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Short communication

Performance evaluation of the re-standardized Abbott Architect anti-HBs assay

Ann H. Wong, Yolanda I.I. Ho*, Raymond W.M. Lai

Department of Microbiology, Prince of Wales Hospital, Hong Kong, China

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ABSTRACT

Background: As defined by World Health Organization (WHO), an antibody level of ≥ 10mIU/mL to hepatitis B virus confers protection. With the launching of Abbott anti-HBs assay re-standardized to the 2nd WHO International Reference Preparation, a positive bias in antibody level would be anticipated. Manufacturer provides limited data for samples around the immune cut-off which has potential implication on vaccine guidance.
Objectives: To evaluate the performance of the re-standardized Abbott Architect anti-HBs assay and to determine the impact of the upward shift.
Study design: A total of 52 samples, including 12 external quality assurance programme samples and 40 clinical samples were tested with both the Abbott 1st WHO standardized and the 2nd WHO re-standardized assay and results compared. The 2nd WHO anti-HBs standard and Acometrix anti-HBs control were also included for comparison.
Results: Verification of the re-standardized assay with the 2nd WHO anti-HBs standard revealed positive bias with mean closer to target value. Overall, the positive bias introduced by the new assay will only affect interpretation of samples with anti-HBs levels > 5.00 to < 10.00 mIU/mL previously tested on the Abbott 1st WHO standardized anti-HBs assay.
Conclusions: Final interpretation of immune status to hepatitis B was not affected by the upward shift following introduction of the new Abbott anti-HBs assay except for previously negative samples with anti-HBs levels between > 5.00 to < 10.00 mIU/mL.

1. Background

Antibody response to hepatitis B surface antigens (anti-HBs) is an important serological marker for sero-conversion or sero-protection resulting from natural infection or vaccination. Anti-HBs level is expressed in International Units (IU) per mL. A cut-off titre of 10 mIU/ml confers immune protection as recommended by WHO [1]. In some countries particularly those with low disease prevalence, a higher antibody level i.e. ≥ 100 mIU/mL detected 4 to 8 weeks following a three or four doses vaccine series is considered as immune [2,3].

The first International Standard (1st IS) for hepatitis B immunoglobulin was established in 1977 [4]. Despite its wide application for calibration of commercial assays, great variation in quantitated levels has been reported with different commercial assays [2,3,5]. Such phenomenon could be attributed to difference in virus surface antigen (HBsAg) employed in different assays, problems with low-avidity antibodies, and difference in vaccine antigen used [2,6].

In our laboratory, the Architect Abbott anti-HBs assay is used for testing anti-HBs in patient sera. In January 2018, Abbott announced that a new anti-HBs assay re-standardized to the 2nd WHO International Reference Preparation, 2008 (Code 07/164) will be launched to replace the 1st WHO standardized assay. A six-point calibration targeted at 0, 10, 50, 100, 500 and 1000 mIU/mL will replace the existing two-point calibration adjustment method. According to the manufacturer, there would be a known positive bias when the 2nd WHO International Standard is tested on the Architect anti-HBs assay [7] such that an upward shift in concentration values would be expected following the conversion. Although linearity and results of 23 spiked samples with concentrations across the measuring range of the assay have been provided by the manufacturer, majority of the spiked samples (17 out of 23) were at anti-HBs level > 100 mIU/mL. Only limited data is available for samples around the immune cut-off level of 10 mIU/mL, thus it has potential implication on guidance of vaccination.

* Corresponding author.
E-mail address: hii801@ha.org.hk (Y.I.I. Ho).

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2. Objectives and study design

The aim of this study was to evaluate the performance of the re-standardized Abbott Architect anti-HBs assay to assess the clinical impact of the anticipated shift. Both external quality assurance programme (EQAP) samples from the Royal College of Pathologists Australasia (RCPA) and clinical samples were included in this study. The EQAP samples (n = 12) were from the years 2016 to 2018 while the clinical samples (n = 40) were from the years 2017 to 2018.

All these samples which had been tested with the 1st WHO standardized anti-HBs assay were retrieved from storage at −70 °C and tested with the re-standardized anti-HBs assay. Linear regression analysis and Bland-Altman difference plot were performed with Microsoft Excel 2010. Precision of the re-standardized assay was assessed by testing the AcroMetrix anti-HBs control (lot number 173424) (Thermo Scientific, USA) five times on four separate runs on different days. The manufacturer claimed that the anti-HBs level of this control material tested with the Abbott Architect AUSAB assay was 13 mIU/mL. Accuracy of the re-standardized assay was assessed with the 2nd WHO anti-HBs Standard diluted to 10 mIU/mL and 100 mIU/mL with the Acrometrix Ultra serum dilution matrix (Thermo Scientific, USA).

3. Results

As compared to the 1st WHO standardized assay, an upward shift in quantitated values was observed for the 8 EQAP samples with antibody levels above the immune cut-off value of 10 mIU/mL (Table 1). Nonetheless, the final interpreted results were not affected (Table 1).

For clinical samples, linear regression analysis of the anti-HBs levels quantitated by both assays showed good correlation ($R^2 = 0.9806$). Again, an upward shift in the anti-HBs levels was observed for all samples tested (Fig. 1).

Bland-Altman difference plot showed that the new assay had a positive mean bias of 25.04 mIU/mL and the bias increased with the quantitated level (Fig. 2).

Based on the in-use Architect anti-HBs assay quantitative results, the clinical samples were categorized into 3 major categories with category 1 = 0.00–5.00 mIU/mL; category 2 = > 5.00 to < 10.00 mIU/mL; and category 3 = ≥10.00 to ≥1000.00 mIU/mL. Results of these 3 categories were shown in Table 2. No discrepancy in immune status interpretation was observed for category 1 and 3 samples (Table 2). For category 2, 11 out of 12 (91.7%) tested previously as non-immune will be interpreted as immune when retested with the 2nd WHO re-standardized anti-HBs assay (Table 2).

Table 1
Anti-HBs levels of RCPA EQAP samples from years 2016 to 2018 measured with Architect 1st WHO standardized anti-HBs assay and Architect 2nd WHO re-standardized anti-HBs assay.

RCPA		Architect 1st WHO standardized anti-HBs assay		Architect 2nd WHO re-standardized anti-HBs assay	
Specimen	Consensus result	anti-HBs level (mIU/mL)	Interpreted result	anti-HBs level (mIU/mL)	Interpreted result
H1:2016:1C	immune	> 1000.0	Immune	> 1000.00	Immune
H3:2016:3D	immune	599.18	Immune	> 1000.00	Immune
H4:2016:4D	immune	40.96	Immune	62.11	Immune
H5:2016:5D	non-immune	0.10	non-immune	0.00	non-immune
H6:2016:6C	immune	15.44	Immune	25.32	Immune
H1:2017:1C	immune	622.99	Immune	> 1000.00	Immune
H2:2017:2C	non-immune	0.00	non-immune	0.00	non-immune
H2:2017:2D	non-immune	0.11	non-immune	0.00	non-immune
H3:2017:3D	immune	382.17	Immune	700.62	Immune
H5:2017:5C	immune	99.69	Immune	185.55	Immune
H1:2018:1C	immune	94.09	Immune	164.50	Immune
H1:2018:1D	non-immune	0.06	non-immune	0.00	non-immune

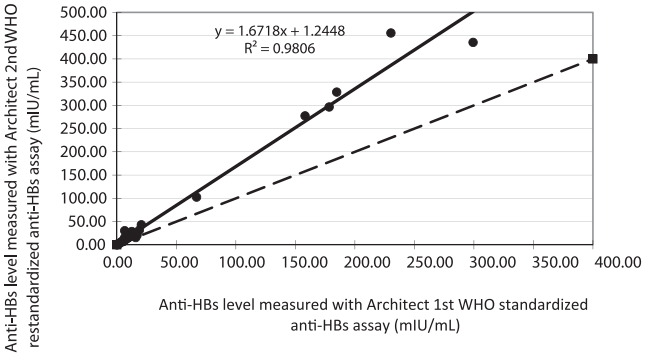

Fig. 1. Graph of Architect 1st WHO standardized anti-HBs assay and Architect 2nd WHO re-standardized anti-HBs assay results for clinical samples (solid line). Dashed line represented expected results with no bias.

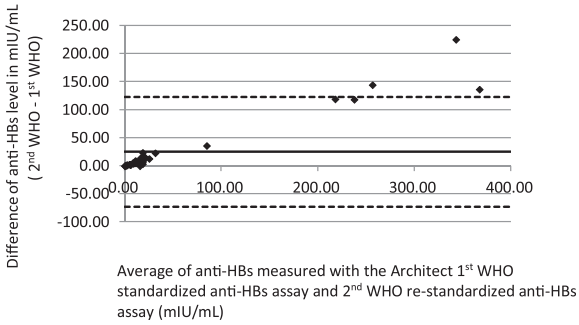

Fig. 2. Bland-Altman plot showing average differences between the Architect 1st WHO standardized anti-HBs assay and Architect 2nd WHO re-standardized anti-HBs assay.

To elucidate the impact of positive bias in the new anti-HBs assay, the 2nd WHO anti-HBs Standard was diluted with AcroMetrix Ultra serum dilution matrix to 10 mIU/mL and 100 mIU/mL and tested in duplicates in two separate runs on different days with both assays. The mean percentage (%) bias of both assays were -20.63 and 13.92 respectively when tested against the target value of 10 mIU/mL, and -21.63 and 15.10 respectively for the target value of 100 mIU/mL (Table 3). The new assay produced results closer to the target values. Thus, it explained the discrepancy in result interpretation for category 2 samples with tested value span around the immune cut-off.

Precision of the assay was assessed with the AcroMetrix control. The mean anti-HBs level quantitated by the new assay was 18.65 mIU/mL (standard deviation = 0.16 mIU/mL and coefficient of variation = 0.85%). This fulfilled the criteria of < 10% set by the manufacturer.

4. Discussion

Quantification of anti-HBs level is crucial in clinical setting as it determines whether an individual has protection to hepatitis B infection. It provides guidance on whether additional doses of vaccine are required for hypo-responders or non-responders. Currently, there is no gold standard for measurement of anti-HBs levels. The report by Huzly et al [2] showed great variation in commercially available assays for quantification of anti-HBs. Our study illustrated this with the AcroMetrix Anti-HBs control which has been calibrated against the 2nd WHO IS. This control material has a claimed value of 13 mIU/mL on the Abbott Architect AUSAB assay. Testing with the Architect 2nd WHO re-standardized anti-HBs assay, however, yielded a value of 18.62 mIU/mL.

Table 2
Anti-HBs levels in categories 1–3 clinical samples measured with Architect 1st WHO standardized anti-HBs assay and Architect 2nd WHO re-standardized anti-HBs assay.

		Architect 1st WHO standardized Anti-HBs assay		Architect 2nd WHO re-standardized Anti-HBs assay	
Category	Sample no	Anti-HBs level (mIU/mL)	Interpreted result	Anti-HBs level (mIU/mL)	interpreted result
1	1	3.70	non-immune	6.15	non-immune
	2	1.13	non-immune	1.45	non-immune
	3	0.18	non-immune	0.67	non-immune
	4	4.52	non-immune	7.36	non-immune
	5	2.28	non-immune	3.05	non-immune
	6	1.39	non-immune	3.02	non-immune
	7	4.87	non-immune	6.99	non-immune
	8	0.09	non-immune	0.00	non-immune
	9	0.18	non-immune	0.00	non-immune
2	10	7.21	Non-immune	10.97	immune
	11	8.91	Non-immune	13.93	immune
	12	5.50	Non-immune	6.95	Non-immmune
	13	9.43	Non-immune	16.09	immune
	14	7.07	Non-immune	11.77	immune
	15	5.96	Non-immune	11.20	immune
	16	7.23	Non-immune	13.66	immune
	17	8.97	Non-immune	14.32	immune
	18	6.68	Non-immune	15.63	immune
	19	9.25	Non-immune	15.97	immune
	20	6.59	Non-immune	30.24	immune
	21	9.21	Non-immune	23.61	immune
3	22	15.83	Immune	15.53	Immune
	23	14.20	Immune	22.90	Immune
	24	11.85	Immune	20.13	Immune
	25	12.51	Immune	20.43	Immune
	26	10.63	Immune	16.53	Immune
	27	20.52	Immune	42.94	Immune
	28	230.30	Immune	455.68	Immune
	29	178.42	Immune	296.53	Immune
	30	184.69	Immune	328.50	Immune
	31	158.17	Immune	277.31	Immune
	32	18.90	Immune	31.57	Immune
	33	14.04	Immune	26.10	Immune
	34	12.48	Immune	28.41	Immune
	35	16.97	Immune	20.52	Immune
	36	13.68	Immune	22.52	Immune
	37	11.78	Immune	19.37	Immune
	38	66.93	Immune	102.58	Immune
	39	15.25	Immune	22.21	Immune
	40	299.29	Immune	435.55	Immune

Table 3
Testing of 2nd WHO anti-HBs at 10 and 100 mIU/mL with the Architect 1st WHO standardized anti-HBs assay and the Architect 2nd WHO re-standardized anti-HBs assay.

2nd WHO anti-HBs titre	Architect 1st WHO standardized anti-HBs assay		Architect 2nd WHO re-standardized anti-HBs assay	
	mIU/mL	%bias	mIU/mL	%bias
10 mIU/mL	7.59	−24.1	11.14	11.40
	8.23	−17.7	11.80	11.80
	7.92	−20.8	11.46	11.46
	8.01	−19.9	12.10	21.00
	Mean = 7.94	Mean = −20.63	Mean = 11.63	Mean = 13.92
100 mIU/mL	74.63	−25.37	112.35	12.35
	81.46	−18.54	117.03	17.03
	76.95	−23.05	114.40	14.40
	80.43	−19.57	116.62	16.62
	Mean = 78.37	Mean = −21.63	Mean = 115.10	Mean = 15.10

Kinn et al compared the performance of the Diasorin LIASION anti-HBs II for the detection of anti-HBs with the Abbott Architect anti-HBs assay and the overall qualitative agreement was fairly satisfactory. Interestingly, 5 samples with distinct discrepancy in that study had values close to the Abbott threshold (7.19–9.68 mIU/mL). When tested with the DiaSorin, it produced results just above the immune cut-off (11.5–19.2 mIU/mL) [8]. The imperfect calibration of the Abbott assay to the WHO standard rather than the lack of sensitivity and specificity was considered by Kinn et al as an explanation for such discrepancy [8]. Recent study by Raven et al reported that when different commercial systems were assessed in quality assessment program using samples at 10 and 100 mIU/mL, the Architect assay has a tendency to give false negative results at 100 mIU/mL [9] which echoes the findings in this study. Thus, previous false negative results with anti-HBs level between > 5.00 to < 10.00 mIU/mL will be resolved with the Architect 2nd WHO re-standardized anti-HBs assay.

5. Conclusion

In conclusion, the Architect 2nd WHO re-standardized anti-HBs assay revealed an upward shift in antibody level as compared with the old Architect assay. Overall interpretation of final immune status was not affected with the introduction of the new assay except for those samples with antibody levels between > 5.00 to < 10.00 mIU/mL.

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Ethical approval

Anonymized samples were tested and ethical approval was not required.

Conflict of interest

The authors declare that we have no conflict of interest.

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References

[1] A.D. Jack, A.J. Hall, N. Maine, et al., What level of hepatitis B antibody is protective? J. Infect. Dis. 179 (1999) 489–492.
[2] D. Huzly, T. Schenk, W. Jilg, et al., Comparison of nine commercially available assays for quantification of antibody response to hepatitis B virus surface antigen, J. Clin. Microbiol. 46 (2008) 1298–1306.
[3] SchryverA. De, B. Claesen, A. Meheus, et al., European survey of hepatitis B vaccination policies for healthcare workers, Eur. J. Public Health 21 (2011) 338–343.
[4] World Health Organization, Anti-hepatitis B Immunoglobulin 626 WHO Tech Resp Ser, 1978, p. 18.
[5] L. Cheng, Q. Guan, J. Zhang, et al., Discrepancies between two automated immunoassay systems in determining hepatitis B virus markers in serum samples with concomitant presence of antigens and antibodies, Ann. Clin. Lab. Sci. 40 (2010) 49–52.
[6] R.A. Heijtink, P.M. Schneeberger, B. Postman, et al., Anti-HBs levels after hepatitis B immunisation depend on test reagents: routinely determined 10 and 100 IU/l seroprotection levels unreliable, Vaccine 20 (2002) 2899–2905.
[7] Abbott product information, P I1004-2017.
[8] S. Kinn, S. Akhavan, H. Agut, et al., Performance of the Diasorin LIASION® anti-HBsII for the detection of hepatitis B surface antibodies in comparison with the Abbott Architect anti-HBs assay, J. Clin. Virol. (2011) 297–302.
[9] S. Raven, J. Hautvast, J.V. Steenbergen, et al., Diagnostic performance of serological assays for anti-HBs testing: results from a quality assessment program, J. Clin. Virol. 87 (2017) 17–22.