STUDY REPORT

EVALUATION OF MP DIAGNOSTICS (MPD) HIV BLOT 2.2 MANUFACTURED BY MP BIOMEDICALS ASIA PACIFIC PTE LTD.

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OBJECT OF THE STUDY

The study was conducted on behalf of MP Biomedicals Asia Pacific Pte Ltd., based on the quotation of 27/5/2011 of the Testing Laboratory for IVD at the Paul-Ehrlich Institut (PEI-IVD), 63225 Langen, Germany. The purpose was to evaluate the sensitivity of the MPD HIV Blot 2.2 manufactured by MP Biomedicals Asia Pacific Pte Ltd. on 17 commercially available HIV seroconversion panel comparatively with the Chiron RIBA HIV-1/HIV-2 SIA manufactured by Chiron Novartis Vaccines and Diagnostics, Inc. Emeryville, CA 94608 (distributed by Ortho Clinical Diagnostics, Inc., Raritan, New Jersey 08869, USA).

ASSAY INFORMATION

Product name:	MP Diagnostics HIV Blot 2.2 Western Blot Assay
Catalogue no.:	11030-036 (36 tests)
Manufacturer:	MP Biomedicals Asia Pacific Pte Ltd.
Intended use	The MPD Diagnostics (MPD) HIV Blot 2.2 is a qualitative enzyme immunoassay for the in vitro detection of antibodies to human immunodeficiency virus type 1 (HIV-1) and type 2 (HIV-2) in human serum or plasma. It is intended for use as a more specific supplemental test on human serum or plasma specimens found repeatedly reactive using screening procedures such as the Enzyme-Linked Immunosorbent Assay (ELISA).
Lot no.:	AE1012
Sample size:	20 µl
Number of determinations per kit:	96 tests per kit
Expiry dates (lot):	01.01.2013
Storage temperature of the kit:	2-8 °C
Assay format:	Western blot
HIV antigens:	gp160, gp120, p66, p55, p51, p39, gp41, p31, p24, and p17, HIV-2 peptide
Conjugate:	Goat anti-human IgG conjugated with alkaline phosphatase. Contains sodium azide as preservative.
Substrate:	Solution of 5-bromo-4-chloro-3-indolyl-phosphate (BCIP) and nitroblue tetrazolium (NBT).
Controls supplied:	Non-reactive control, strong reactive control, weak Reactive control
Cut-off	N/A
Reading	Visual
Interpretation of results:	See table 1 below.

INTRODUCTION

The intended use, explanation of the tests and the principles of the test procedure are quoted from the instructions for use (IFU) of the two test kits:

1. MPD HIV Blot 2.2

"The MPD HIV Blot 2.2 is an independent supplemental test of high specificity to further confirm the presence of antibodies to HIV-1 and/or HIV-2" (reactive in ELISA). "The nitrocellulose strips are incorporated with separated, bound antigenic proteins from partially purified inactivated HIV-1 (gp160, gp120, p66, p55, p51, p39, gp41, p31, p24, and p17) using electrophoretic blotting, plus a specific HIV-2 synthetic peptide on the same strips. Individual nitrocellulose strips are incubated with diluted serum or plasma and controls. Specific antibodies to HIV-1 and HIV-2 if present in the specimens will bind to the HIV-1 proteins and HIV-2 peptide on the strips. The strips are washed to remove unbound materials. Antibodies that bind specifically to HIV proteins can be visualized using a series of reactions with goat anti-human IgG conjugated with alkaline phosphatase and the substrate BCIP/NBT. Positive and negative control serum specimens are run

simultaneously to allow identification of viral proteins."

2. Chiron RIBA HIV-1/HIV-2 Strip Immunoblot Assay (Chiron RIBA HIV-1/2 SIA)

"The Chiron RIBA HIV-1/HIV-2 SIA is an in vitro qualitative enzyme immunoassay (EIA) for the detection of antibodies to Human Immunodeficiency Virus Types 1 and 2 in human serum or plasma. It is intended for use in the confirmation of specimens found to be repeatedly reactive using a licensed screening procedure for anti-HIV-1 and/oranti-HIV-2. Additionally, it may be used to characterize antigenic specificity within the scope of HIV diagnosis."

The Chiron RIBA HIV-1/HIV-2 SIA is a three stage test which utilizes individual HIV-1/HIV-2 antigens immobilized as bands on nitrocellulose strips (the same matrix used in Western blots). In the first stage, the specimen or assay control is diluted and incubated with the strip. Antibodies to HIV-1, if present, will react to the corresponding HIV-1 antigen bands but not with the HIV-2 envelope peptide band on the nitrocellulose strip.HIV-2 antibodies, if present in the specimen, will bind to the HIV-2 envelope peptide band and to p26 present in theHIV-1 p24/HIV-2 p26 antigen band. HIV-2 antibodies, if present, will not cross react to the gp120 band. Due to homology between HIV-1 and HIV-2 in the gag, pol and env regions cross-reactivity may be observed in the HIV-1 p24 (present in the HIV-1 p24/HIV-2 p26 band), gp41 equivalent, or p31 antigens coated on the strip. Removal of unbound serum/plasma components is accomplished by aspiration and washing. In the second stage, the strip is incubated in the presence of a peroxidase-labeled goat anti-human IgG conjugate. The conjugate binds to the human IgG portion of the antigen-antibody complexes. Removal of unbound conjugate is accomplished by aspiration and subsequent wash steps. In the third stage, a colorimetric enzyme detection system composed of hydrogen peroxide and 4-chloro-1-naphthol is added. If bound conjugate is present, the enzymatic reaction will produce an insoluble blue-black colored reaction product at each antigen, peptide or control band. The color reaction involves the initial divalent oxidation of the peroxidase enzyme by hydrogen peroxide. Subsequent reduction of peroxidase to its initial state by two successive univalent interactions with soluble 4-chloro-1-naphthol results in the insoluble blue-black colored reaction product. After the development of color on the strip, the reaction is stopped by removal of the reactants and final wash steps. The visual band patterns which develop on each individual strip are the result of specific antibody being bound to each of the individual recombinant antigens and/or peptides on that strip. The reactivity of specimens towards each antigen band is determined by comparing the intensity of the individual antigen band with that of the Level I and Level II human IgG internal control bands blotted onto each strip.

The Interpretation criteria of test results for the two test kits are shown in **table 1**.

MPD HIV Blot 2.2

No viral specific bands present	Negative
Detection of p17 antibodies only, no other bands	Negative
Detection of 2 env (gp160/gp41and gp120) and GAG (p17, p24, p55) or POL (p31, p51, p66)	HIV-1 positive
Detection of 2 env (gp160/gp41 and gp120) and GAG (p17, p24, p55) or pol, (p31, p51, p66) and HIV-2 specific band is visible	HIV-1 positive with HIV-2 indicated
Any viral specific bands present but pattern does not meet criteria for Positive	Indeterminate
Any viral specific bands present but pattern does not meet criteria for Positive but HIV-2 specific band is visible.	Indeterminate with HIV-2 indicated

Chiron RIBA HIV-1/HIV-2 SIA

No bands present having 1+ or greater reactivity	Negative
1+ or greater reactivity to gp41 equivalent and any other HIV-1 antigen band, but <1+ reactivity to HIV-2 envelope peptide band	Positive for HIV-1 only
1+ or greater reactivity to the HIV-2 envelope peptide and any other HIV-1	Positive for HIV-2 only

band, but < 1+ reactivity to the gp120 band	
1+ or greater reactivity to HIV-1 gp41 equivalent, HIV-1 gp120 and the HIV-2 envelope peptide bands	Positive for HIV-1 and HIV-2
1+ or greater reactivity present to HIV bands, but the pattern does not meet the criteria for HIV-1 positive or HIV-2 positive	Indeterminate

MATERIAL AND METHODS

Assays

Six kits of one lot (no AE1012) of MPD HIV Blot 2.2 article no. 11030-036 (kit with 36 tests) were provided by MP Biomedicals Asia Pacific Pte Ltd. to the Testing Laboratory at the Paul Ehrlich Institute (PEI-IVD). Regarding Chiron RIBA HIV-1/HIV-2 SIA the manual version (product code 933130) was used. The details for the test dates, and the lots used are included in **table 2**.

Table 2: Lots used, expiry dates of the lots, and test dates

	Product name	**MPD HIV Blot 2.2**			**RIBA HIV-1/HIV-2 SIA**			
	Manufacturer	**MP Biomedicals**			**Chiron**			
	Cat.-no.	11030-036			933130			
No.	Supplier	Panel #	Lot	Test date	Expiry date	Lot	Test date	Expiry date	Panel samples
1	Seracare	PRB939	11030-037	14.06.2011	01.01.2013	109325	30.09.2011	31.12.2011	4-7
1	Seracare	PRB939	11030-036	14.06.2011	01.01.2013	XA1372	25.07.2007	31.10.2008	8-9
2	Seracare	PRB965	11030-036	14.06.2011	01.01.2013	109325	30.09.2011	31.12.2011	1-6
3	Seracare	PRB966	11030-036	14.06.2011	01.01.2013	109325	05.08.2011	31.12.2011	1-6
4	Seracare	PRB967	11030-036	14.06.2011	01.01.2013	109325	08.08.2011	31.12.2011	1-6
5	Seracare	PRB968	11030-036	14.06.2011	01.01.2013	109325	05.08.2011	31.12.2011	1-10
6	Seracare	PRB969	11030-036	17.06.2011	01.01.2013	109325	08.08.2011	31.12.2011	1-10
7	Seracare	PRB970	11030-036	15.06.2011	01.01.2013	109325	20.07.2011	31.12.2011	1-4
8	Seracare	PRB971	11030-036	15.06.2011	01.01.2013	109325	09.08.2011	31.12.2011	1-4
9	Seracare	PRB972	11030-036	15.06.2011	01.01.2013	109325	08.08.2011	31.12.2011	1-6
10	ZMC	6240	11030-036	16.06.2011	01.01.2013	109325	09.08.2011	31.12.2011	1-13
11	ZMC	6243	11030-037	15.06.2011	01.01.2013	109325	30.09.2011	31.12.2011	1-6
11	ZMC	6243	11030-036	15.06.2011	01.01.2013	XA1372	31.07.2007	31.10.2008	7-10
12	ZMC	6245	11030-036	15.06.2011	01.01.2013	109325	09.08.2011	31.12.2011	1-11
13	ZMC	6246	11030-036	16.06.2011	01.01.2013	109325	20.07.2011	31.12.2011	1-21
14	ZMC	9017	11030-036	15.06.2011	01.01.2013	95272	23.06.2009	20.01.2011	1-11
15	ZMC	9019	11030-036	16.06.2011	01.01.2013	109325	20.07.2011	31.12.2011	1-3
16	ZMC	9032	11030-037	14.06.2011	01.01.2013	109325	30.09.2011	31.12.2011	1-7
16	ZMC	9032	11030-036	16.06.2011	01.01.2013	WA8037	26.07.2007	21.06.2008	8-14
17	ZMC	12008	11030-036	16.06.2011	01.01.2013	109325	08.08.2011	31.12.2011	1-13

Seroconversion panels employed

The seroconversion panels were selected by MP Biomedicals based on, i) the criteria of the Common Technical Specifications (2009/886/EC) [CTS] of the IVD Directive 98/79/EC ("Sero-conversion HIV samples", "Early sero-conversion HIV samples" and general principle 3.1.8.3), ii) start with negative bleed (defined as negative by antibody test); iii) have narrow bleeding intervals (defined by MP as ≤10 days from negative to positive bleed), and include at least a positive or indeterminate results for HIV Western blots and/or immunoblots. As a result, a total of 17 commercially available seroconversion samples as displayed in **table 5** were selected by MP Biomedicals HIV Blot 2.2 and tested at PEI-IVD. The panels were from Zeptometrix, (375 West Street, W. Bridgewater, MA 02379, USA) and SeraCare, (25 Kenwood Circle, Suite 14A, MA 02038, USA).

Laboratory testing

The 17 seroconversion panels were tested at PEI-IVD. One technician carried out the testing for each test kit

according to the respective instructions for use (IFU): for MPD HIV Blot 2.2 the IFU version MAE 0011-GER-2, revision date 07/09; for RIBA HIV-1/HIV-2 SIA the IFU version 03-2008. Prior to testing the samples were centrifuged at 10,000 rpm, 10 minutes (Centrifuge 5403, Eppendorf, 22339 Hamburg, Germany).

Discrepancy resolution
Determinations with the MPD HIV Blot 2.2 were carried out in single measurements. Results discrepant from the assigned sample status were repeat tested also in single measurement.

Analysis of sensitivity in seroconversion panels
The detection of HIV in the seroconversion panels by MPD HIV Blot 2.2 was compared with the Chiron RIBA HIV-1/HIV-2 SIA. The number of bleedings detected positive for HIV 1/2 was counted for each panel and compared between the two test kits. The average score for the sensitivity of an assay was the total number of positive bleeds divided by the total number of panels tested. Timing of anti-HIV detection was the time difference (days) in detection of the 1st HIV positive sample of a panel between MPD HIV Blot 2.2 and Chiron RIBA HIV-1/HIV-2 SIA. The overall day delay was then the total day difference divided by the number of panels tested. In the case where an assay is negative or indeterminate over all seroconversion follow up bleeds, two additional days are assigned.

RESULTS

HIV-1 Seroconversion Sensitivity
The results are displayed in **tables 3-5**. Panel PRB939 did not meet the criteria for a narrow bleed interval because the interval in the actual seroconversion between panel members 939-8 to 939-9 is 80 days and panel PRB971 was negative in all bleeds for both, MPD HIV Blot 2.2 and Chiron RIBA HIV-1/HIV-2 SIA. Thus, 15 seroconversion panels out of the total 17 remained that were qualified according to the CTS and which were used for the comparative analysis, with 147 samples in total.

Overall, MPD HIV Blot 2.2 was positive in 23 bleeds, indeterminate in 20 bleeds and negative in 104 bleeds. Chiron RIBA HIV-1/HIV-2 SIA was positive in 25 bleeds, indeterminate in 16, negative in 103 and invalid in 3 bleeds.

Table 3 shows the performance of kit based on detection of Positives, whereas table 4 shows the performance based on detection of Indeterminates/Positives.

Table 3: Performance of Kit based on Positives Detection

Performance	Panels	No of Panels
MPD and Chiron have equal detection of positives	PRB967, PRB 968, ZMC6245, ZMC6246, ZMC9017, ZMC9019, ZMC12008	7
MPD detected Positives earlier	ZMC6240, PRB965	2
Chiron detected Positives earlier	PRB969, PRB970, ZMC9032	3

Table 4: Performance of Kit based on Positives and/or Indeterminates

Performance	Panels	No of Panels
MPD and Chiron have equal detection of Positives and Indeterminates	PRB965, PRB966, PRB968, PRB969, PRB970, ZMC6243, ZMC6245, ZMC6246, ZMC9019, PRB9032,	10
MPD detected Positives or Indeterminates earlier	PRB967, PRB972, ZMC6240, ZMC12008	4
Chiron detected Positives or Indeterminates earlier	PRB9017	1

Referring to table 3, in seven seroconversion panels, MPD HIV Blot 2.2 and Chiron RIBA HIV-1/HIV-2 SIA were positive in the same panel follow-up samples. In two panels, MPD HIV Blot 2.2 detected HIV one bleed earlier than Chiron RIBA HIV-1/HIV-2. In one panel, RIBA HIV-1/HIV-2 SIA was two bleeds earlier positive (ZMC9032) and in another two panels (PRB969, PRB970) RIBA HIV-1/HIV-2 SIA was one bleed earlier compared to MPD HIV Blot 2.2.

Referring to table 4, in ten seroconversion panels, MPD HIV Blot 2.2 and Chiron RIBA HIV-1/HIV-2 SIA were positive or indeterminate in the same panel follow-up samples. In four panels, MPD HIV Blot 2.2 detected HIV (as indeterminates or positives) earlier than Chiron RIBA HIV-1/HIV-2. Chiron RIBA HIV-1/HIV-2 detected HIV (as indeterminates or positives) earlier than MPD HIV Blot 2.2 in one panel.

Referring to table 5 showing the detailed results, altogether this represents on average a score of 0.14 more positive bleeds per panel for RIBA HIV-1/HIV-2 SIA compared to MPD HIV Blot 2.2. With respect to timing of HIV detection, this represents an overall day delay in detection of HIV for MPD HIV Blot 2.2 of one day in total or 0.07 days per panel. However, if indeterminates are considered, MPD HIV Blot 2.2 would have 43 samples reacting positive or indeterminate, compared to 41 samples with RIBA HIV-1/HIV-2 SIA. This is a score of 0.14 more indeterminate/positive bleeds per panel for MPD HIV Blot 2.2 than RIBA HIV-1/HIV-2 SIA and an overall earlier detection of 8 days for MPD HIV Blot 2.2 or 0.53 days per panel. This is appropriate because the instructions for use of MPD HIV Blot 2.2. require that indeterminate blots require testing of sequential samples and because the CTS define “seroconversion HIV samples” for confirmatory assays as positive or indeterminate.

It is noteworthy that the MPD HIV Blot 2.2 was indeterminate in those follow up samples where RIBA HIV-1/HIV-2 SIA reacted earlier positive than MPD HIV Blot 2.2. The band pattern in these samples was gp160 and p24 positive corresponding to the pattern gp41 (product of gp160) and p24/p26 positive in RIBA HIV-1/HIV-2 SIA in the same samples. This may be important for Western blot interpretation by different regulatory authorities (WHO, FDA, CDC, ARC, DVV Germany, etc.), which in some cases require one HIV env only for positive reading. This implies that Western blot with the presence of just gp160 and p24 will be considered positive by the respective regulatory authorities leading to twelve additional positive panel members for MPD HIV Blot 2.2: 968-8, 969-8, 970-4, 972-6, 6243-10, 6245-9, 6246-17, 9017-10, 9032-9, 9032-10, 9032-11, 12008-11.

In two panels (ZMC6246 and ZMC9032) MPD HIV Blot 2.2 showed non-specific reactivity characterized by bands that remain unchanged over the course of the panel and that have no corresponding antibody reactivity in HIV 1/2 screening assays. The blot results with this band were therefore considered negative. Also with RIBA HIV-1/HIV-2 SIA in panel ZMC12008 there was “+/-“ reactivity for p24/p26 across all panel members and with ZMC9032 with gp41, p31 and p24/p26 below the level I reference band. In case of panel ZMC9032 this non-specificity may have interfered with the actual sensitivity determination in RIBA HIV-1/HIV-2 SIA.

In the case of RIBA HIV-1/HIV-2 SIA, 3 invalid results were observed with weak or missing control bands in samples PRB970-1, ZMC6246-5, ZMC6246-16.

Table 5: Seroconversion sensitivity of MPD HIV Blot 2.2 on 17 HIV seroconversion panels in comparison to Chiron RIBA HIV-1/HIV-2 SIA

			Test name	**MPD HIV Blot 2.2**											**Chiron RIBA HIV-1/HIV-2 SIA**							
			Manufacturer	MP Biomedicals Asia Pacific Pte Ltd, 2 Pioneer Place, Singapore 627885											Chiron Novartis Vaccines and Diagnostics, Inc. Emeryville, CA 94608							
			Catalogue.-no.	AE1012											933130							
			Lot no.	11030-036											Various (see table 2)							
			Expiry date	01.01.2013																		
			Method	Rapid assay (60´, 60´, 15´, RT)											Manual							
	Supplier	Panel #	Day since 1st bleed	HIV2	p17	p24	p31	gp41	p51	p55	p66	gp120	gp160	Result	gp120	gp41	HIV2	p31	p24/26	Result	Day diff. (pos)	Day diff. (ind/pos)
1	Seracare	PRB965-1	0	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	neg		
	Seracare	PRB965-2	5	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	neg		
	Seracare	PRB965-3	7	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	neg		
	Seracare	PRB965-4	12	-	-	-	-	-	-	-	-	-	+	ind	-	1+	-	-	-	ind		0
	Seracare	PRB965-5	14	-	-	+	-	-	-	-	-	+	+	pos	-	2+	-	-	-	ind	-7	
	Seracare	PRB965-6	21	-	-	+	-	-	-	-	-	+	+	pos	-	4+	-	-	2+	pos		
2	Seracare	PRB966-1	0	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	neg		
	Seracare	PRB966-2	2	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	neg		
	Seracare	PRB966-3	20	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	neg		
	Seracare	PRB966-4	22	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	neg		
	Seracare	PRB966-5	30	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	neg		
	Seracare	PRB966-6	35	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	neg		
	Seracare	PRB966-7	37	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	neg		
	Seracare	PRB966-8	44	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	neg		
	Seracare	PRB966-9	48	-	-	-	-	-	-	-	-	-	-	neg	-	+/-	-	+/-	-	neg		
	Seracare	PRB966-10	51	-	-	-	-	-	-	-	-	-	+	ind	-	3+	-	+/-	-	ind	0	0
3	Seracare	PRB967-1	0	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	neg		
	Seracare	PRB967-2	3	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	neg		
	Seracare	PRB967-3	7	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	neg		
	Seracare	PRB967-4	17	-	-	-	-	-	-	-	-	-	+	ind	-	+/-	-	-	-	neg		-2
	Seracare	PRB967-5	19	-	-	-	-	-	-	-	-	-	+	ind	-	1+	-	-	-	ind		
	Seracare	PRB967-6	24	-	-	+	+	-	-	-	-	+	+	pos	-	3+	-	-	2+	pos	0	

4	Seracare	PRB968-1	0	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	neg		
	Seracare	PRB968-2	3	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	neg		
	Seracare	PRB968-3	8	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	neg		
	Seracare	PRB968-4	10	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	neg		
	Seracare	PRB968-5	15	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	neg		
	Seracare	PRB968-6	17	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	neg		
	Seracare	PRB968-7	26	-	-	-	-	-	-	-	-	-	-	neg	-	+/-	-	-	-	neg		
	Seracare	PRB968-8	28	-	-	+	-	-	-	-	-	-	+	ind	-	1+	-	-	-	ind		0
	Seracare	PRB968-9	33	-	-	+	-	-	-	-	-	+	+	pos	-	4+	-	+/-	3+	pos	0	
	Seracare	PRB968-10	35	-	-	+	-	-	-	-	-	+	+	pos	-	4+	-	+/-	3+	pos		
5	Seracare	PRB969-1	0	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	neg		
	Seracare	PRB969-2	29	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	neg		
	Seracare	PRB969-3	48	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	neg		
	Seracare	PRB969-4	53	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	neg		
	Seracare	PRB969-5	55	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	neg		
	Seracare	PRB969-6	61	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	neg		
	Seracare	PRB969-7	63	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	neg		
	Seracare	PRB969-8	70	-	-	+	-	-	-	-	-	-	+	ind	+/-	2+	-	-	1+	pos	2	0
	Seracare	PRB969-9	72	-	-	+	-	-	-	-	-	+	+	pos	+/-	3+	-	-	3+	pos		
	Seracare	PRB969-10	77	-	-	+	-	-	-	+	-	+	+	pos	1+	4+	-	-	4+	pos		
6	Seracare	PRB970-1	0	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	invalid		
	Seracare	PRB970-2	7	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	neg		
	Seracare	PRB970-3	10	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	neg		
	Seracare	PRB970-4	14	-	-	+	-	-	-	-	-	-	+	ind	2+	-	-	-	2+	pos	2	0
7	Seracare	PRB972-1	0	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	neg		
	Seracare	PRB972-2	3	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	neg		
	Seracare	PRB972-3	11	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	neg		
	Seracare	PRB972-4	14	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	neg		
	Seracare	PRB972-5	18	-	-	-	-	-	-	-	-	-	-	neg	-	+/-	-	-	-	neg		
	Seracare	PRB972-6	21	-	-	+	-	-	-	-	-	-	+	ind	-	+/-	-	-	-	neg	0	-2
8	ZMC	6240-1	0	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	neg		
	ZMC	6240-2*	2	n.t.	n.t.	n.t.	n.t.	n.t.	n.t.	n.t.	n.t.	n.t.	n.t.	N/A	n.t.	n.t.	n.t.	n.t.	n.t.	N/A		
	ZMC	6240-3	7	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	neg		
	ZMC	6240-4	9	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	neg		

	ZMC	6240-5	14	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	neg		
	ZMC	6240-6	16	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	neg		
	ZMC	6240-7	21	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	neg		
	ZMC	6240-8	23	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	neg		
	ZMC	6240-9	28	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	neg		
	ZMC	6240-10	30	-	-	-	-	-	-	-	-	-	+	ind	-	+/-	-	-	-	neg		-6
	ZMC	6240-11	36	-	-	+	-	-	-	-	-	+	+	pos	-	4+	-	-	+/-	ind		
	ZMC	6240-12	45	-	-	+	-	-	-	+	-	+	+	pos	-	4+	-	-	3+	pos	-9	
	ZMC	6240-13	53	-	-	+	-	+	-	+	-	+	+	pos	-	4+	-	-	3+	pos		
9	ZMC	6243-1	0	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	neg		
	ZMC	6243-2	5	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	neg		
	ZMC	6243-3	7	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	neg		
	ZMC	6243-4	12	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	neg		
	ZMC	6243-5	18	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	neg		
	ZMC	6243-6	20	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	neg		
	ZMC	6243-7	25	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	neg		
	ZMC	6243-8	28	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	neg		
	ZMC	6243-9	33	-	-	-	-	-	-	-	-	-	+	ind	-	2+	-	-	-	ind	0	0
	ZMC	6243-10	35	-	-	+	-	-	-	-	-	-	+	ind	-	3+	-	-	-	ind		
10	ZMC	6245-1	0	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	neg		
	ZMC	6245-2	46	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	neg		
	ZMC	6245-3	52	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	neg		
	ZMC	6245-4	56	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	neg		
	ZMC	6245-5	61	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	neg		
	ZMC	6245-6	67	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	neg		
	ZMC	6245-7	70	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	neg		
	ZMC	6245-8	74	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	neg		
	ZMC	6245-9	77	-	-	+	-	-	-	-	-	-	+	ind	-	2+	-	-	+/-	ind		0
	ZMC	6245-10	83	-	-	+	-	-	-	-	-	+	+	pos	-	4+	-	-	3+	pos	0	
	ZMC	6245-11	85	-	-	+	-	-	-	-	-	+	+	pos	-	4+	-	-	3+	pos		
11	ZMC	6246-1	0	-	-	-	-	-	-	-	+	-	-	neg[5)]	-		-	-	-	neg		
	ZMC	6246-2	2	-	-	-	-	-	-	-	+	-	-	neg[5]	-	-	-	-	-	neg		
	ZMC	6246-3	12	-	-	-	-	-	-	-	+	-	-	neg[5)]	-	-	-	-	-	neg		
	ZMC	6246-4	14	-	-	-	-	-	-	-	+	-	-	neg[5]	-	-	-	-	-	neg		
	ZMC	6246-5	19	-	-	-	-	-	-	-	+	-	-	neg[5)]	-	-	-	-	-	invalid		

	ZMC	6246-6	22	-	-	-	-	-	-	-	+	-	-	neg[5]	-	-	-	-	-	neg		
	ZMC	6246-7	26	-	-	-	-	-	-	-	+	-	-	neg[5)]	-	-	-	-	-	neg		
	ZMC	6246-8	29	-	-	-	-	-	-	-	+	-	-	neg[5]	-	-	-	-	-	neg		
	ZMC	6246-9	33	-	-	-	-	-	-	-	+	-	-	neg[5)]	-	-	-	-	-	neg		
	ZMC	6246-10	36	-	-	-	-	-	-	-	+	-	-	neg[5]	-	-	-	-	-	neg		
	ZMC	6246-11	40	-	-	-	-	-	-	-	+	-	-	neg[5)]	-	-	-	-	-	neg		
	ZMC	6246-12	43	-	-	-	-	-	-	-	+	-	-	neg[5]	-	-	-	-	-	neg		
	ZMC	6246-13	47	-	-	-	-	-	-	-	+	-	-	neg[5)]	-	-	-	-	-	neg		
	ZMC	6246-14	50	-	-	-	-	-	-	-	+	-	-	neg[5]	-	-	-	-	-	neg		
	ZMC	6246-15	54	-	-	-	-	-	-	-	+	-	-	neg[5)]	-	-	-	-	-	neg		
	ZMC	6246-16	57	-	-	-	-	-	-	-	+	-	-	neg[5)]	-	-	-	-	-	invalid		
	ZMC	6246-17	61	-	-	+	-	-	-	-	+	-	+	ind	-	2+	-	-	+/-	ind		0
	ZMC	6246-18	64	-	-	+	-	-	-	-	+	+	+	pos	-	3+	-	-	2+	pos	0	
	ZMC	6246-19	70	-	-	+	-	+	-	-	+	+	+	pos	-	3+	-	-	3+	pos		
	ZMC	6246-20	72	-	-	+	-	+	-	-	+	+	+	pos	-	3+	-	-	4+	pos		
	ZMC	6246-21	77	-	-	+	-	+	-	-	+	+	+	pos	4+	-	-	-	4+	pos		
12	ZMC	9017-1	0	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	neg		
	ZMC	9017-2	3	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	neg		
	ZMC	9017-3	7	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	neg		
	ZMC	9017-4	10	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	neg		
	ZMC	9017-5	14	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	neg		
	ZMC	9017-6	17	-	-	-	-	-	-	-	-	-	-	neg	-	1+	-	-	-	ind		7
	ZMC	9017-7	21	-	-	-	-	-	-	-	-	-	-	neg	-	2+	-	-	-	ind		
	ZMC	9017-8	24	-	-	-	-	-	-	-	-	-	+	ind	-	2+	-	-	-	ind		
	ZMC	9017-9	28	-	-	+	-	-	-	-	-	-	+	ind	-	3+	-	-	-	ind		
	ZMC	9017-10	32	-	-	+	-	-	-	-	-	-	+	ind	-	4+	-	-	-	ind		
	ZMC	9017-11	35	-	-	+	-	-	-	-	-	+	+	pos	-	4+	-	-	1+	pos	0	
13	ZMC	9019-1	0	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	neg		
	ZMC	9019-2	3	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	neg		
	ZMC	9019-3	8	-	+	+	+	+	+	+	+	+	+	pos	+/-	4+	-	2+	4+	pos	0	0
14	ZMC	9032-1	0	-	-	+	-	-	-	-	-	-	-	neg[5)]	-	-	-	-	-	neg		
	ZMC	9032-2	2	-	-	+	-	-	-	-	-	-	-	neg[5]	-	+/-	-	+/-	+/-	neg		
	ZMC	9032-3	7	-	-	+	-	-	-	-	-	-	-	neg[5)]	-	+/-	-	+/-	+/-	neg		
	ZMC	9032-4	10	-	-	+	-	-	-	-	-	-	-	neg[5]	-	+/-	-	+/-	+/-	neg		
	ZMC	9032-5	14	-	-	+	-	-	-	-	-	-	-	neg[5)]	-	+/-	-	+/-	+/-	neg		

	ZMC	9032-6	17	-	-	+	-	-	-	-	-	-	-	neg[5]	-	+/-	-	+/-	+/-	neg		
	ZMC	9032-7	22	-	-	+	-	-	-	-	-	-	-	neg[5)]	-	+/-	-	+/-	+/-	neg		
	ZMC	9032-8	24	-	-	+	-	-	-	-	-	-	-	neg[5]	-	+/-	-	+/-	+/-	neg		
	ZMC	9032-9	29	-	-	+	-	-	-	-	-	-	+	ind	-	+/-	-	+/-	1+	ind		
	ZMC	9032-10	36	-	-	+	-	-	-	-	-	-	+	ind	-	2+	-	+/-	4+	pos	13	0
	ZMC	9032-11	38	-	-	+	-	-	-	-	-	-	+	ind	-	2+	-	+/-	4+	pos		
	ZMC	9032-12	49	-	-	+	-	-	-	-	-	+	+	pos	-	2+	-	+/-	4+	pos		
	ZMC	9032-13	51	-	-	+	-	-	-	-	-	+	+	pos	-	2+	-	+/-	4+	pos		
	ZMC	9032-14	56	-	-	+	-	-	-	-	-	+	+	pos	+/-	2+	-	+/-	4+	pos		
15	ZMC	12008-1	0	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	+/-	neg		
	ZMC	12008-2	2	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	+/-	neg		
	ZMC	12008-3	7	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	+/-	neg		
	ZMC	12008-4	9	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	+/-	neg		
	ZMC	12008-5	14	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	+/-	neg		
	ZMC	12008-6	16	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	+/-	neg		
	ZMC	12008-7	21	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	+/-	neg		
	ZMC	12008-8	23	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	+/-	neg		
	ZMC	12008-9	28	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	+/-	neg		
	ZMC	12008-10	33	-	-	-	-	-	-	-	-	-	-	neg	-	+/-	-	-	+/-	neg		
	ZMC	12008-11	35	-	-	+	-	-	-	-	-	-	+	ind	-	+/-	-	-	+/-	neg		-5
	ZMC	12008-12	40	-	-	+	-	-	-	-	-	+	+	pos	-	3+	-	-	2+	pos	0	
	ZMC	12008-13	42	-	-	+	-	-	-	-	-	+	+	pos	-	3+	-	-	2+	pos		
		Total number of samples tested												147						147		
		Total positive												23						25		
		Total indeterminate												20						16		
		Total negative												104						103		
		Invalid												0						3		
		Positive score [1)]												1.53						1.67		
		Total day delay																			1	
		Day delay per panel [2)]												0.07								
		Positive/indeterminate score [3)]												2.87						2.73		
		Total day delay																				-8
		Day delay per panel [4)]												-0.53								

16	Seracare PRB939-1	0	-	-	-	-	-	-	-	-	-	-	neg	n.t.	n.t.	n.t.	n.t.	n.t.	N/A		
	Seracare PRB939-2	2	-	-	-	-	-	-	-	-	-	-	neg	n.t.	n.t.	n.t.	n.t.	n.t.	N/A		
	Seracare PRB939-3	7	-	-	-	-	-	-	-	-	-	-	neg	n.t.	n.t.	n.t.	n.t.	n.t.	N/A		
	Seracare PRB939-4	9	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	neg		
	Seracare PRB939-5	14	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	neg		
	Seracare PRB939-6	16	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	neg		
	Seracare PRB939-7	21	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	neg		
	Seracare PRB939-8	23	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	neg		
	Seracare PRB939-9	103	-	+	+	+	+	+	-	+	+	+	pos	1+	4+	-	2+	2+	pos	0	0
17	Seracare PRB971-1	0	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	neg		
	Seracare PRB971-2	7	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	neg		
	Seracare PRB971-3	10	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	neg		
	Seracare PRB971-4	14	-	-	-	-	-	-	-	-	-	-	neg	-	-	-	-	-	neg	N/A	N/A

Footnotes and Abbreviations:

1) Total positives divided by 15 panels
2) Total day delay (positives) divided by 15 panels
3) Total positives & indeterminates divided by 15 panels
4) Total day delay (positives & indeterminates) divided by 15 panels; minus sign = earlier detection MPD HIV Blot 2.2
5) Non-specific reactivity to p66 in ZMC6246, p24 in ZMC9032 respectively, is suspected because other assays showed absence of antibody to HIV and because these bands remained unchanged over the course of the panel; therefore these panel members are interpreted as negative.

Green = positive result; blue-green = indeterminate result
+/- band intensity < level I IgG reference band in RIBA HIV-1/HIV-2 SIA
* Sample 6240-2 exhausted
Neg = negative; pos = positive; ind = indeterminate; ZMC = Zeptometrix; Day diff. = day difference in detection of HIV; n.t. = not tested; N/A = not applicable

SUMMARY

MPD HIV Blot 2.2 manufactured by MP Biomedicals Asia Pacific Pte Ltd. was tested at the Paul-Ehrlich-Institut in order to evaluate the serocoversion sensitivity in comparison with the Chiron RIBA HIV-1/HIV-2 SIA. In summary, if only positive bleeds are considered, MPD HIV Blot 2.2 detected HIV on average 0.14 bleed per panel later than Chiron RIBA HIV-1/HIV-2 SIA or 0.07 day per panel later. This represents only a small difference between the two assays.

On the other hand, if indeterminates and positives are considered, MPD HIV Blot 2.2 detected HIV on average 0.14 bleed per panel earlier than Chiron RIBA HIV-1/HIV-2 SIA or 0.53 days per panel earlier. In addition, the indeterminate results by MPD HIV Blot 2.2 showed a similar band reaction pattern as Chiron RIBA HIV-1/HIV-2 SIA.

Overall the seroconversion sensitivity of MPD HIV Blot 2.2 and RIBA HIV-1/2 SIA are comparable and both assays reacted similarly in the same panel follow up samples.

28/11/2011

Date Signed by Dr. H. Scheiblauer (PEI-IVD)