

# Performance evaluation of the Determine® HIV-1/2 Ag/Ab Combo, a novel 4<sup>th</sup> generation rapid test



Tomer Keren, Thierry Guidasci, Baruch Rivetz, Falk Fish

Inverness Medical Innovations, Inc., 51 Sawyer Road, Suite 200, Waltham, MA 02453 USA

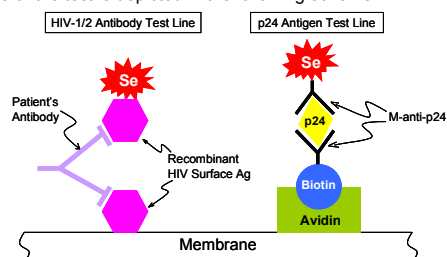


## Introduction

Calling for a move towards universal access to HIV prevention, treatment, care and support, the WHO, UNAIDS and health organizations are encouraging the use of rapid HIV diagnostics as a tool to effectively address the epidemic. Widely used 3<sup>rd</sup> generation antibody assays do not detect acute/early infection. Fourth generation assays, able to shorten the "diagnostic window" by detecting both antibodies and p24 antigen, are setting the standard in HIV case identification. However, most 4<sup>th</sup> generation tests are in EIA format and do not differentiate the two markers. Also, EIA's are intended for deployment in central labs and are not accessible in remote areas where the need is significant. Inverness Medical has developed the Determine® HIV-1/2 Ag/Ab Combo, the first single rapid immunochromatographic test strip, which provides simultaneous and differentiated detection of HIV-1 p24 antigen and antibodies to HIV-1/2. The test format is on the highly regarded, compact and the easy-to-use Determine® platform. The performance of the new Determine® Combo has been evaluated in-house and at 9 clinical sites across Africa, Asia, Europe and Latin America. The aim of this study was to assess the performance of the Determine® HIV-1/2 Ag/Ab Combo in comparison to 3<sup>rd</sup> and other 4<sup>th</sup> generation HIV tests. The test is CE marked and USAID approved.

## Method

The principle of the test is depicted in the following scheme:

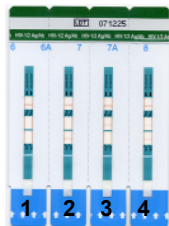


A procedural control bar is incorporated in the assay device.

Fifty µL of each sample were applied to the sample pad. The results were read 20 minutes thereafter.

### Interpretation of the test results:

1. Negative
2. Positive for Antibodies
3. Positive for HIV-1 p24 antigen
4. Positive for both Antibodies and p24 antigen



## Specimens

**Positive** specimens included a total of 1179 samples from the chronic stage of the infection of patients with: HIV-2, HIV-1, HIV-1 group O, HIV-1 non B subtypes (A, C, D, F, J, G, H, I, K, CRF01\_AE, CRF02\_AG, CRF06\_cpx, CRF09\_cpx, CRF11\_cpx, CRF35\_AD and BD) and HIV-1 and HIV-2 samples from patients at the stage of high risk, pre- and per-seroconversion. Both retrospective (frozen serum/plasma) and prospective (fresh whole blood and serum/plasma) samples were included.

**Negative** specimens included a total of 2343 confirmed serum, plasma or whole blood specimens of blood donors, pregnant women, disease states other than HIV and potential interferences.

## Results

### Overall Sensitivity

The diagnostic sensitivity of Determine® HIV-1/2 Ag/Ab Combo on the 1179 patients at the chronic stage of infection is calculated to be 100%.

Types	Number of Specimens Tested	Positive by Determine® HIV-1/2 Ag/Ab Combo	Sensitivity
HIV-1	885	885	100.00%
HIV-1 group O	7	7	100.00%
HIV-1 non B subtypes*	154	154	100.00%
HIV-2	133	133	100.00%
<b>Total</b>	<b>1179</b>	<b>1179</b>	<b>100.00%</b>

\*Subtypes: A, C, D, F, G, H, I, K, and CRF AE, AG, AD, BD, 06, 09 and 011.

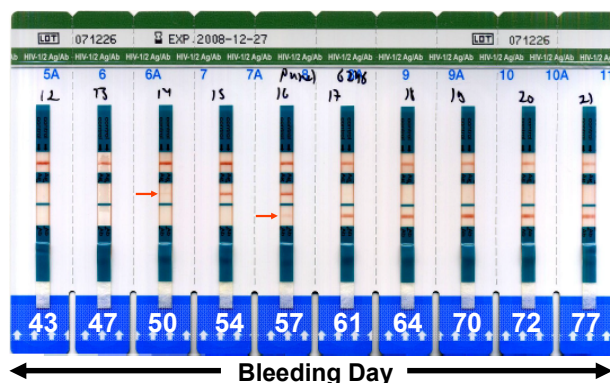
### Overall Specificity

Population	Number of Specimens Tested	Negative by Determine® HIV-1/2 Ag/Ab Combo Ab Test line	Specificity (%) of the Antibody Test line	Negative by Determine® HIV-1/2 Ag/Ab Combo Ag Test line	Specificity (%) of the Antigen Test line
Seronegative specimens	1783	1769	99.21%	1776	99.61%
Pregnant women	200	200	100.00%	199	99.50%
Disease States Other than HIV and Potentially Interfering Substances*	360	356	98.89%	360	100.00%
<b>Total</b>	<b>2343</b>	<b>2325</b>	<b>99.23%</b>	<b>2335</b>	<b>99.66%</b>

\*IV drug users, rheumatoid factor, cancer, alcoholic cirrhosis, autoimmune (ANA), high cholesterol, lipemic, high bilirubin, hemolyzed, anti mouse IgG and other viral or bacterial infections, multiparous, (HBV, HCV, HTLV, CMV, Toxo IgG, Syphilis, HSV 1/2, EBV, Flu vaccinated patients and Chlamydia IgG/IgM).

## Sensitivity for Early/Acute Phase Infection

The capability of any HIV diagnostic to detect early (=acute) infection can be demonstrated by testing commercial seroconversion panels. The following image reflects the results of testing panel BCP 6246 with the Determine® HIV-1/2 Ag/Ab Combo test:



The Determine® Combo was further tested on 32 additional commercial seroconversion panels. The results are summarized in the following table:

Panel	Bleeding Day with First Positive Result					"Window" Shortened by (days):
	Reference Test Results		Determine® HIV-1/2	Determine® HIV-1/2		
	p24 Ag	3rd Gen EIA	Ag/Ab Combo	p24 Ag	Antibody	
BBI-AB	28	28	28	28	33	0
BBI-AE	3	7	7	3	7	4
BBI-AF	15	28	28	28	28	0
BBI-AG	27	34	27	27	34	0
BBI-AI	0	7	7	0	7	7
BBI-AL	14	21	-	14	-	8
BBI-AM	0	9	9	0	9	9
BBI-AN-E	16	103	103	21	103	82
BBI-AP	7	11	11	7	11	4
BBI-AQ	9	18	18	18	18	0
BBI-AS	12	14	19	12	19	7
BBI-AT	2	14	14	7	14	7
BBI-AU	13	13	15	13	15	2
BBI-AV	18	20	18	18	20	0
BBI-BA	8	19	19	11	19	8
BBI-BB	10	14	17	10	17	7
BBI-BC	7	7	10	7	10	3
BBI-BD	17	-	-	-	-	0
BBI-BE	3	12	12	7	12	5
BBI-BG	23	28	28	23	28	5
BBI-BH	9	15	15	9	15	6
BBI-BI	0	9	9	7	12	2
BBI-J	14	26	26	14	26	12
BBI-L	0	0	0	0	9	0
BBI-Q	53	60	60	53	65	7
BBI-W	37	47	84	47	84	37
BBI-Y	44	44	44	44	44	0
BBI-Z	7	27	27	7	27	20
BCPI-6240	23	28	28	23	30	5
BCPI-6244	28	33	33	33	35	0
BCPI-6246	50	57	57	50	57	7
BCPI-9020	95	98	95	95	95	0
BCPI-9021	50	57	57	50	57	7

The results show that, depending on the sampling frequency in the individual panels, Determine® HIV-1/2 Ag/Ab Combo is able to identify infection in 2-87 days earlier than 3<sup>rd</sup> generation antibody tests. In 23 out of the 33 panels, the Combo test detected infection prior to its detection by the antibody only test.

## Sensitivity to Acute / Early Infection Specimens from Around the World

Sensitivity of the Determine® Combo to acute/primary HIV Infection was evaluated by testing pre and per- seroconversion specimens from various geographical areas. All those specimens tested negative by standard 3<sup>rd</sup> generation EIA's. The results are summarized in the following table:

Country	Number of Specimens	Positive by Determine® Combo	Sensitivity (%)
Belgium	10	8	80.00
UK	18	16	88.89
Thailand	7	2	28.57
France	81	81	100.00
Colombia	1	1	100.00
<b>Total</b>	<b>117</b>	<b>108</b>	<b>92.31</b>

## Conclusions

The Determine® HIV-1/2 Ag/Ab Combo is the first rapid 4<sup>th</sup> generation test exhibiting a high level of performance comparable to 4<sup>th</sup> generation commercial ELISAs. Determine® HIV-1/2 Ag/Ab Combo is a milestone in rapid HIV screening allowing for shortening of the "diagnostic window" to detect pre- and per-seroconversion phases of HIV infection, differential determination of the status of each marker during the window period and use in HIV rapid testing algorithms in areas with limited laboratory infrastructure.