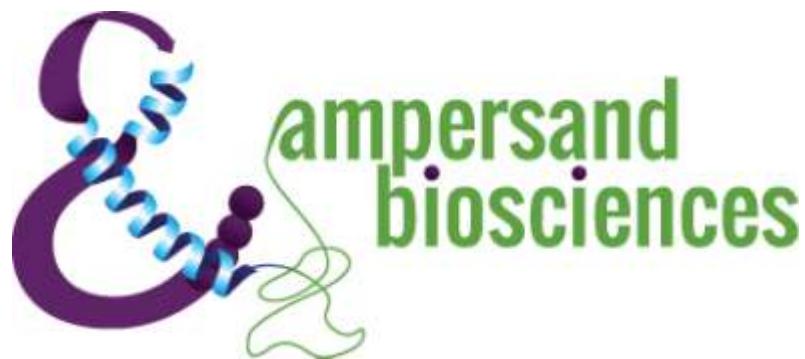


Mouse High Sensitivity Cytokine Panel 1

Kit # M102-K

Validation Report Version 1.0

May 13, 2024



## **1. Assay Description:**

A multiplex assay was developed and validated for the measurement of Mouse IFN $\gamma$ , IL-1 $\alpha$ , IL-1 $\beta$ , IL-10, IL-12p40, IL-12p70, IL-13, IL-2, IL-4, IL-5, IL-6, KC/GRO and TNF $\alpha$ . The kit is microsphere-based and consist of using antigen-specific antibodies covalently coupled to magnetic Luminex beads and biotinylated detection antibodies in a capture-sandwich format. All incubations take place at room temperature in a 96-well plate. 30  $\mu$ L of standard, controls or sample are added to the appropriate wells, followed by 10  $\mu$ L of blocker and 10  $\mu$ L of multiplexed capture-antibody microspheres. The plate is incubated for 2 hours at ambient temperature on a plate shaker. After washing 3 times, 40 $\mu$ L of detection antibodies are added to each well, thoroughly mixed, and incubated 1 hour at ambient temperature on a plate shaker. The Streptavidin-Phycoerythrin conjugate (SA-PE) working solution is then added to the plate and incubated for 30 minutes. The plate is then washed 3 times and the beads are resuspended in 100  $\mu$ L of wash buffer. After shaking on a plate shaker for 5 minutes, the plate is then analyzed on the Luminex 200 Analyzer.

## **2. Control and Sample Description:**

Control	Description
Control 1	Normal Mouse Serum (33%) spiked with Recombinant IL-1 $\alpha$ , IL-1 $\beta$ , IL-10, IL-12p70, IL-13, IL-2, IL-4, IL-5, and IL-6.
Control 2	Normal Mouse Serum (8.3%) spiked with Recombinant IFN $\gamma$ , IL-1 $\alpha$ , IL-1 $\beta$ , IL-10, IL-12p40, IL-12p70, IL-13, IL-2, IL-4, IL-5, IL-6, KC/GRO, and TNF $\alpha$ .

Sample	Description
Serum 1	Normal Mouse Serum spiked with Recombinant IFN $\gamma$ , IL-1 $\alpha$ , IL-1 $\beta$ , IL-10, IL-12p40, IL-12p70, IL-13, IL-2, IL-4, IL-5, IL-6, KC/GRO, and TNF $\alpha$ .
Serum 2	Normal Mouse Serum spiked with Recombinant IL-1 $\alpha$ , IL-1 $\beta$ , IL-12p70, IL-13, and IL-5.
Plasma 1	Normal Mouse Plasma spiked with Recombinant IL-1 $\beta$ , IL-12p70, and IL-5.
Plasma 2	Normal Mouse Plasma spiked with Recombinant IL-1 $\beta$ , IL-12p70, IL-13, IL-2, IL-4, and IL-5.

## **3. LLOQ, LDD and Curves:**

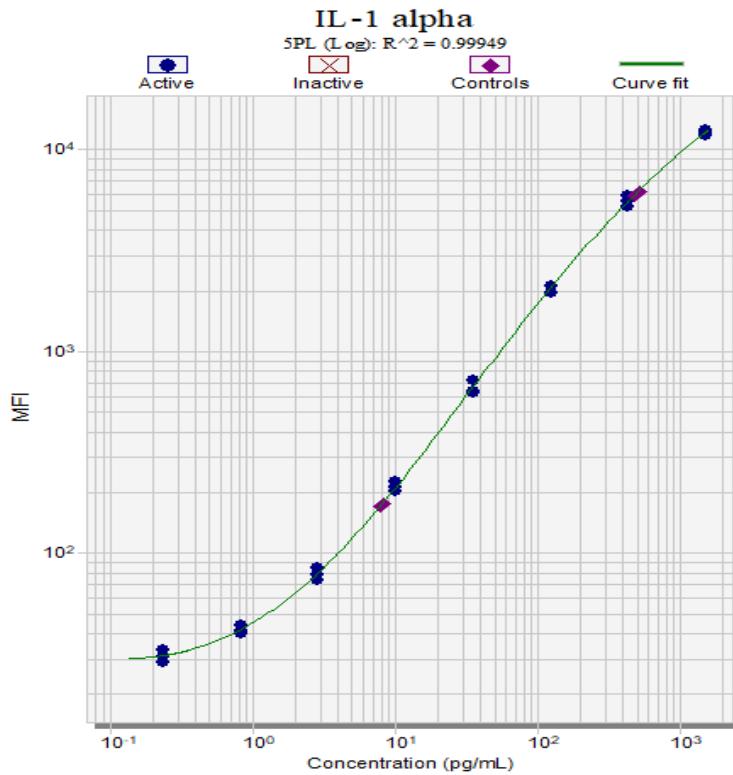
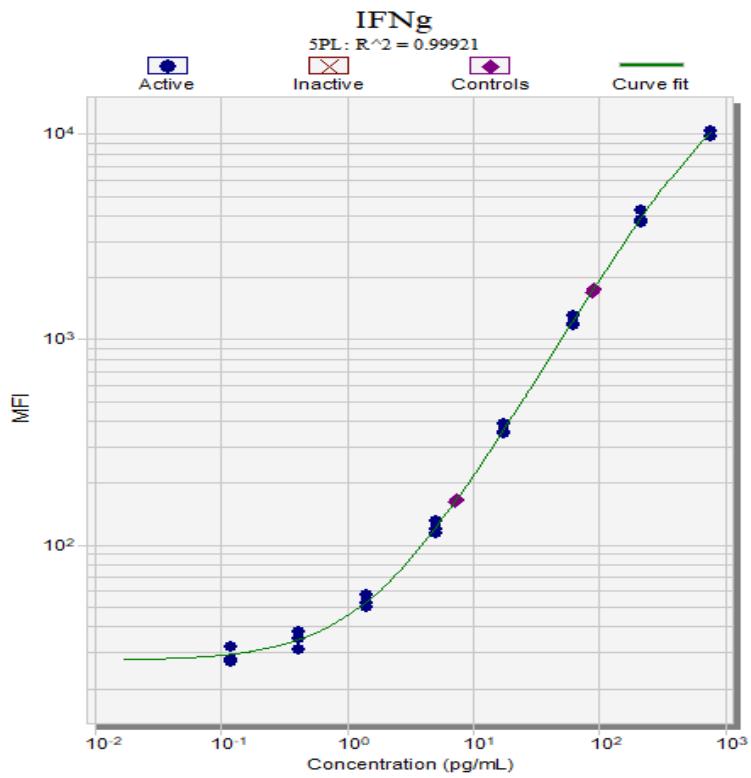
**LDD:** MFI (Median Fluorescent Intensity) for 20 replicates of the standard curve diluent was averaged and two (2) standard deviations added. This value was calculated to concentration off the standard curve.

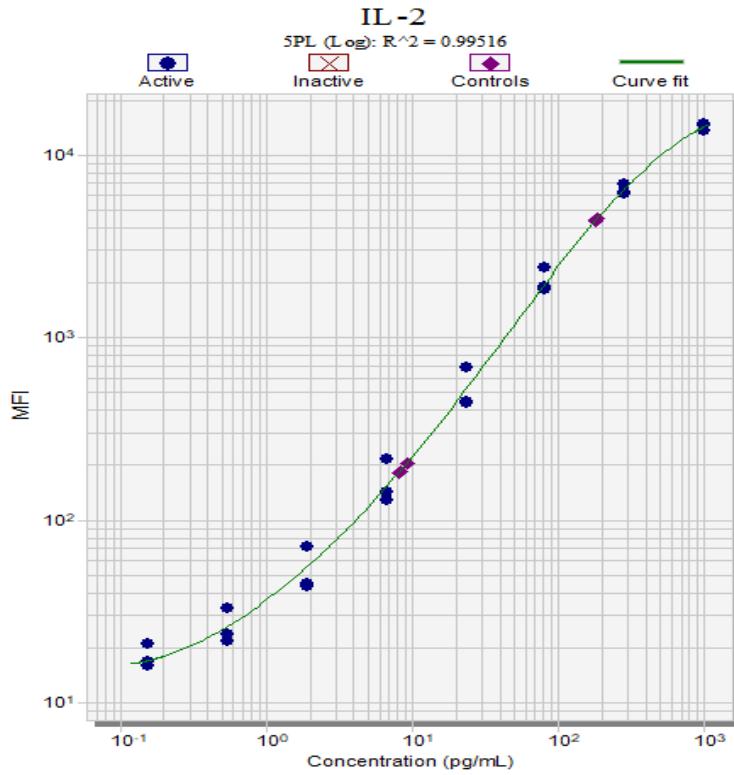
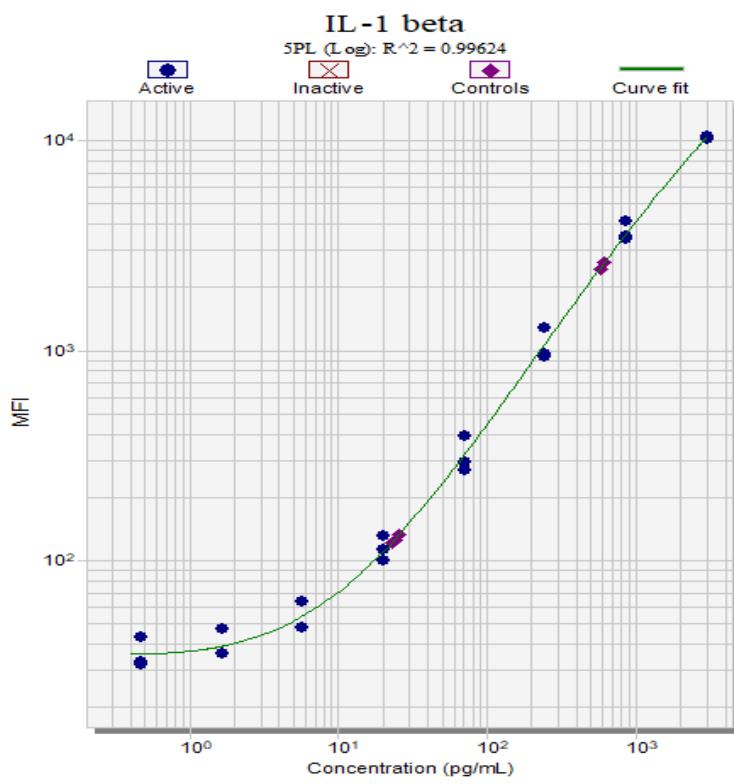
**LLOQ:** LLOQ was assessed by diluting a low serum sample for 8, 2-fold dilutions in triplicate. The LLOQ represents the value at which 30% CV was attained, with linearity with 70-130%. If that value calculates lower than the LOD, then the LLOQ value is equal to the LOD.

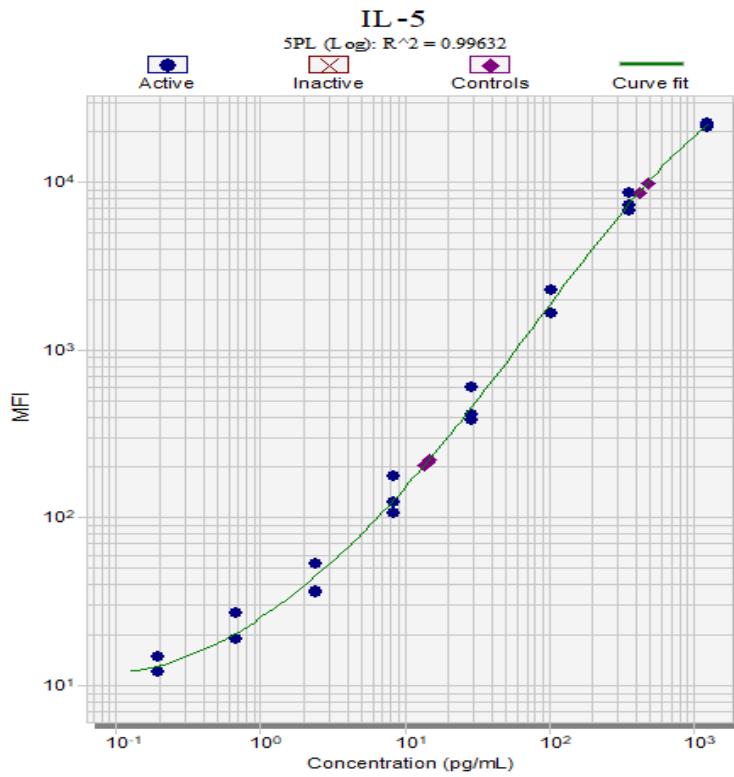
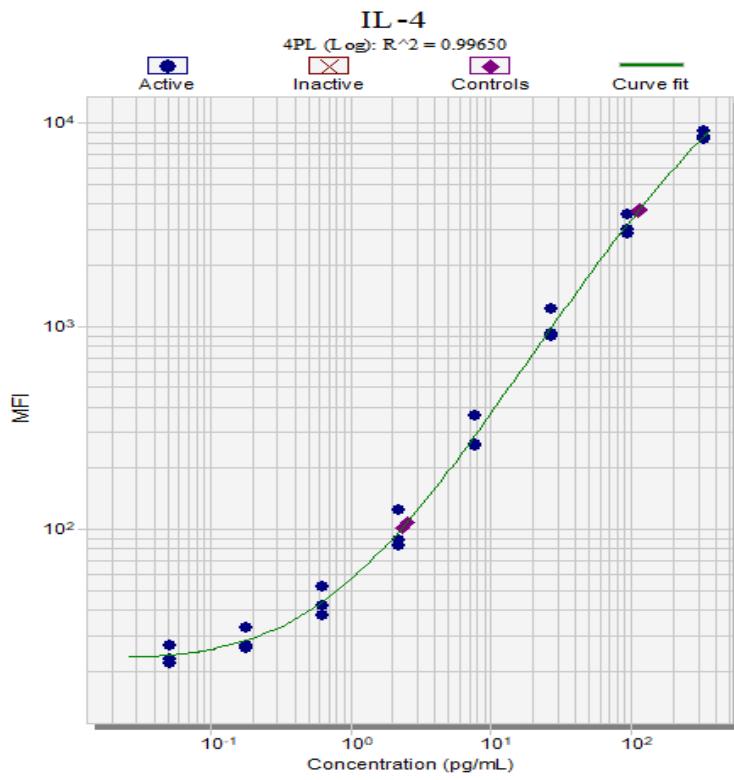
		<b>S1</b>	<b>S8</b>	<b>LDD</b>	<b>LLOQ</b>
<b>IFN<math>\gamma</math></b>	<b>pg/mL</b>	0.12	750	0.46	0.46
<b>IL-1<math>\alpha</math></b>	<b>pg/mL</b>	0.23	1500	0.65	1.2
<b>IL-1<math>\beta</math></b>	<b>pg/mL</b>	0.47	3000	1.5	1.5
<b>IL-10</b>	<b>pg/mL</b>	0.19	1250	0.30	0.30
<b>IL-12p40</b>	<b>pg/mL</b>	0.78	5000	8.0	8.0
<b>IL-12p70</b>	<b>pg/mL</b>	2.33	15000	8.2	8.2
<b>IL-13</b>	<b>pg/mL</b>	0.16	1000	0.38	1.2
<b>IL-2</b>	<b>pg/mL</b>	0.16	1000	0.58	0.90
<b>IL-4</b>	<b>pg/mL</b>	0.051	330	0.10	0.10
<b>IL-5</b>	<b>pg/mL</b>	0.19	1250	0.20	0.20
<b>IL-6</b>	<b>pg/mL</b>	0.23	1500	0.20	0.20
<b>KC/GRO</b>	<b>pg/mL</b>	0.23	1500	1.2	1.2
<b>TNF<math>\alpha</math></b>	<b>pg/mL</b>	0.078	500	0.10	0.10

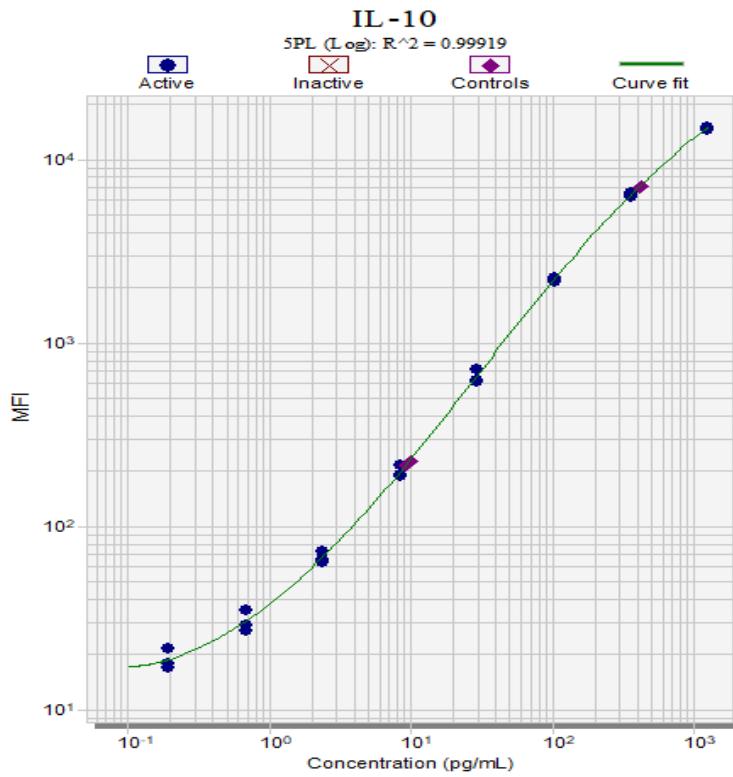
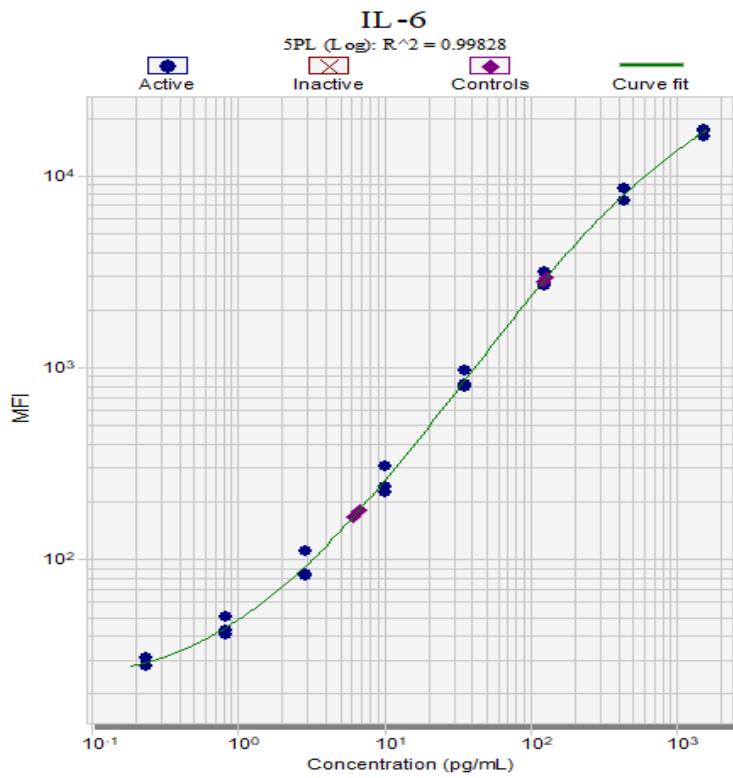
<b>X Dilution Factor</b>		<b>S1</b>	<b>S8</b>	<b>LDD</b>	<b>LLOQ</b>
<b>IFN<math>\gamma</math></b>	<b>pg/mL</b>	0.59	3750	2.3	2.3
<b>IL-1<math>\alpha</math></b>	<b>pg/mL</b>	1.2	7500	3.3	6.0
<b>IL-1<math>\beta</math></b>	<b>pg/mL</b>	2.3	15000	7.5	7.5
<b>IL-10</b>	<b>pg/mL</b>	0.97	6250	1.5	1.5
<b>IL-12p40</b>	<b>pg/mL</b>	3.9	25000	40	40
<b>IL-12p70</b>	<b>pg/mL</b>	12	75000	41	41
<b>IL-13</b>	<b>pg/mL</b>	0.78	5000	1.9	6.0
<b>IL-2</b>	<b>pg/mL</b>	0.78	5000	2.9	4.5
<b>IL-4</b>	<b>pg/mL</b>	0.26	1650	0.50	0.50
<b>IL-5</b>	<b>pg/mL</b>	0.97	6250	1.0	1.0
<b>IL-6</b>	<b>pg/mL</b>	1.2	7500	1.0	1.0
<b>KC/GRO</b>	<b>pg/mL</b>	1.2	7500	6.0	6.0
<b>TNF<math>\alpha</math></b>	<b>pg/mL</b>	0.39	2500	0.50	0.50

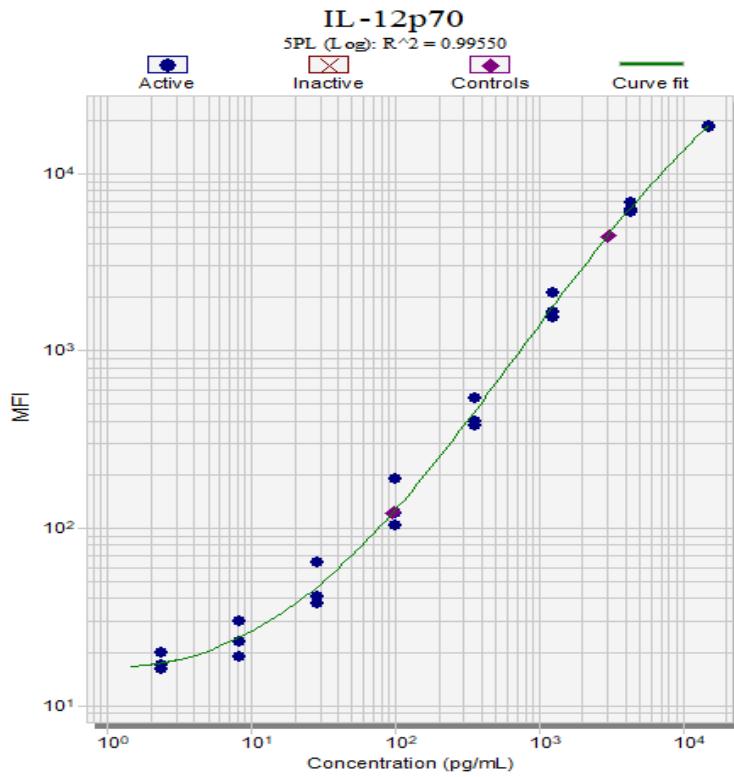
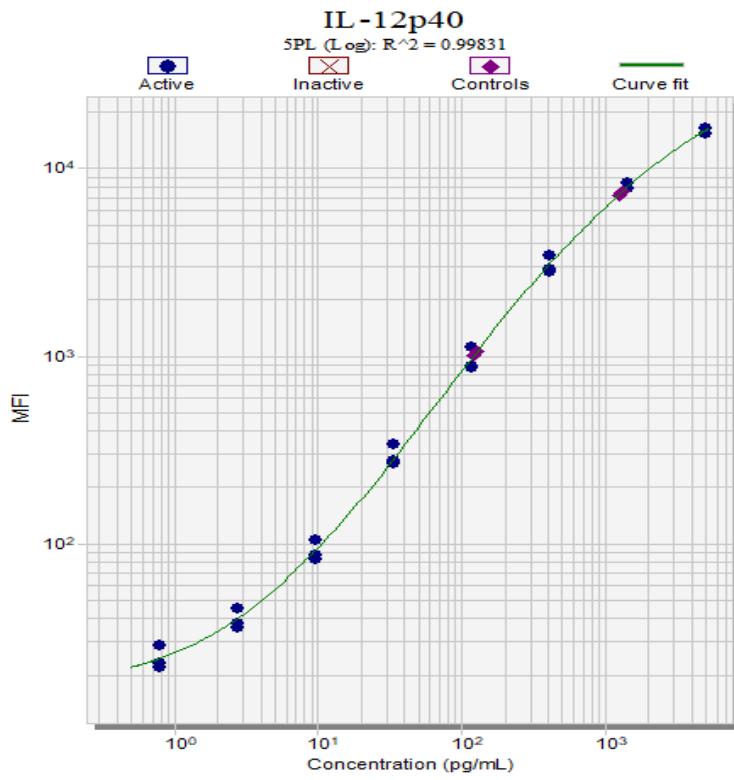
**Curves:** Curves were calculated using the best fit function in Plate Viewer Software. The S1 is the lowest level standard and the S8 is the highest.

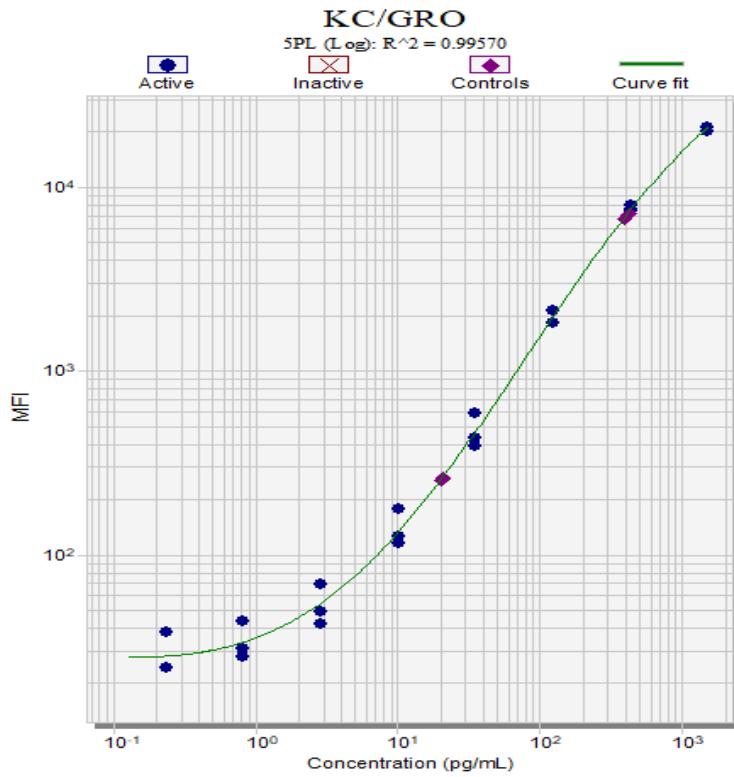
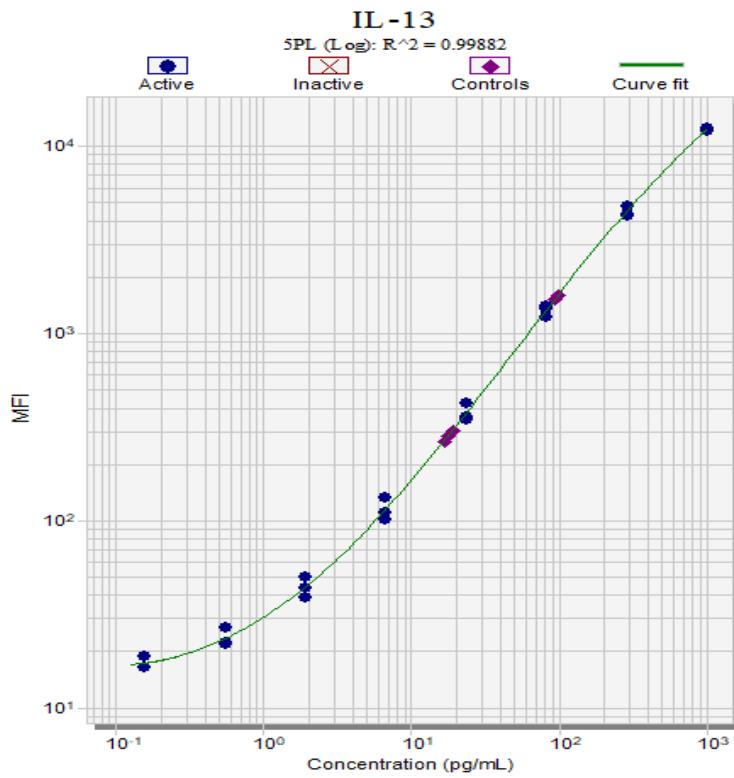


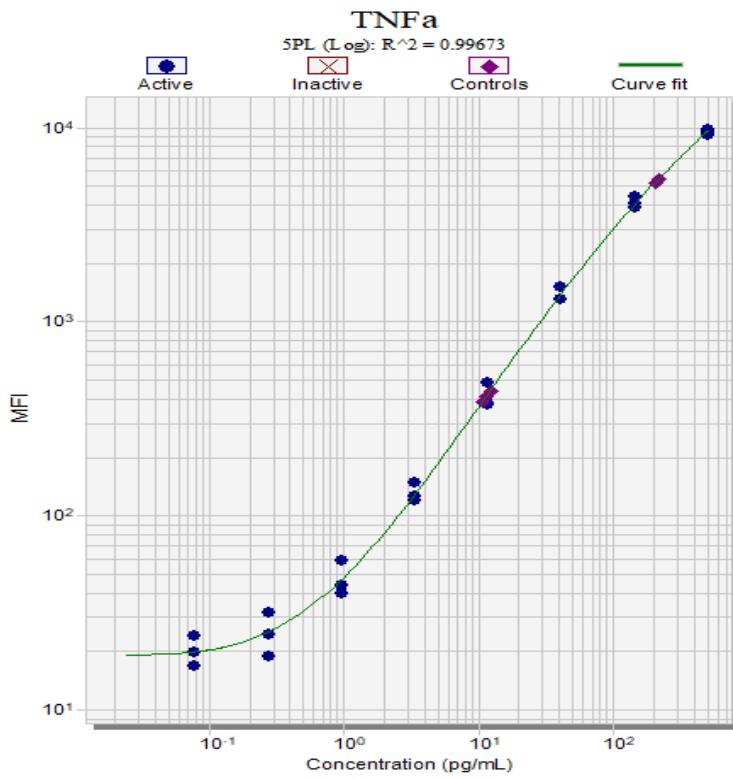












#### 4. Precision:

Control samples were run in triplicate over 3 runs over 2 days with 2 analysts. Precision is the % CV of each run (intra, each run; inter, over 3 runs). Acceptance for precision is <20% CV. All assays meet acceptance for precision.

IFNy		1	2	3	Inter
<b>Control 1</b>	<b>Mean</b>	6.8	7.1	7.2	<b>7.0</b>
	% CV	4%	3%	3%	<b>4%</b>
<b>Control 2</b>	<b>Mean</b>	84	87	82	<b>84</b>
	% CV	3%	3%	4%	<b>3%</b>

IL-1 $\alpha$		1	2	3	Inter
<b>Control 1</b>	<b>Mean</b>	7.7	7.7	8.7	<b>8.0</b>
	% CV	2%	6%	4%	<b>7%</b>
<b>Control 2</b>	<b>Mean</b>	494	484	457	<b>478</b>
	% CV	4%	3%	2%	<b>4%</b>

IL-1 $\beta$		1	2	3	Inter
<b>Control 1</b>	<b>Mean</b>	21	26	24	<b>24</b>
	% CV	3%	3%	1%	<b>8%</b>
<b>Control 2</b>	<b>Mean</b>	556	609	497	<b>554</b>
	% CV	4%	5%	2%	<b>9%</b>

IL-10		1	2	3	Inter
<b>Control 1</b>	<b>Mean</b>	10	9.4	10	<b>10</b>
	% CV	3%	5%	2%	<b>5%</b>
<b>Control 2</b>	<b>Mean</b>	391	426	343	<b>387</b>
	% CV	4%	2%	1%	<b>10%</b>

IL-12p40		1	2	3	Inter
<b>Control 1</b>	<b>Mean</b>	122	133	126	<b>127</b>
	% CV	6%	5%	3%	<b>6%</b>
<b>Control 2</b>	<b>Mean</b>	1117	1143	1187	<b>1149</b>
	% CV	1%	4%	3%	<b>4%</b>

IL-12p70		1	2	3	Inter
<b>Control 1</b>	<b>Mean</b>	85	92	94	<b>90</b>
	% CV	3%	3%	3%	<b>5%</b>
<b>Control 2</b>	<b>Mean</b>	2773	2973	2647	<b>2798</b>
	% CV	1%	3%	2%	<b>5%</b>

IL-13		1	2	3	Inter
<b>Control 1</b>	<b>Mean</b>	17	14	20	<b>17</b>
	% CV	3%	1%	3%	<b>14%</b>
<b>Control 2</b>	<b>Mean</b>	87	91	90	<b>89</b>
	% CV	2%	2%	2%	<b>3%</b>

IL-2		1	2	3	Inter
<b>Control 1</b>	<b>Mean</b>	7.0	7.1	7.6	<b>7.2</b>
	% CV	3%	4%	1%	<b>4%</b>
<b>Control 2</b>	<b>Mean</b>	168	170	175	<b>171</b>
	% CV	3%	3%	4%	<b>4%</b>

IL-4		1	2	3	Inter
<b>Control 1</b>	<b>Mean</b>	2.1	2.2	2.3	<b>2.2</b>
	% CV	5%	3%	6%	<b>5%</b>
<b>Control 2</b>	<b>Mean</b>	103	109	98	<b>103</b>
	% CV	1%	2%	0%	<b>5%</b>

IL-5		1	2	3	Inter
<b>Control 1</b>	<b>Mean</b>	12	13	13	<b>13</b>
	% CV	5%	2%	9%	<b>7%</b>
<b>Control 2</b>	<b>Mean</b>	408	458	341	<b>402</b>
	% CV	1%	4%	2%	<b>13%</b>

IL-6		1	2	3	Inter
<b>Control 1</b>	<b>Mean</b>	5.3	5.8	6.2	<b>5.8</b>
	% CV	3%	1%	3%	<b>7%</b>
<b>Control 2</b>	<b>Mean</b>	114	132	119	<b>122</b>
	% CV	2%	2%	6%	<b>7%</b>

KC/GRO		1	2	3	Inter
<b>Control 1</b>	<b>Mean</b>	19	21	21	<b>20</b>
	% CV	1%	1%	2%	<b>6%</b>
<b>Control 2</b>	<b>Mean</b>	344	396	363	<b>368</b>
	% CV	2%	1%	2%	<b>6%</b>

TNF $\alpha$		1	2	3	Inter
<b>Control 1</b>	<b>Mean</b>	10	11	11	<b>11</b>
	% CV	2%	3%	4%	<b>6%</b>
<b>Control 2</b>	<b>Mean</b>	195	197	175	<b>189</b>
	% CV	3%	1%	1%	<b>6%</b>

##### 5. Linearity:

Linearity was assessed using 2 serum and 2 plasma samples spiked with the standard and diluted 1:2 for 8 dilutions. Percent Recovery was calculated using the calculated value (with kit dilution) as expected (observed x dilution / expected concentration X 100). The acceptance range for linearity is 70-130% recovery for all values above the LLOQ. All assays meet acceptance criteria.

IFNy		Serum 1	Serum 2	Plasma 1	Plasma 2
pg/ml					
1:5		2375	2600	3330	2315
1:10		1130	1270	1270	1315
1:20		557	705	607	849
1:40		270	346	272	329
2		95%	98%	76%	114%
4		99%	111%	96%	129%
8		97%	98%	90%	78%

IL-1 $\alpha$		Serum 1	Serum 2	Plasma 1	Plasma 2
pg/ml					
1:5		511	458	545	538
1:10		258	257	271	305
1:20		131	140	138	171
1:40		67	77	70	75
2		101%	112%	99%	113%
4		102%	109%	102%	112%
8		103%	110%	101%	88%

IL-1β		Serum 1	Serum 2	Plasma 1	Plasma 2
pg/ml					
1:5		4520	4115	3835	4000
1:10		2075	1940	1725	2170
1:20		965	1088	847	1325
1:40		425	501	393	494
2		92%	94%	90%	109%
4		93%	112%	98%	122%
8		88%	92%	93%	75%

IL-10		Serum 1	Serum 2	Plasma 1	Plasma 2
pg/ml					
1:5		37	35	41	34
1:10		20	17	18	20
1:20		10	10	10	12
1:40		5.5	5.3	5.0	5.4
2		106%	100%	89%	116%
4		104%	120%	106%	118%
8		108%	102%	104%	94%

IL-12p40		Serum 1	Serum 2	Plasma 1	Plasma 2
pg/ml					
1:5		332	272	443	322
1:10		188	149	204	179
1:20		106	87	105	98
1:40		54	45	53	42
2		113%	109%	92%	111%
4		112%	117%	103%	110%
8		103%	102%	101%	86%

IL-12p70		Serum 1	Serum 2	Plasma 1	Plasma 2
pg/ml					
1:5		2780	2340	2495	2365
1:10		1105	991	923	1029
1:20		487	479	380	520
1:40		202	224	172	210
2		79%	85%	74%	87%
4		88%	97%	82%	101%
8		83%	93%	91%	81%

IL-13		Serum 1	Serum 2	Plasma 1	Plasma 2
pg/ml					
1:5		147	154	261	197
1:10		87	98	130	114
1:20		54	58	69	74
1:40		31	35	37	37
2		119%	128%	100%	116%
4		124%	119%	106%	130%
8		115%	118%	108%	101%

IL-2		Serum 1	Serum 2	Plasma 1	Plasma 2
pg/ml					
1:5		65	57	55	49
1:10		24	23	22	22
1:20		11	11	9.2	12
1:40		5.0	5.5	4.1	4.9
2		76%	80%	81%	88%
4		88%	96%	83%	108%
8		93%	102%	90%	84%

IL-4		Serum 1	Serum 2	Plasma 1	Plasma 2
pg/ml					
1:5		32	29	32	32
1:10		16	15	16	17
1:20		8.8	8.4	7.8	10
1:40		4.0	4.3	3.9	4.1
2		99%	102%	100%	110%
4		112%	114%	97%	114%
8		91%	104%	101%	84%

IL-5		Serum 1	Serum 2	Plasma 1	Plasma 2
pg/ml					
1:5		617	511	547	340
1:10		245	201	198	219
1:20		91	81	73	127
1:40		38	33	30	65
2		79%	79%	72%	129%
4		74%	81%	74%	116%
8		84%	82%	82%	102%

<b>IL-6</b>				
<b>pg/ml</b>	<b>Serum 1</b>	<b>Serum 2</b>	<b>Plasma 1</b>	<b>Plasma 2</b>
<b>1:5</b>	41	30	36	30
<b>1:10</b>	17	13	13	13
<b>1:20</b>	7.4	6.0	6.1	7.6
<b>1:40</b>	3.1	2.9	2.6	2.8
<b>2</b>	81%	85%	73%	89%
<b>4</b>	88%	94%	92%	115%
<b>8</b>	85%	98%	84%	73%

<b>KC/GRO</b>				
<b>pg/ml</b>	<b>Serum 1</b>	<b>Serum 2</b>	<b>Plasma 1</b>	<b>Plasma 2</b>
<b>1:5</b>	38	1075	238	741
<b>1:10</b>	18	688	154	473
<b>1:20</b>	8.0	430	90	256
<b>1:40</b>	4.6	250	46	140
<b>2</b>	92%	128%	130%	128%
<b>4</b>	91%	125%	116%	108%
<b>8</b>	114%	116%	103%	109%

<b>TNF<math>\alpha</math></b>				
<b>pg/ml</b>	<b>Serum 1</b>	<b>Serum 2</b>	<b>Plasma 1</b>	<b>Plasma 2</b>
<b>1:5</b>	76	50	58	48
<b>1:10</b>	36	23	27	27
<b>1:20</b>	18	14	14	16
<b>1:40</b>	8.9	7.3	6.9	8.1
<b>2</b>	95%	94%	93%	112%
<b>4</b>	97%	117%	104%	120%
<b>8</b>	101%	107%	100%	101%

**Freeze/thaw stability:** Samples were assessed for freeze-thaw stability after 1, 2, and 3 F/T cycles. All values were within the acceptance range of 80-120% for freeze-thaw samples compared to the non-freeze thawed samples indicating that samples could be freeze-thawed up to 3 times without a loss in signal.

	IFN $\gamma$				
	pg/ml	Serum 1	Serum 2	Plasma 1	Plasma 2
Value	FT-0X	852	776	880	674
	FT-1X	908	750	891	704
	FT-2X	953	819	897	746
	FT-3X	731	712	824	672
% Control	FT-0X	100%	100%	100%	100%
	FT-1X	107%	97%	101%	105%
	FT-2X	112%	106%	102%	111%
	FT-3X	86%	92%	94%	100%

	IL-1 $\alpha$				
	pg/ml	Serum 1	Serum 2	Plasma 1	Plasma 2
Value	FT-0X	966	755	906	887
	FT-1X	945	710	890	888
	FT-2X	989	727	904	945
	FT-3X	808	635	794	813
% Control	FT-0X	100%	100%	100%	100%
	FT-1X	98%	94%	98%	100%
	FT-2X	102%	96%	100%	107%
	FT-3X	84%	84%	88%	92%

	IL-1 $\beta$				
	pg/ml	Serum 1	Serum 2	Plasma 1	Plasma 2
Value	FT-0X	8725	7340	6965	7390
	FT-1X	9900	7125	7090	7695
	FT-2X	9985	7490	7185	7810
	FT-3X	7740	6810	6670	7200
% Control	FT-0X	100%	100%	100%	100%
	FT-1X	113%	97%	102%	104%
	FT-2X	114%	102%	103%	106%
	FT-3X	89%	93%	96%	97%

	<b>IL-10</b>				
	<b>pg/ml</b>	<b>Serum 1</b>	<b>Serum 2</b>	<b>Plasma 1</b>	<b>Plasma 2</b>
Value	FT-0X	1685	1445	2080	1435
	FT-1X	1760	1425	2050	1440
	FT-2X	1780	1460	2120	1495
	FT-3X	1515	1285	1970	1400
% Control	FT-0X	100%	100%	100%	100%
	FT-1X	104%	99%	99%	100%
	FT-2X	106%	101%	102%	104%
	FT-3X	90%	89%	95%	98%

	<b>IL-12p40</b>				
	<b>pg/ml</b>	<b>Serum 1</b>	<b>Serum 2</b>	<b>Plasma 1</b>	<b>Plasma 2</b>
Value	FT-0X	1430	1210	2095	1625
	FT-1X	1445	1175	2120	1760
	FT-2X	1440	1180	2260	1745
	FT-3X	1360	1085	2120	1640
% Control	FT-0X	100%	100%	100%	100%
	FT-1X	101%	97%	101%	108%
	FT-2X	101%	98%	108%	107%
	FT-3X	95%	90%	101%	101%

	<b>IL-12p70</b>				
	<b>pg/ml</b>	<b>Serum 1</b>	<b>Serum 2</b>	<b>Plasma 1</b>	<b>Plasma 2</b>
Value	FT-0X	9665	8300	9440	8195
	FT-1X	10300	8165	9330	8490
	FT-2X	10885	8730	9330	8920
	FT-3X	8925	7960	9200	8180
% Control	FT-0X	100%	100%	100%	100%
	FT-1X	107%	98%	99%	104%
	FT-2X	113%	105%	99%	109%
	FT-3X	92%	96%	97%	100%

	<b>IL-13</b>				
	<b>pg/ml</b>	<b>Serum 1</b>	<b>Serum 2</b>	<b>Plasma 1</b>	<b>Plasma 2</b>
Value	FT-0X	121	34	181	26
	FT-1X	118	34	191	25
	FT-2X	121	43	192	27
	FT-3X	123	35	188	29
% Control	FT-0X	100%	100%	100%	100%
	FT-1X	98%	100%	105%	98%
	FT-2X	100%	127%	106%	105%
	FT-3X	101%	103%	104%	111%

	<b>IL-2</b>				
	<b>pg/ml</b>	<b>Serum 1</b>	<b>Serum 2</b>	<b>Plasma 1</b>	<b>Plasma 2</b>
Value	FT-0X	965	768	938	768
	FT-1X	1028	758	940	747
	FT-2X	1067	796	958	791
	FT-3X	825	711	915	703
% Control	FT-0X	100%	100%	100%	100%
	FT-1X	106%	99%	100%	97%
	FT-2X	111%	104%	102%	103%
	FT-3X	85%	93%	98%	92%

	<b>IL-4</b>				
	<b>pg/ml</b>	<b>Serum 1</b>	<b>Serum 2</b>	<b>Plasma 1</b>	<b>Plasma 2</b>
Value	FT-0X	665	550	679	655
	FT-1X	679	524	710	669
	FT-2X	709	554	706	690
	FT-3X	576	504	677	634
% Control	FT-0X	100%	100%	100%	100%
	FT-1X	102%	95%	105%	102%
	FT-2X	107%	101%	104%	105%
	FT-3X	87%	92%	100%	97%

	<b>IL-5</b>				
	<b>pg/ml</b>	<b>Serum 1</b>	<b>Serum 2</b>	<b>Plasma 1</b>	<b>Plasma 2</b>
Value	FT-0X	1380	1035	1230	1190
	FT-1X	1465	1070	1240	1195
	FT-2X	1435	1120	1315	1260
	FT-3X	1265	970	1220	1125
% Control	FT-0X	100%	100%	100%	100%
	FT-1X	106%	103%	101%	100%
	FT-2X	104%	108%	107%	106%
	FT-3X	92%	94%	99%	95%

	<b>IL-6</b>				
	<b>pg/ml</b>	<b>Serum 1</b>	<b>Serum 2</b>	<b>Plasma 1</b>	<b>Plasma 2</b>
Value	FT-0X	2130	1610	2280	1940
	FT-1X	2255	1570	2360	1900
	FT-2X	2280	1695	2425	2020
	FT-3X	1725	1395	2275	1805
% Control	FT-0X	100%	100%	100%	100%
	FT-1X	106%	98%	104%	98%
	FT-2X	107%	105%	106%	104%
	FT-3X	81%	87%	100%	93%

	KC/GRO				
	pg/ml	Serum 1	Serum 2	Plasma 1	Plasma 2
Value	FT-0X	1390	1415	2030	1710
	FT-1X	1455	1385	2020	1650
	FT-2X	1585	1520	2105	1715
	FT-3X	1195	1300	1950	1575
% Control	FT-0X	100%	100%	100%	100%
	FT-1X	105%	98%	100%	96%
	FT-2X	114%	107%	104%	100%
	FT-3X	86%	92%	96%	92%

	TNF $\alpha$				
	pg/ml	Serum 1	Serum 2	Plasma 1	Plasma 2
Value	FT-0X	1115	836	929	748
	FT-1X	1099	813	979	763
	FT-2X	1129	816	974	755
	FT-3X	965	756	951	715
% Control	FT-0X	100%	100%	100%	100%
	FT-1X	99%	97%	105%	102%
	FT-2X	101%	98%	105%	101%
	FT-3X	87%	90%	102%	96%

**6. Bench Top Stability:** Samples were assessed bench top stability and 2hr RT, and 2 & 4Hr 4°C to determine if the samples were stable on the bench prior to the assay or if refrigeration was required. All values were within the acceptance range of 80-120% for samples compared to the bench top samples indicating that no loss in activity will occur during the testing of the samples, with the exception of IL-13 at 2 hr RT. Samples are kept on ice during processing to avoid any loss of signal.

	IFN $\gamma$				
	pg/ml	Serum 1	Serum 2	Plasma 1	Plasma 2
Value	0 HR	5068	4373	4920	747
	2hr RT	5280	4561	5205	701
	2hr 4C	5439	4677	5169	880
	4hr 4C	4764	4682	4951	735
% Control	0 HR	100%	100%	100%	100%
	2hr RT	104%	104%	106%	94%
	2hr 4C	107%	107%	105%	118%
	4hr 4C	94%	107%	101%	98%

	<b>IL-1α</b>				
	<b>pg/ml</b>	<b>Serum 1</b>	<b>Serum 2</b>	<b>Plasma 1</b>	<b>Plasma 2</b>
Value	0 HR	883	733	932	1019
	2hr RT	775	561	664	756
	2hr 4C	881	741	871	1007
	4hr 4C	769	670	741	953
% Control	0 HR	100%	100%	100%	100%
	2hr RT	88%	76%	71%	74%
	2hr 4C	100%	101%	93%	99%
	4hr 4C	87%	91%	79%	94%

	<b>IL-1β</b>				
	<b>pg/ml</b>	<b>Serum 1</b>	<b>Serum 2</b>	<b>Plasma 1</b>	<b>Plasma 2</b>
Value	0 HR	8165	7510	7360	8230
	2hr RT	7970	7225	7720	7520
	2hr 4C	8530	8620	7455	9095
	4hr 4C	7880	8640	7090	7545
% Control	0 HR	100%	100%	100%	100%
	2hr RT	98%	96%	105%	91%
	2hr 4C	104%	115%	101%	111%
	4hr 4C	97%	115%	96%	92%

	<b>IL-10</b>				
	<b>pg/ml</b>	<b>Serum 1</b>	<b>Serum 2</b>	<b>Plasma 1</b>	<b>Plasma 2</b>
Value	0 HR	1535	1430	2115	1505
	2hr RT	1450	1345	1920	1380
	2hr 4C	1580	1550	2050	1565
	4hr 4C	1475	1450	1965	1425
% Control	0 HR	100%	100%	100%	100%
	2hr RT	94%	94%	91%	92%
	2hr 4C	103%	108%	97%	104%
	4hr 4C	96%	101%	93%	95%

	<b>IL-12p40</b>				
	<b>pg/ml</b>	<b>Serum 1</b>	<b>Serum 2</b>	<b>Plasma 1</b>	<b>Plasma 2</b>
Value	0 HR	1350	1265	2335	1820
	2hr RT	1340	1190	2275	1730
	2hr 4C	1425	1165	2210	1800
	4hr 4C	1310	1155	2250	1730
% Control	0 HR	100%	100%	100%	100%
	2hr RT	99%	94%	97%	95%
	2hr 4C	106%	92%	95%	99%
	4hr 4C	97%	91%	96%	95%

	<b>IL-12p70</b>				
	<b>pg/ml</b>	<b>Serum 1</b>	<b>Serum 2</b>	<b>Plasma 1</b>	<b>Plasma 2</b>
Value	0 HR	9380	8865	10050	9055
	2hr RT	9470	8445	10025	8705
	2hr 4C	10550	9920	9530	10165
	4hr 4C	8455	9805	9045	8760
% Control	0 HR	100%	100%	100%	100%
	2hr RT	101%	95%	100%	96%
	2hr 4C	112%	112%	95%	112%
	4hr 4C	90%	111%	90%	97%

	<b>IL-13</b>				
	<b>pg/ml</b>	<b>Serum 1</b>	<b>Serum 2</b>	<b>Plasma 1</b>	<b>Plasma 2</b>
Value	0 HR	134	33	194	26
	2hr RT	94	32	121	26
	2hr 4C	102	31	141	28
	4hr 4C	105	32	146	28
% Control	0 HR	100%	100%	100%	100%
	2hr RT	70%	95%	63%	97%
	2hr 4C	76%	92%	73%	105%
	4hr 4C	78%	95%	75%	108%

	<b>IL-2</b>				
	<b>pg/ml</b>	<b>Serum 1</b>	<b>Serum 2</b>	<b>Plasma 1</b>	<b>Plasma 2</b>
Value	0 HR	877	811	959	822
	2hr RT	918	812	984	723
	2hr 4C	953	981	980	905
	4hr 4C	824	962	929	728
% Control	0 HR	100%	100%	100%	100%
	2hr RT	105%	100%	103%	88%
	2hr 4C	109%	121%	102%	110%
	4hr 4C	94%	119%	97%	89%

	<b>IL-4</b>				
	<b>pg/ml</b>	<b>Serum 1</b>	<b>Serum 2</b>	<b>Plasma 1</b>	<b>Plasma 2</b>
Value	0 HR	595	553	692	713
	2hr RT	613	561	699	665
	2hr 4C	637	627	716	777
	4hr 4C	585	594	668	689
% Control	0 HR	100%	100%	100%	100%
	2hr RT	103%	102%	101%	93%
	2hr 4C	107%	113%	103%	109%
	4hr 4C	98%	107%	97%	97%
	<b>IL-5</b>				

	<b>pg/ml</b>	<b>Serum 1</b>	<b>Serum 2</b>	<b>Plasma 1</b>	<b>Plasma 2</b>
<b>Value</b>	0 HR	1320	1160	1325	1285
	2hr RT	1260	1095	1275	1210
	2hr 4C	1425	1225	1290	1380
	4hr 4C	1255	1190	1225	1200
<b>% Control</b>	0 HR	100%	100%	100%	100%
	2hr RT	95%	94%	96%	94%
	2hr 4C	108%	106%	97%	107%
	4hr 4C	95%	103%	92%	93%

	<b>IL-6</b>				
	<b>pg/ml</b>	<b>Serum 1</b>	<b>Serum 2</b>	<b>Plasma 1</b>	<b>Plasma 2</b>
<b>Value</b>	0 HR	1820	1640	2320	2070
	2hr RT	1855	1695	2225	1920
	2hr 4C	1995	1860	2415	2390
	4hr 4C	1695	1895	2345	1895
<b>% Control</b>	0 HR	100%	100%	100%	100%
	2hr RT	102%	103%	96%	93%
	2hr 4C	110%	113%	104%	115%
	4hr 4C	93%	116%	101%	92%

	<b>KC/GRO</b>				
	<b>pg/ml</b>	<b>Serum 1</b>	<b>Serum 2</b>	<b>Plasma 1</b>	<b>Plasma 2</b>
<b>Value</b>	0 HR	1290	1440	2050	1785
	2hr RT	1215	1355	1940	1650
	2hr 4C	1325	1730	2050	2005
	4hr 4C	1190	1750	1935	1625
<b>% Control</b>	0 HR	100%	100%	100%	100%
	2hr RT	94%	94%	95%	92%
	2hr 4C	103%	120%	100%	112%
	4hr 4C	92%	122%	94%	91%

	<b>TNF<math>\alpha</math></b>				
	<b>pg/ml</b>	<b>Serum 1</b>	<b>Serum 2</b>	<b>Plasma 1</b>	<b>Plasma 2</b>
<b>Value</b>	0 HR	984	807	944	815
	2hr RT	1035	854	1020	740
	2hr 4C	1085	883	1010	811
	4hr 4C	904	884	952	778
<b>% Control</b>	0 HR	100%	100%	100%	100%
	2hr RT	105%	106%	108%	91%
	2hr 4C	110%	109%	107%	100%
	4hr 4C	92%	109%	101%	95%