Evaluation of the Bio-Rad Genscreen HIV Ag-Ab ULTRA

Introduction

The Bio-Rad Genscreen HIV Ag-Ab ULTRA assay is an enzyme immunoassay for the detection of HIV p24 antigen and antibodies to HIV1 (groups M and O) and HIV2 in human serum or plasma.

Methods

Specificity

Four thousand eight hundred and ninety two specimens from a single blood donor population and 350 specimens from two presumed antibody status negative populations presenting at separate diagnostic laboratories, were tested on the Bio-Rad Genscreen HIV Ag-Ag ULTRA assay. All samples initially reactive were retested in duplicate. Samples repeatedly reactive were tested in the NRL HIV-1/2 confirmatory algorithm to determine the true status. Specimens that were confirmed as HIV antigen and/or antibody positive were excluded from the specificity and negative delta calculations.

Sensitivity

A total of 285 confirmed anti-HIV positive specimens were tested on the Bio-Rad Genscreen HIV Ag-Ab ULTRA assay: 245 specimens were anti-HIV-1 positive and 40 specimens were anti-HIV-2 positive. Sensitivity and positive delta values were calculated for the overall results for the anti HIV positive samples; positive delta values were also calculated separately for the anti HIV-1 and anti HIV-2 positive samples.

A total of 74 specimens, collected as a series of sequential samples from seven individuals undergoing HIV seroconversion were also tested. The results of testing these seven seroconversion panels on the Bio-Rad Genscreen HIV Ag-Ab ULTRA assay were compared with the results of testing the same panels on the benchmark assay (BMA). The mean difference in detecting a reactive sample was determined

False Reactivity

Forty-three specimens that had given falsely reactive results on previously evaluated anti-HIV assays were tested on the Bio-Rad Genscreen HIV Ag-Ab ULTRA assay. Data from testing these samples were used to determine the degree of common false reactivity between the Bio-Rad Genscreen HIV Ag-Ab ULTRA assay and these other currently registered anti-HIV assays.

Reproducibility

A QC sample of appropriate antibody reactivity and a QC sample of appropriate p24 antigen reactivity for the Bio-Rad Genscreen HIV Ag-Ab ULTRA assay were selected and tested at prescribed intervals. A total of 150 replicates of the antibody QC sample and 127 replicates of the p24 antigen QC sample were tested.

Reproducibility was separately assessed for antigen and antibody by determining mean, standard deviation (S.D.) and coefficient of variation (CV). Outlying results were identified using Grubb's test.

Results

Specificity

Blood Donor Samples

Five of 4892 specimens from the blood donor population were repeatedly reactive on the Bio-Rad Genscreen HIV Ag-Ab ULTRA assay. None of the repeat reactors was confirmed positive. This gave an estimated specificity of 99.90% and a negative delta value of 4.7.

Table 1: Results of testing anti-HIV negative blood donor samples on the Bio-Rad Genscreen HIV Ag-Ab ULTRA assay (with corresponding specificity and delta values).

Samples Tested (n)	Initial Reactors (n)	Repeat Reactors (n)	
		True Positives	False Positives
4892	**59**	**0**	**5**

Specificity	**99.90% (95%CI 99.75% - 99.96%)**
δ value	**– 4.72**

Anti-HIV Negative Diagnostic Samples

Six of 350 specimens from two presumed anti-HIV negative populations were repeatedly reactive on the Bio-Rad Genscreen HIV Ag-Ab ULTRA assay. None of the repeat reactors was confirmed positive. This gave an estimated specificity of 98.29% and a negative delta value of 3.1.

Table 2: Results of testing presumed anti-HIV negative diagnostic samples on the Bio-Rad Genscreen HIV Ag-Ab ULTRA assay (with corresponding specificity and delta values).

Laboratory ID	Samples Tested (n)	Initial Reactors (n)	Repeat Reactors (n)	
			True Positives	False Positives
1	147	2	0	2
2	203	4	0	4
TOTAL	**350**	**6**	**0**	**6**

Specificity	**98.29% (95% CI 96.12 – 99.30%)**
δ value	**– 3.1**

Sensitivity

Confirmed anti HIV Positive Samples

All 245 confirmed anti-HIV-1 positive samples and all 40 anti-HIV-2 positive samples were reactive on the Bio-Rad Genscreen HIV Ag-Ab ULTRA assay. This gave an estimated sensitivity of 100% and positive delta values of 13.0 for HIV-1, 14.8 for HIV-2 and an overall positive delta value of 12.9.

Table 3: Results of testing anti-HIV-1 and anti HIV-2 positive samples on the Bio-Rad Genscreen HIV Ag-Ab ULTRA assay (with corresponding sensitivity and delta values).

Type of Sample	Number of Positive Samples Tested	Number Correct	δ value
anti HIV-1 Positive	245	245	13.0
anti HIV-2 Positive	40	40	14.8
TOTAL:	**285**	**285**	**12.9**

Sensitivity	**100% (95%CI 98.3% - 100%)**
δ value	**+12.9**

Seroconversion Samples

In all seven seroconversion panels the Bio-Rad Genscreen HIV Ag-Ab ULTRA assay detected reactivity before the BMA. The mean difference in seroconversion sensitivity was — 2.57 samples (range — 1 to — 5 samples).

False Reactivity

Thirty nine of 43 samples falsely reactive on other previously evaluated assays were non-reactive on the Bio-Rad Genscreen HIV Ag-Ab ULTRA assay. One of five samples falsely reactive on the Bio-Rad Genscreen HIV-1/2 EIA, one of 26 samples falsely reactive on the Abbott Prism HIV-1/2 ChLIA, one of seven samples falsely reactive on the Abbott Murex HIV Ag/Ab Combination EIA and the one sample falsely reactive on the Bio-Rad Genscreen Plus HIV Ag/Ab EIA were also falsely reactive on the Bio-Rad Genscreen HIV Ag-Ab ULTRA assay.

Table 4: Results of testing falsely reactive samples on the Bio-Rad Genscreen HIV Ag-Ab ULTRA assay.

Assay Producing False Reactivity	Bio-Rad Genscreen HIV Ag-Ab ULTRA Assay		Samples Showing Common False Reactivity (n)
	Samples Tested (n)	Samples Non-reactive (n)	
Bio-Rad Genscreen HIV-1/2 EIA	5	4	1
Bio-Rad Genscreen Plus Ag/Ab EIA	1	0	1
Abbott Prism HIV-1/2 ChLIA	26	25	1
Abbott Murex HIV Ag/Ab Combination EIA	7	6	1
Abbott ARCHITECT HIV Ag/Ab Combo	4	4	0
TOTAL	**43**	**39**	**4**

Reproducibility

Combined, analysed results from a total of 150 replicates in 73 test runs for the antibody QC sample Pelispy Type 38 from the two testing sites demonstrated a coefficient of variation of < 20%.
Combined, analysed results from a total of 127 replicates from 65 test runs for the antigen QC sample p24 AA 03dil from the two testing sites also demonstrated a coefficient of variation of < 20%.

Table 5: Results of testing the antibody QC sample Pelispy Type 38 and the antigen QC sample p24 AA 03dil on the Bio-Rad Genscreen HIV Ag-Ab ULTRA assay.

QC Sample	Test Runs (n)	Observations (n)	Mean (S/Co)	S.D.	C.V. (%)
Pelispy Type 38 (Ab)	73	150	2.71	0.45	16.45
p24 AA 03dil (Ag)	65	127	2.01	0.27	13.51

The Bio-Rad Genscreen HIV Ag-Ab ULTRA assay was recommended for registration on the Australian Register of Therapeutic Goods.

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