

V-PLEX® Chemokine Panel 1: Original vs. Gen B



White Paper

Introduction

MSD continues to improve its products in response to customer feedback. As part of this commitment, the V-PLEX Chemokine Panel 1 has been updated to enhance performance and strengthen long-term supply. In this whitepaper, we discuss the key performance metrics of the V-PLEX Chemokine Panel 1 Gen B Kit and the original V-PLEX Chemokine Panel 1 Kit for both the human and NHP assays.

Table 1: Kits compared in this white paper.

Description	Part Number Original	Part Number Gen B
V-PLEX Chemokine Panel 1 (human) Kit	K15047D	K15705D
V-PLEX Chemokine Panel 1 (NHP) Kit	K15055D	K15712D

Methods

The V-PLEX Chemokine Panel 1 Gen B Kit uses updated antibodies for Eotaxin and MIP-1 α , as well as a new assay diluent to reduce matrix effects and a new read buffer for a shorter protocol. These changes also lead to improved sensitivity and increased longevity of critical reagents. Additionally, the Eotaxin assay is now available in the Gen B NHP kit.

The V-PLEX Chemokine Panel 1 Gen B Kit does not include the low sensitivity IL-8 (HA) assay. If IL-8 is of interest, the V-PLEX Human IL-8 Kit (K151RAD) and the V-PLEX NHP IL-8 Kit (K156RAD) are available for purchase.

Table 2: Differences between the original and Gen B versions of the V-PLEX Chemokine Panel 1 Kits.

Description	Panel 1	Panel 1 Gen B
Assay generation – Eotaxin	B	C
Assay generation – MIP-1 α	B	C
Assay – IL-8 (HA)	Yes	No
NHP cross-reactivity – Eotaxin	No	Yes
Assay diluent	Diluent 43	Diluent 57
Read buffer	Read Buffer T (2X)	MSD GOLD™ Read Buffer B
Read buffer incubation	Yes (10 min)	No

Calibration Curve Comparison

The calibrators in the V-PLEX Chemokine Panel 1 Gen B Kit are anchored to the calibrators in the V-PLEX Chemokine Panel 1 Kit to ensure consistent quantitation between kits. As shown in Figure 1, all calibration curves from the V-PLEX Chemokine Panel 1 (human) Gen B Kit (red) have equivalent or higher sensitivity and range relative to the calibration curves from the V-PLEX Chemokine Panel 1 Kit (blue).

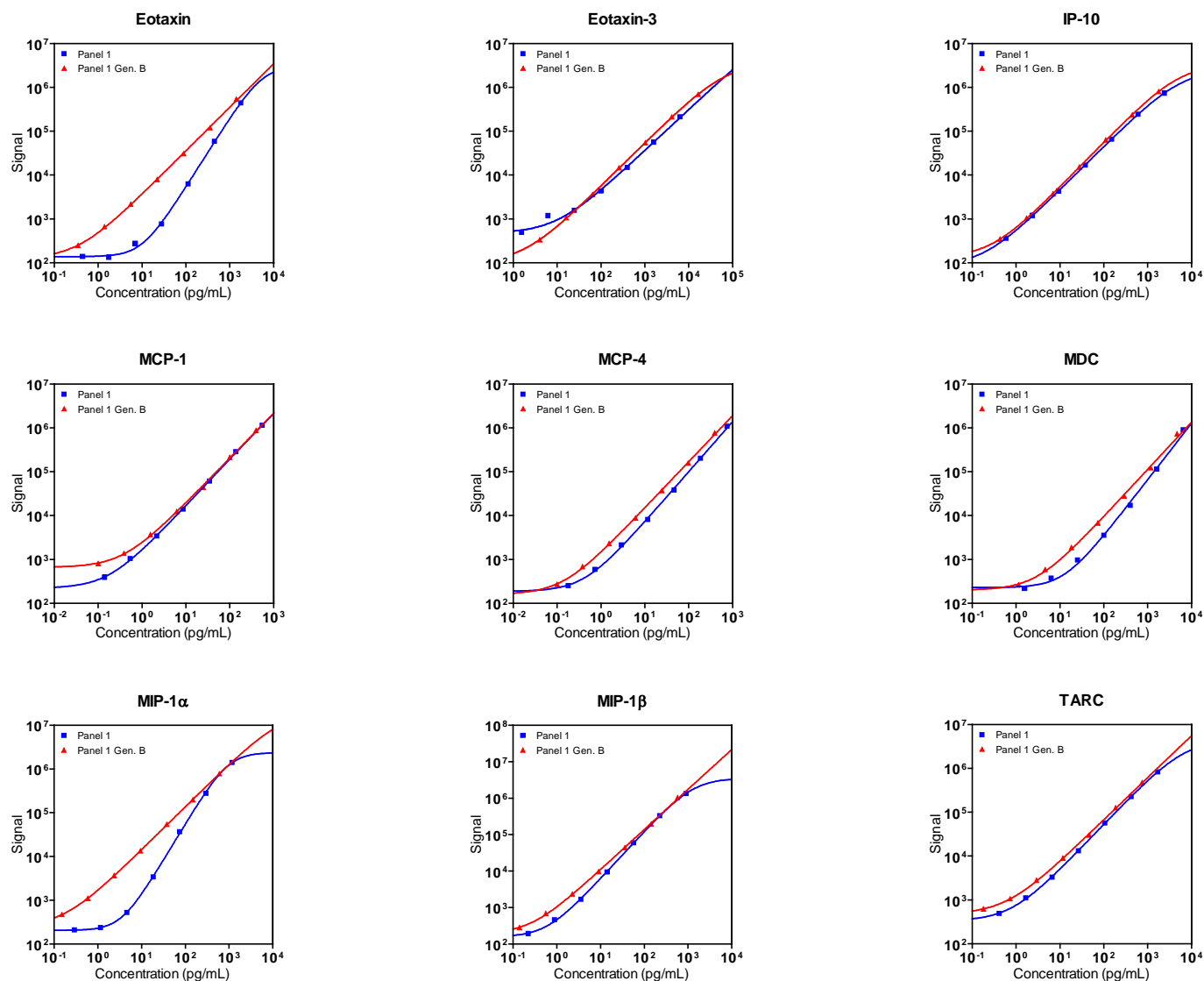


Figure 1: Comparison of calibration curves for the original and Gen B versions of the V-PLEX Chemokine Panel 1 Kits.

Sensitivity Comparison

For comparison of sensitivity, the LLOD is a calculated concentration corresponding to 2.5 standard deviations above the background (zero calibrator). The ULOQ is the highest concentration at which the CV of the calculated concentration is $\leq 20\%$ and the recovery of each analyte is within 80-120% (75-125% for TARC and MDC) of the nominal value. The LLOQ is the lowest concentration at which the CV of the calculated concentration is $\leq 20\%$ and the recovery of each analyte is within 80-120% of the nominal value.

As shown in Table 3, all assays in the Gen B Kit have equivalent or improved sensitivity. Antibody changes for the Eotaxin and MIP-1 α assays significantly improved Hill slope, LLOD, and LLOQ. Additionally, all assays benefitted from reagent and protocol changes in the Gen B kit. Top-of-curve and ULOQ concentrations were adjusted to maximize dynamic range when using Methodical Mind™ reader software.

Table 3: Comparison of sensitivity for the original and Gen B versions of the V-PLEX Chemokine Panel 1 Kits.

Analyte	Hill slope		Median LLOD (pg/mL)		LLOQ (pg/mL)		ULOQ (pg/mL)	
	Panel 1	Panel 1 Gen B	Panel 1	Panel 1 Gen B	Panel 1	Panel 1 Gen B	Panel 1	Panel 1 Gen B
Eotaxin	1.59	0.995	3.26	0.20	12.3	2.14	1,120	910
Eotaxin-3	0.937	0.987	1.77	1.44	10.2	11.1	3,750	11,100
IP-10	0.991	1.02	0.37	0.12	1.37	1.29	500	1,240
MCP-1	1.06	1.03	0.09	0.16	1.09	1.45	375	247
MCP-4	1.14	1.05	0.18	0.05	1.49	0.518	472	234
MDC	1.29	1.08	1.22	1.00	88.3	6.43	3,700	2,930
MIP-1 α	1.67	0.987	3.02	0.05	13.8	0.357	743	403
MIP-1 β	1.30	1.10	0.17	0.09	1.88	1.20	520	390
TARC	1.05	0.965	0.22	0.11	3.32	2.14	1,120	481

Sample Testing Comparison

We tested 30 human samples (commercially sourced serum, EDTA plasma, heparin plasma, citrate plasma, urine, and CSF) and 30 NHP samples (commercially sourced serum, EDTA plasma, and urine from rhesus and cynomolgus macaque species) at 4-fold dilution side-by-side with the Gen B and original V-PLEX Chemokine Panel 1 Kits. Results are presented below for human (Table 4) and NHP (Table 5) samples.

Table 4: Normal human samples tested in the V-PLEX Chemokine Panel 1 Gen B Kit.

Sample Type	Statistic	Eotaxin	MIP-1 β	Eotaxin-3	TARC	IP-10	MIP-1 α	MCP-1	MDC	MCP-4
Serum (N=5)	Median (pg/mL)	112	55.9	17.8	466	129	6.09	165	603	151
	Range (pg/mL)	29.7–229	32.9–489	12.9–33.1	182–1230	65–455	3.58–28.1	121–313	548–1390	63.4–191
	% Detected	100%	100%	80%	100%	100%	100%	100%	100%	100%
	% Quantified	100%	100%	0%	100%	100%	100%	100%	100%	100%
EDTA Plasma (N=5)	Median (pg/mL)	108	34.3	22.7	218	203	4.27	65.6	660	71.9
	Range (pg/mL)	56.6–140	25.5–477	8.09–28.4	72.3–394	128–853	2.71–28.3	55.4–75.1	636–1460	30.7–86.7
	% Detected	100%	100%	100%	100%	100%	100%	100%	100%	100%
	% Quantified	100%	100%	0%	100%	100%	100%	100%	100%	100%
Heparin Plasma (N=5)	Median (pg/mL)	641	31.4	237	141	270	4.10	117	541	295
	Range (pg/mL)	124–1120	25.0–36.2	145–356	68.8–162	149–682	3.13–5.86	94.6–169	365–811	57.9–362
	% Detected	80%	80%	60%	80%	80%	80%	80%	80%	80%
	% Quantified	80%	80%	60%	80%	80%	80%	80%	80%	80%
Citrate Plasma (N=5)	Median (pg/mL)	105	27.6	16.2	93.1	183	3.59	75.8	549	37.2
	Range (pg/mL)	65.5–146	18.9–461	14.4–23.4	31.1–137	108–765	2.49–28.1	64.6–92.4	531–1130	26.4–66.6
	% Detected	100%	100%	80%	100%	100%	100%	100%	100%	100%
	% Quantified	100%	100%	0%	100%	100%	100%	100%	100%	100%
Urine (N=5)	Median (pg/mL)	5.71	3.23	ND	0.735	23.0	0.894	210	5.80	5.62
	Range (pg/mL)	5.00–14.1	0.498–53.8	NA	NA	1.77–38.8	0.326–1.46	24.3–624	4.23–6.34	0.622–35.0
	% Detected	80%	100%	0%	20%	80%	40%	100%	60%	100%
	% Quantified	20%	20%	0%	0%	60%	20%	100%	0%	80%
CSF (N=5)	Median (pg/mL)	1.22	9.18	ND	1.64	146	1.48	231	26.3	0.239
	Range (pg/mL)	1.00–8.69	4.92–27.8	NA	0.599–7.13	67.0–299	0.845–7.96	185–1320	NA	0.167–1.27
	% Detected	60%	100%	0%	100%	100%	100%	100%	20%	100%
	% Quantified	20%	60%	0%	20%	100%	60%	80%	20%	0%

Table 5: Normal NHP samples tested in the V-PLEX Chemokine Panel 1 Gen. B Kit.

Sample Type	Statistic	Eotaxin	MIP-1 β	Eotaxin-3	TARC	IP-10	MIP-1 α	MCP-1	MDC	MCP-4
Serum (N=10)	Median (pg/mL)	232	139	ND	1.21	895	14.8	292	242	9.53
	Range (pg/mL)	52.0–376	93.1–203	NA	0.564–4.23	335–4130	9.41–24.9	181–493	94.9–431	2.69–36.0
	% Detected	100%	100%	0%	90%	100%	100%	100%	100%	100%
	% Quantified	100%	100%	0%	0%	100%	100%	100%	100%	100%
EDTA Plasma (N=10)	Median (pg/mL)	52.7	84.9	ND	1.50	694	1.86	75.9	318	6.65
	Range (pg/mL)	24.5–158	26.3–254	NA	0.617–4.14	432–2140	0.984–3.78	45.7–113	137–1050	2.06–20.4
	% Detected	100%	100%	0%	100%	100%	100%	100%	100%	100%
	% Quantified	100%	100%	0%	0%	100%	70%	100%	100%	90%
Urine (N=10)	Median (pg/mL)	ND	ND	ND	ND	ND	ND	8.63	ND	ND
	Range (pg/mL)	NA	NA	NA	NA	NA	NA	0.548–33.2	NA	NA
	% Detected	0%	0%	0%	0%	0%	0%	80%	0%	0%
	% Quantified	0%	0%	0%	0%	0%	0%	50%	0%	0%

Concordance Comparison

Tested samples were plotted to evaluate concordance between the original and Gen B versions of the V-PLEX Chemokine Panel 1 Kit. Samples with in-well concentration outside the quantitative range were excluded from the analysis. Results for human (Figure 2) and NHP (Figure 3) samples are shown below. Assays show good agreement across both versions of the kit.

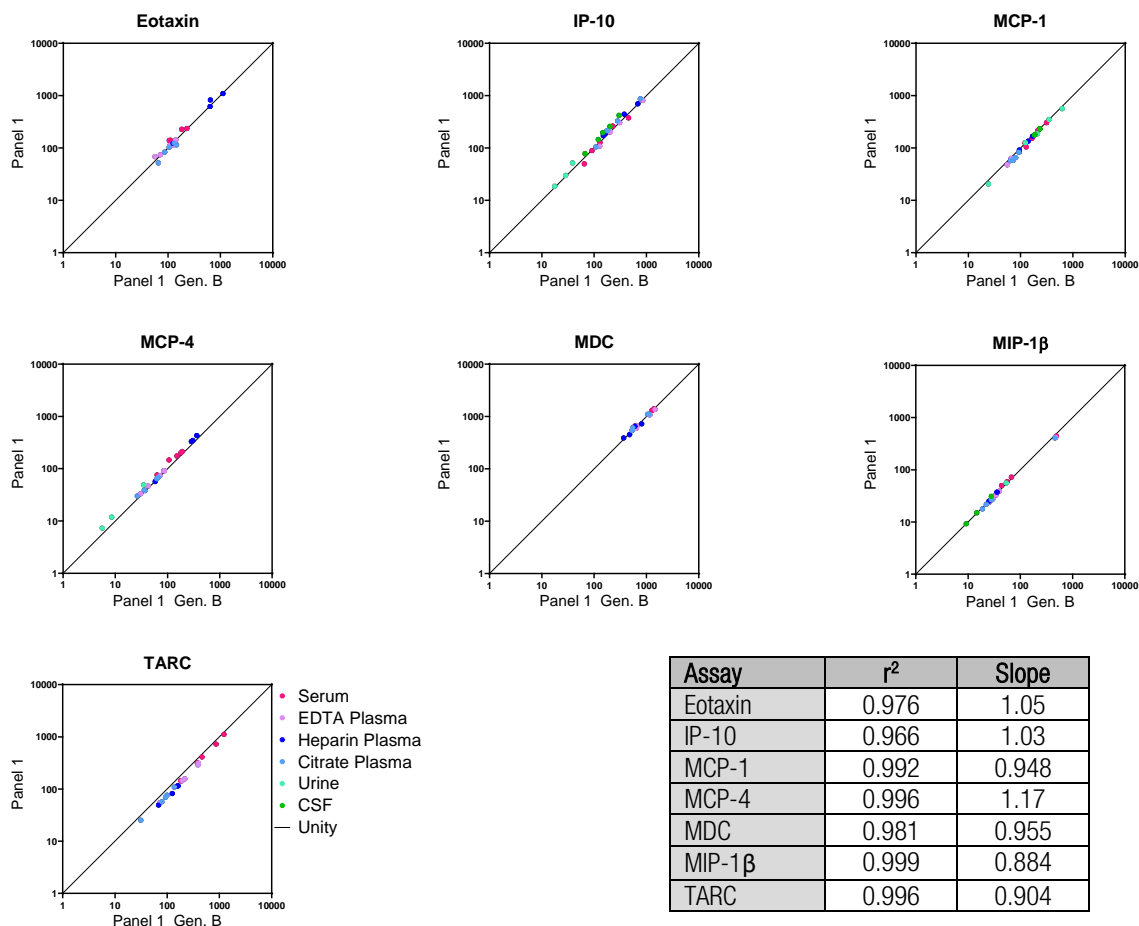


Figure 2: Human sample concordance between the V-PLEX Chemokine Panel 1 Kit (y-axis) and the V-PLEX Chemokine Panel 1 Gen B Kit (x-axis).

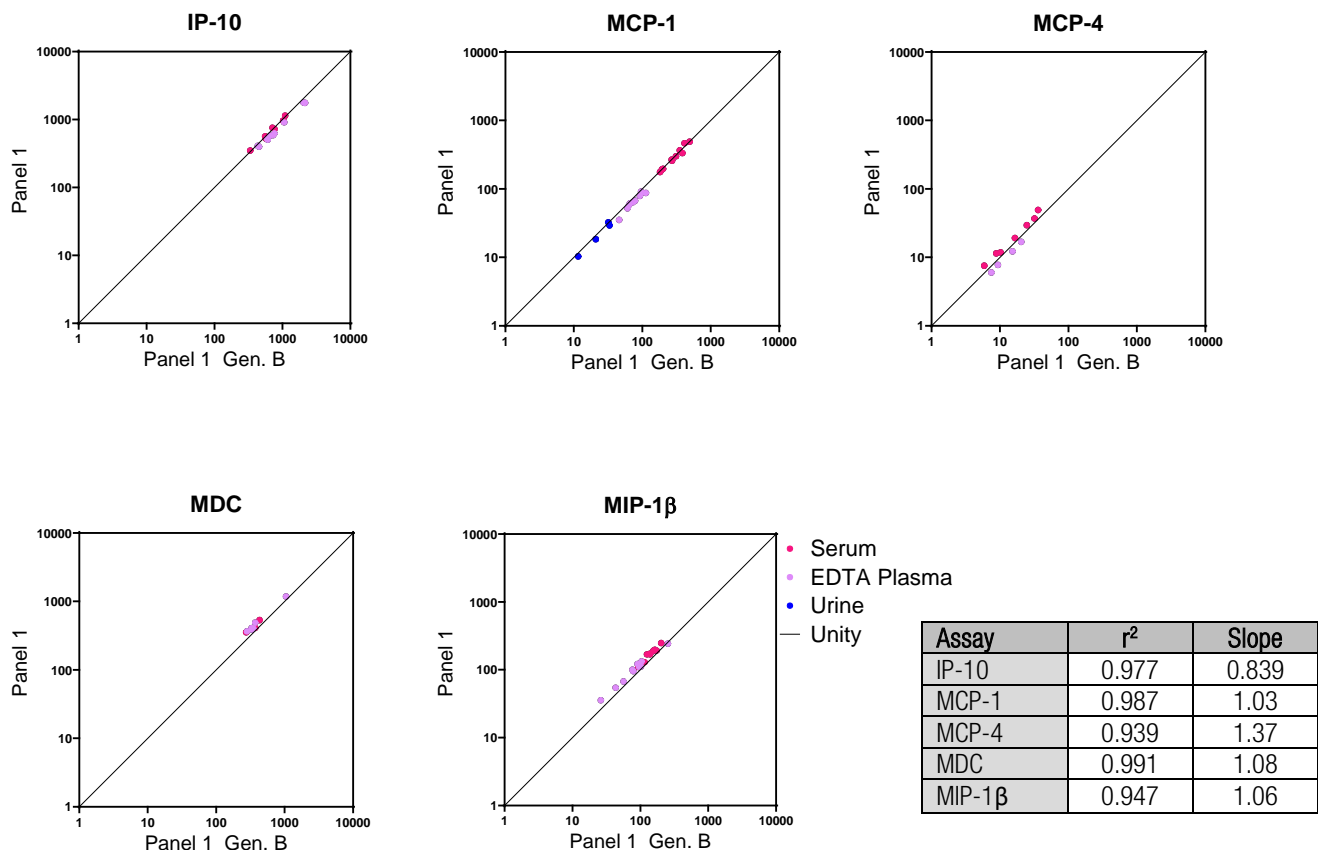


Figure 3: NHP sample concordance between the V-PLEX Chemokine Panel 1 Kit (y-axis) and the V-PLEX Chemokine Panel 1 Gen B Kit (x-axis).

Summary

The V-PLEX Chemokine Panel 1 Gen B Kit is an analytically validated and improved version of the V-PLEX Chemokine Panel 1 Kit with updated antibodies as well as a new assay diluent, read buffer, and protocol for improved sensitivity and increased longevity of critical reagents. The V-PLEX Chemokine Panel 1 Gen B Kit demonstrates reliable sample quantitation and strong concordance with the V-PLEX Chemokine Panel 1 Kit. More information and data can be found in the product insert for each panel.

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