Panel B	0.0648	0.1337	0.00007	0.083	0.0187	1	0.4482
MESO Scale Ultra High Sensitive Cytokine Panel	0.2554	0.267	0.069	0.4018	0.2843	0.4482	1

Notes:

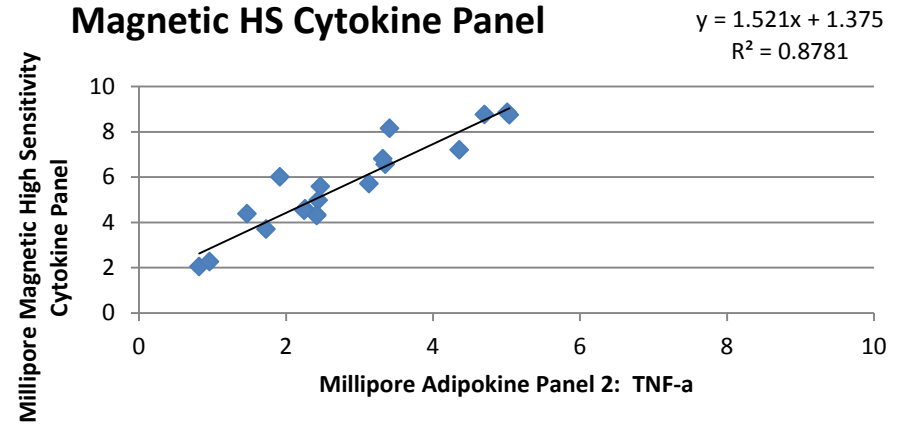
All samples are serum, and from the n=20 serum set lot 081611 (aka: SCS081611) All samples from same processing methods including: Normal Pooling, Aliquoting, Ultra-Cold Storage, Thawing and Room Temperature Exposure. Alternate populations and/or draw tube type and/or specimen condition may yield different results based on the levels of endogenous TNF-a (i.e. matrix and/or standard curve level)

Outliers may negatively impact R2 correlation values. See scatter plots in the following pages for in-depth review.

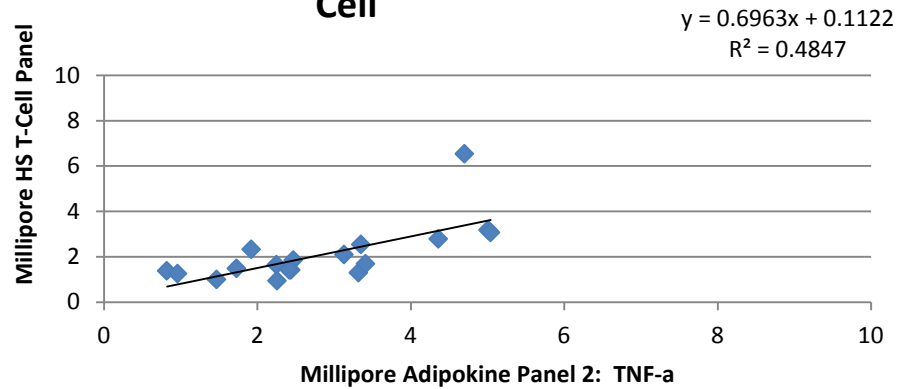
**Serum TNF-a: Adipokine Panel 2 vs. Metabolic Hormone Panel**



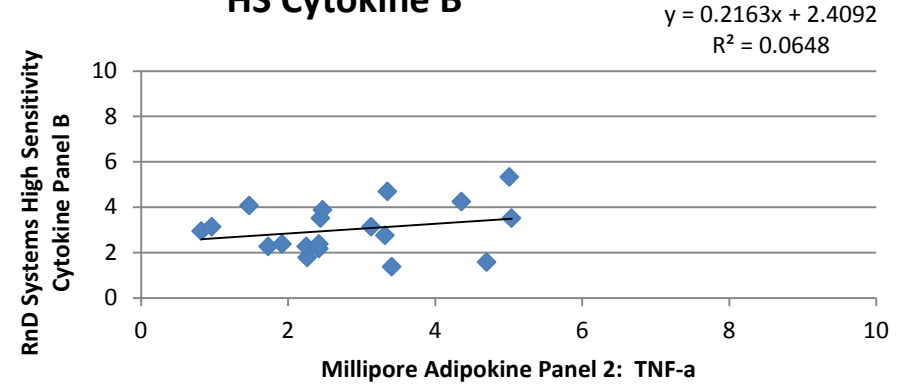
**Serum TNF-a: Adipokine Panel 2 vs. Magnetic HS Cytokine Panel**



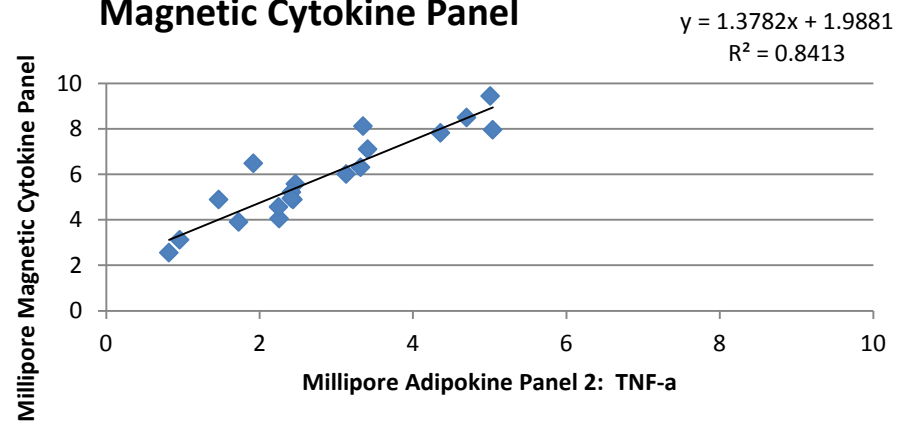
**Serum TNF-a: Adipokine Panel 2 vs. HS T-Cell**



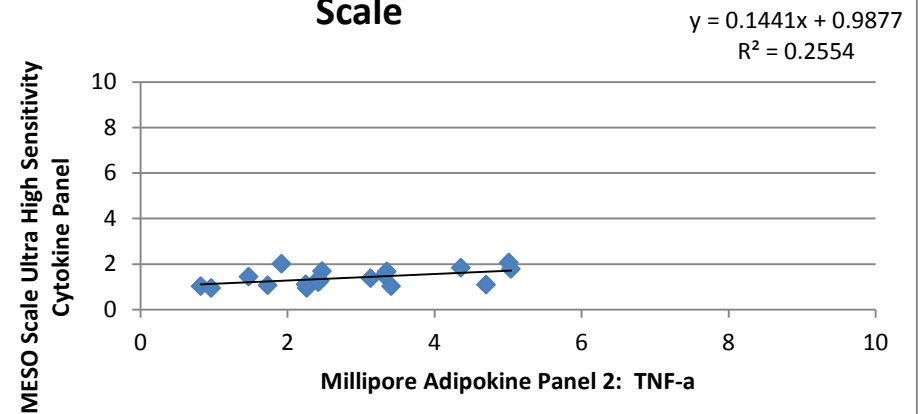
**Serum TNF-a: Adipokine Panel 2 vs. RnD HS Cytokine B**



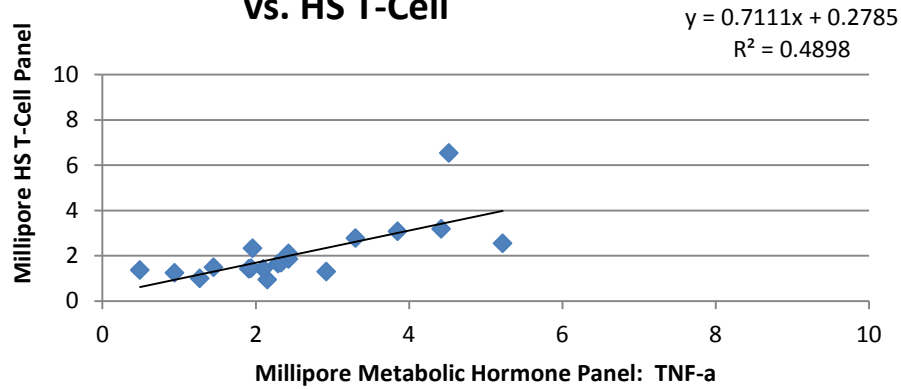
**Serum TNF-a: Adipokine Panel 2 vs. Magnetic Cytokine Panel**



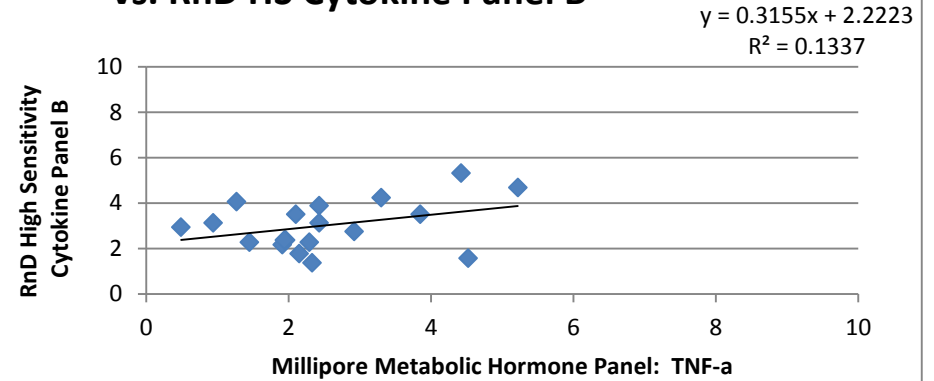
**Serum TNF-a: Adipokine Panel 2 vs. MESO Scale**



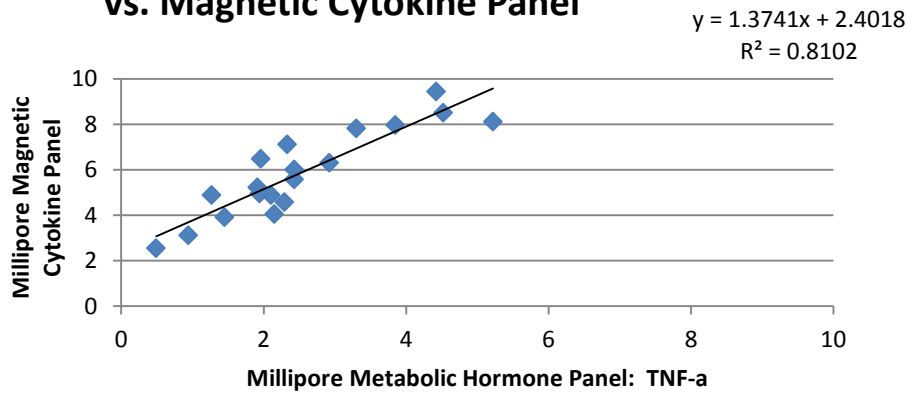
### Serum TNF-a: Metabolic Hormone Panel vs. HS T-Cell



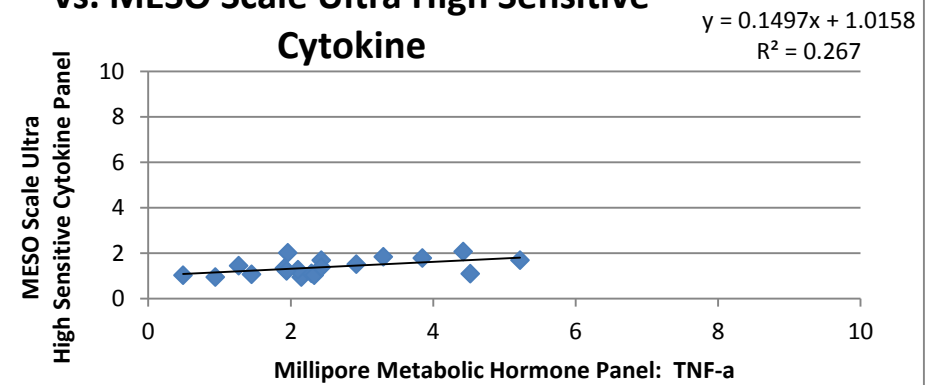
### Serum TNF-a: Metabolic Hormone Panel vs. RnD HS Cytokine Panel B



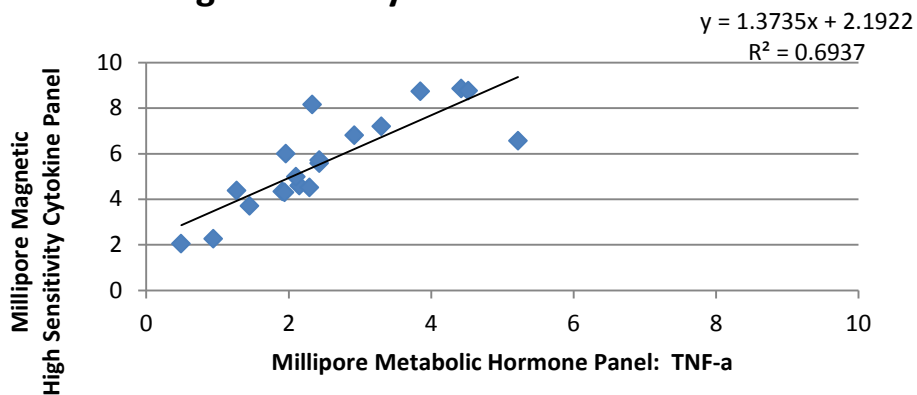
### Serum TNF-a: Metabolic Hormone Panel vs. Magnetic Cytokine Panel



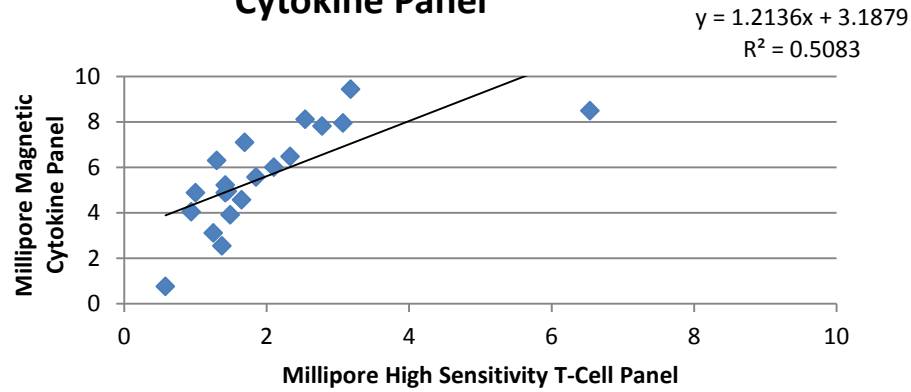
### Serum TNF-a: Metabolic Hormone Panel vs. MESO Scale Ultra High Sensitive Cytokine



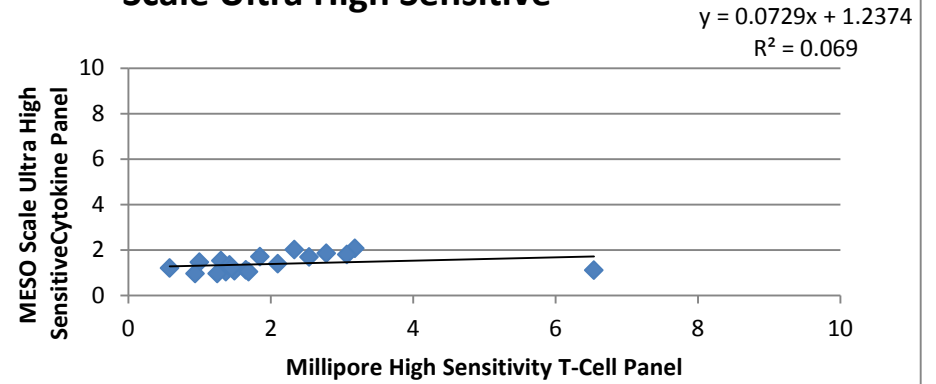
### Serum TNF-a: Metabolic Hormone Panel vs. Magnetic HS Cytokine Panel



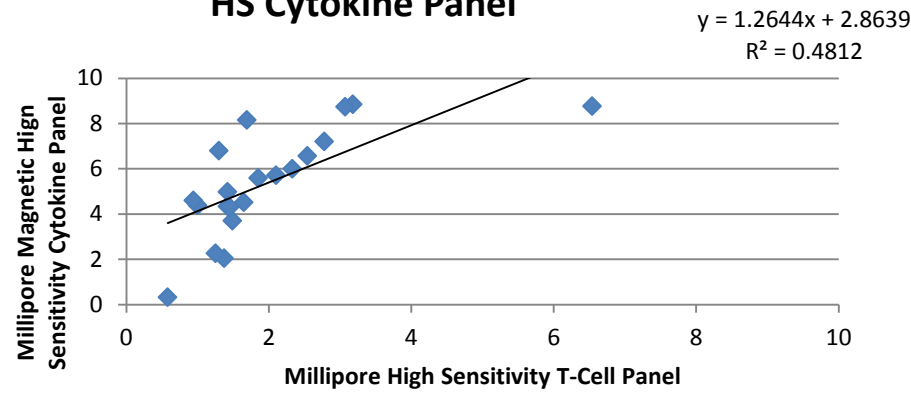
**Serum TNF-a: HS T-Cell Panel vs. Magnetic Cytokine Panel**



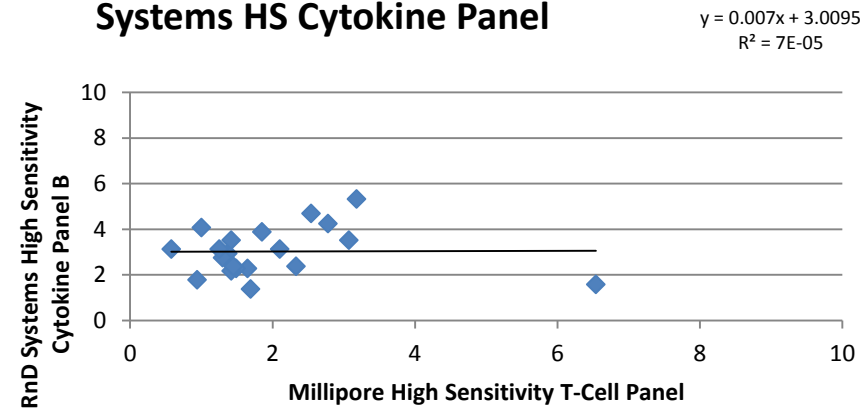
**Serum TNF-a: HS T-Cell Panel vs. MESO Scale Ultra High Sensitive**



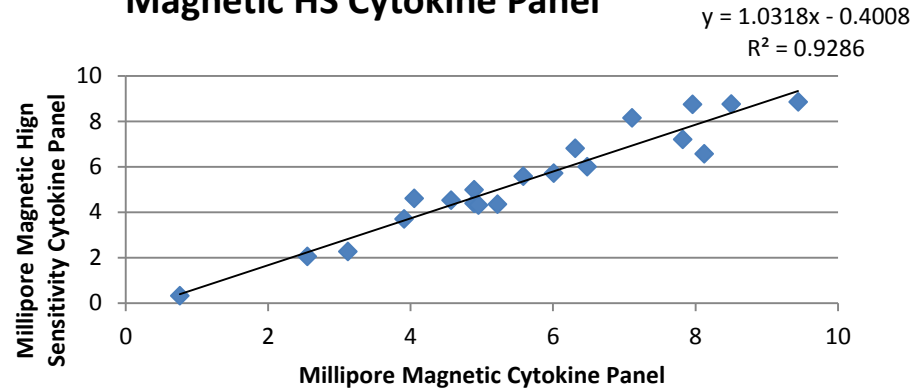
**Serum TNF-a: HS T-Cell Panel vs. Magnetic HS Cytokine Panel**



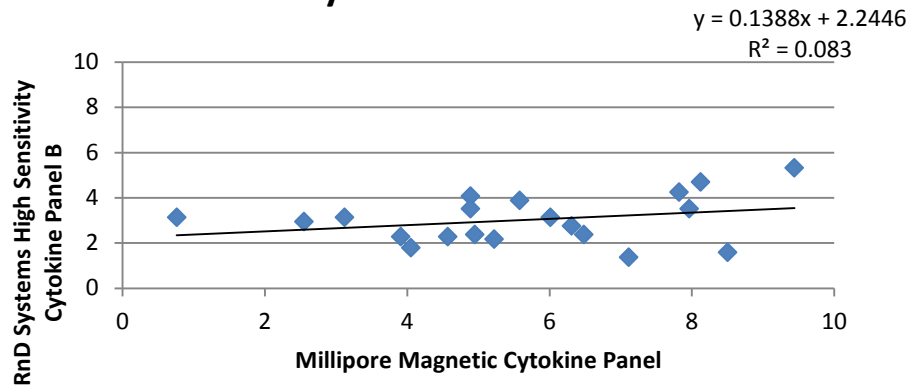
**Serum TNF-a: HS T-Cell Panel vs. RnD Systems HS Cytokine Panel**



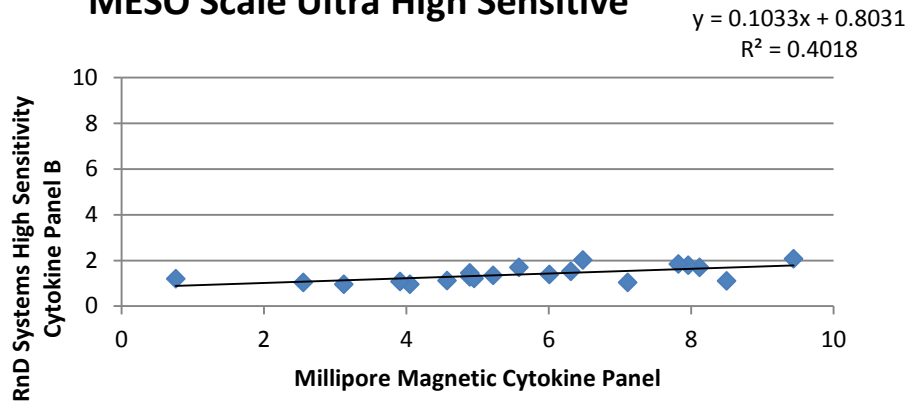
### Serum TNF-a: Magnetic Cytokine Panel vs. Magnetic HS Cytokine Panel

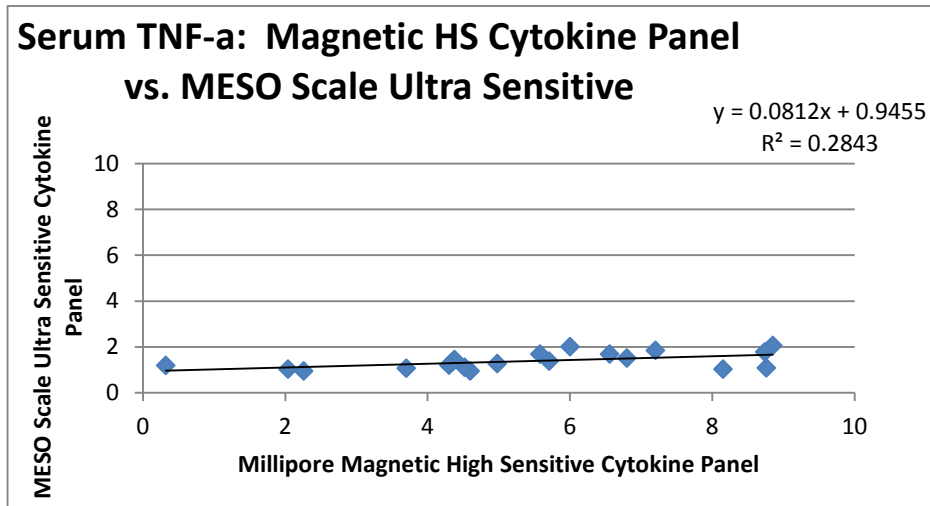
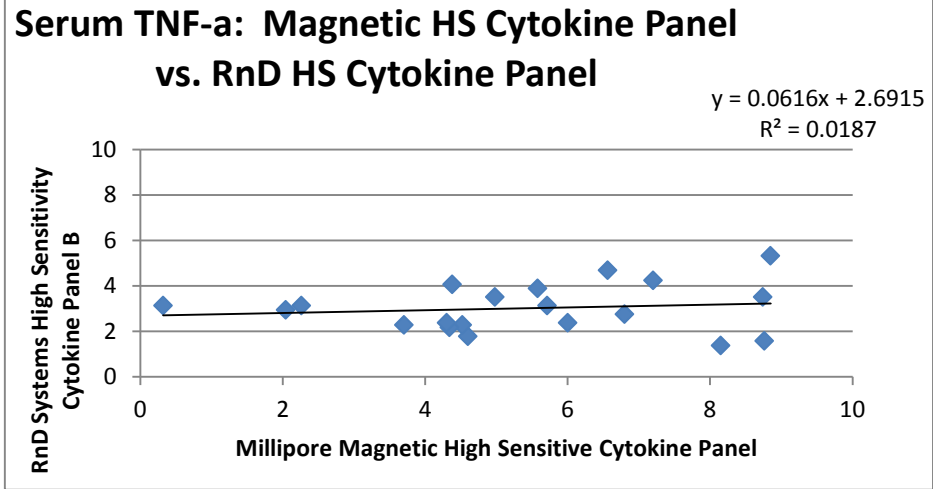


### Serum TNF-a: Magnetic Cytokine Panel vs. RnD HS Cytokine Panel



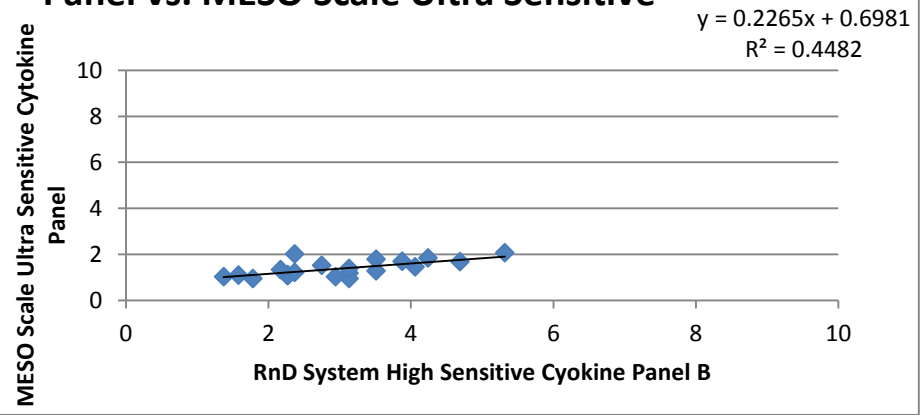
### Serum TNF-a: Magnetic Cytokine Panel vs. MESO Scale Ultra High Sensitive







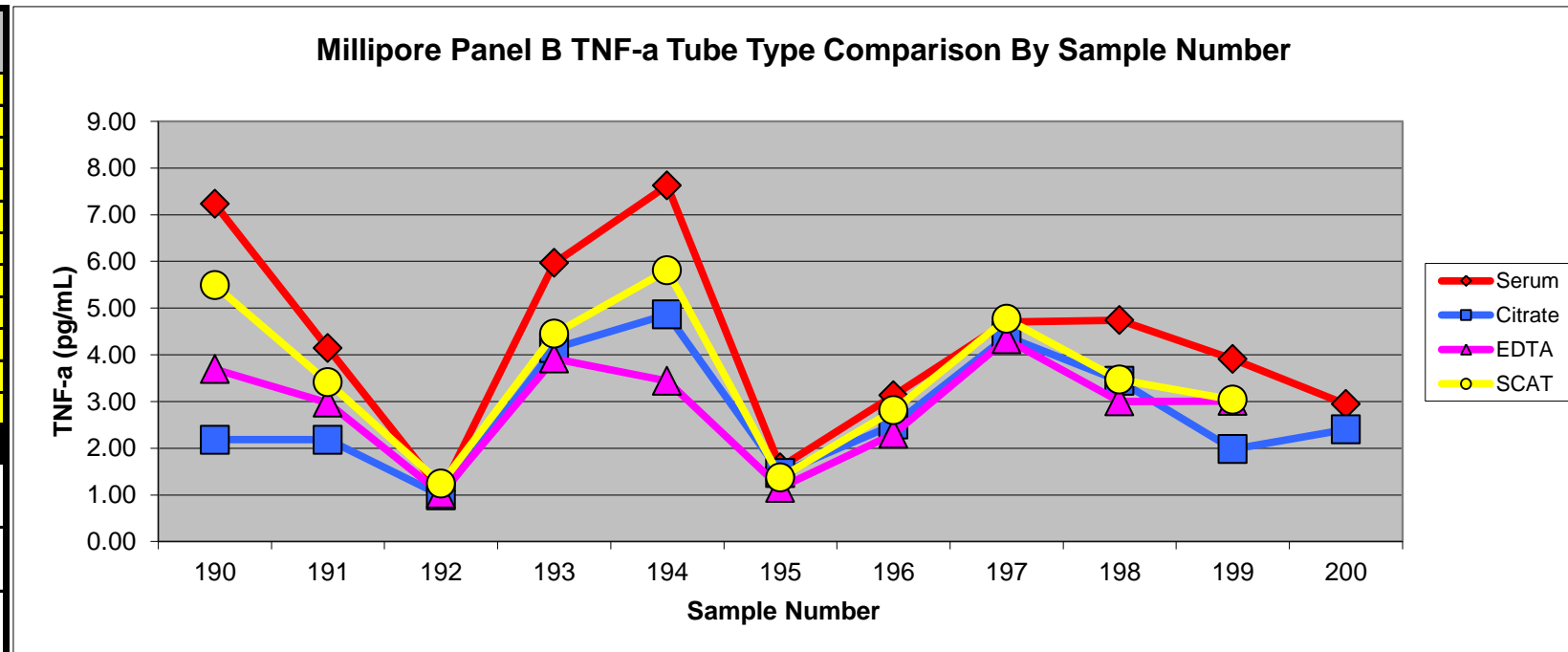
### Serum TNF-a: RnD System High Sensitive Panel vs. MESO Scale Ultra Sensitive



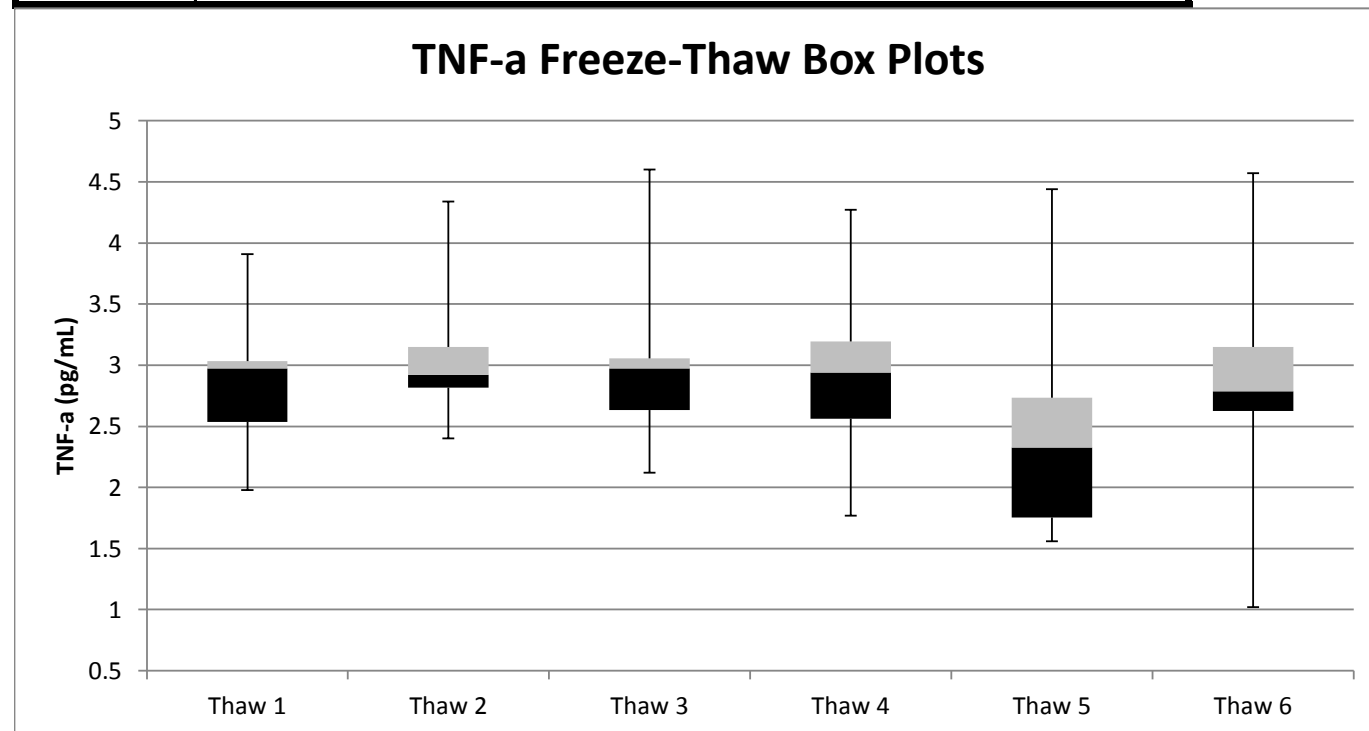
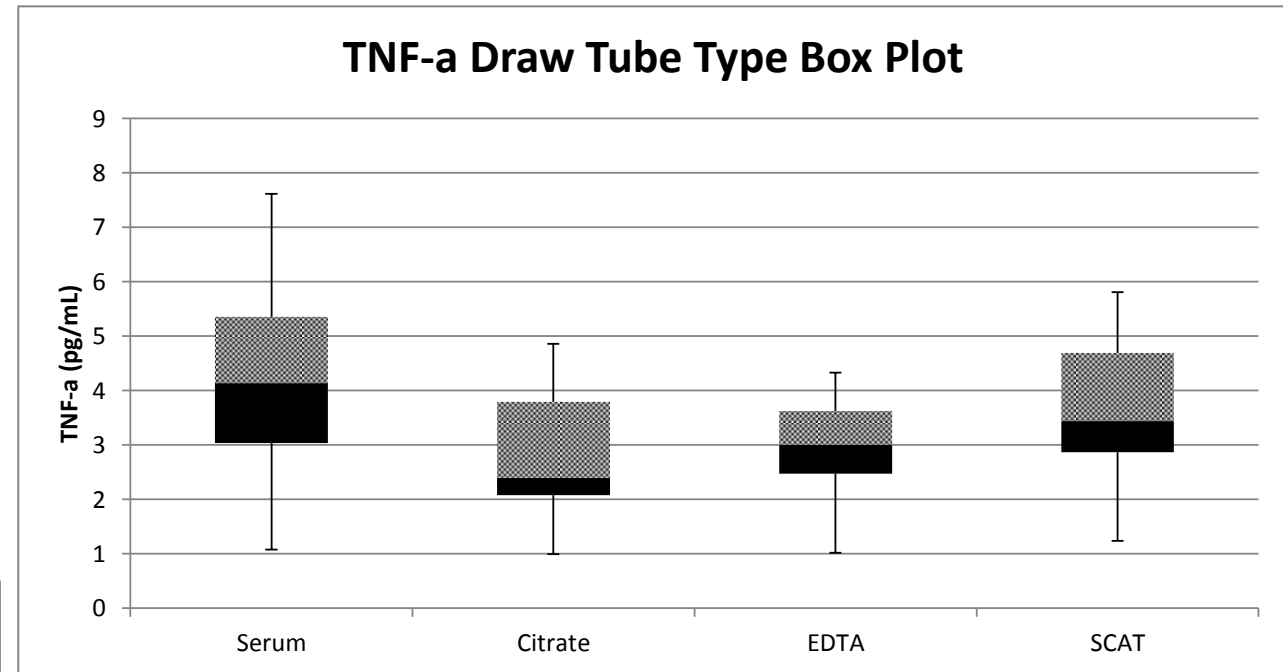
**Millipore Panel B  
TNF-a**

**Tube Type and Freeze Thaw Comparison**

Sample Number	Serum	Citrate	EDTA	SCAT
190	7.23	2.18	3.69	5.49
191	4.14	2.18	2.97	3.41
192	1.08	0.99	1.02	1.24
193	5.97	4.14	3.92	4.45
194	7.62	4.86	3.44	5.81
195	1.59	1.46	1.15	1.37
196	3.13	2.50	2.31	2.81
197	4.70	4.39	4.33	4.77
198	4.74	3.44	3.00	3.47
199	3.91	1.98	3.01	3.04
200	2.94	2.40	3.01	3.04
<b>Average</b>	<b>4.28</b>	<b>2.77</b>	<b>2.88</b>	<b>3.59</b>
Sample Type Average	3.38			
Sample Type Stdev	0.70			
Sample Type CV%	20.64			



Sample Number	Thaw 1	Thaw 2	Thaw 3	Thaw 4	Thaw 5	Thaw 6
199 Serum	3.91	4.34	4.60	4.27	4.44	4.57
199 Citrate	1.98	2.80	2.53	2.50	2.53	2.60
199 EDTA	3.01	2.87	2.94	2.74	1.56	2.87
199 SCAT	3.04	3.21	3.01	3.21	2.12	3.24
200 Serum	2.94	2.97	3.07	3.14	2.80	2.70
200 Citrate	2.40	2.40	2.12	1.77	1.63	1.02
<b>Average</b>	<b>2.88</b>	<b>3.10</b>	<b>3.05</b>	<b>2.94</b>	<b>2.51</b>	<b>2.83</b>
Freeze/Thaw Average	2.88					
Freeze/Thaw Stdev	0.21					
Freeze/Thaw CV%	7.18					



Range of Standard Curve: 0.13 - 10,000 pg/mL
Manufacturer Defined Minimum Detectable Concentration: 0.14 pg/mL
Manufacturer Defined Intra-Assay CV% = 1.4 - 7.9 %
Manufacturer Defined Inter-Assay CV% <21%
Manufacturer Recommended Sample Type: Serum or Plasma
There appears to be a great deal of variance amongst some samples (190, 194) but not others (192, 196, 197) that may be contributing to the apparent sample type difference. Furthermore, most of these samples are reading at the bottom of the curve where there is a known increase in sample variance. Finally, there does not appear to be much effect from the freeze/thaw data. The CV% (7.18%) falls into the expected range for Intra-Assay CV%. The differences in Thaws may be attributed to nothing more than random bounce.