Evaluation of a novel HBV DNA test from fingerstick capillary blood at the point-of-care as a tool to enhance clinical management

Behzad Hajarizadeh[1], Jacob George[2], Miriam T Levy[3], Ian Wong[4], Jess Howell[5], Gesalit Cabrera[1], Elise Tu[1], Marianne Martinello[1,6], Tanya L Applegate[1]*, Gail V Matthews[1,7]*

1. The Kirby Institute, University of New South Wales, Sydney; 2. Storr Liver Centre, Westmead Hospital & The Westmead Institute for Medical Research, University of Sydney, Sydney; 3. Liverpool Hospital & South-Western Clinical School, University of New South Wales, Sydney; 4. Blacktown Hospital, Sydney; 5 St Vincent's Hospital & the Burnet Institute, Melbourne; 6. Prince of Wales Hospital, Sydney; 7. St Vincent's Hospital, Sydney
*T Applegate and G Matthews are joint senior authors

Introduction

HBV viral load tests are essential to guide hepatitis B antiviral treatment eligibility and effectiveness.

Many people are unable to access standard-of-care tests, particularly those in remote areas or resource-limited settings given high cost, or limited infrastructure.

The GeneXpert® Systems - the most commonly used molecular point-of-care platform globally - have the potential to offer simple and affordable HBV viral load tests.

However, current Xpert® HBV Viral Load tests require phlebotomy and sample centrifugation, limiting its potential to be conducted at the point-of-care in primary health care and other decentralized settings.

Aims

To evaluate diagnostic performance of the point-of-care Xpert® HBV Viral Load assay, using fingerstick capillary blood samples, compared to standard-of-care assay using venous blood plasma samples.

Methods

Study participants & setting: Individuals diagnosed with chronic HBV infection (HBsAg positive) who were ≥18 years old were eligible, irrespective of their HBV viral load, HBeAg or treatment status. Participants were enrolled from six hospital-based liver disease clinics in Australia.

Study procedures: After informed consent, two blood samples were collected from each participant.

- Fingerstick capillary blood (100µL): Tested using Xpert® HBV Viral Load assay (lower limit of quantification, 10 IU/mL).
- Venepuncture whole blood (600µL plasma): Tested using COBAS® AmpliPrep/COBAS® TaqMan® HBV DNA Test (standard-of-care)

Analysis: Fingerstick samples were diluted 1 in 10 for testing. The original HBV viral load reads on Xpert® were multiplied by 10 and the adjusted values were used in the analysis. Given the adjustment and to enable comparative analyses, the lower limit of quantification for adjusted viral load was changed to 100 IU/mL and was used for sensitivity and specificity analysis.

The positive and negative agreement of the Xpert® for HBV viral load ≥100 IU/mL (vs. <100 IU/mL or undetectable) were evaluated and compared to the standard-of-care. Agreement measurements of assays were assessed using Bland–Altman bias plot.

This study has been approved by the St Vincent's Hospital Sydney Human Research Ethics Committee (2021/ETH11051).

Results

To date, **178 participants** have been enrolled (target n=250)

Table 1: Baseline participants' characteristics

Gender	Male	96 (54%)
	Female	82 (46%)
Age, median (IQR), years		46 (39-59)
Current HBV clinical management	Initial assessment	24 (13%)
	Monitoring only	76 (43%)
	Treatment	78 (44%)
HBeAg	Negative	141 (79%)
	Positive	33 (19%)
	Unknown	4 (2%)
Cirrhosis	No	108 (90%)
	Yes	11 (6%)
	Unknown	7 (4%)

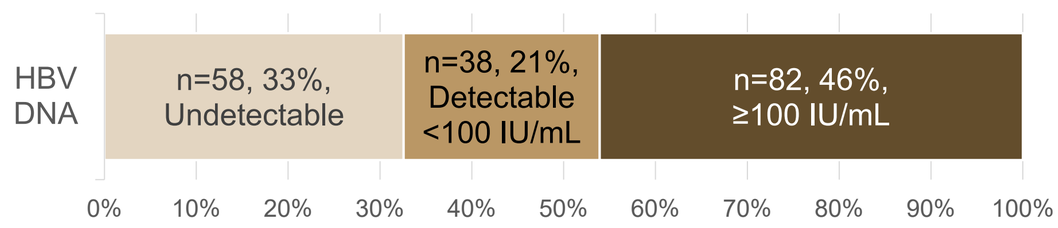

Figure 1: HBV viral load by the standard-of-care assay

Table 2: Positive and negative agreement of HBV viral loads measured by the Xpert® using fingerstick capillary blood sample and standard-of-care assay

		HBV viral load measured by standard-of-care assay on 600µL plasma		
		≥100 IU/mL	<100 IU/mL*	Total
HBV viral load measured by Xpert® on 100µL capillary blood	≥100 IU/mL	79	10	89
	<100 IU/mL*	3	86	89
	Total	82	96	**178**

Among all participants (n=178), 82 had HBV viral load ≥100 IU/mL, detected by the standard-of-care. Of those participants, **the Xpert® detected similar HBV viral load ≥100 IU/mL in 96.3% (95%CI: 93.6%, 99.1%)**.

Among participants who received treatment (n=78), 67 had HBV viral load <100 IU/mL or undetectable, by the standard-of-care. Of those participants, **the Xpert® detected similar HBV viral load <100 IU/mL or undetectable in 93.1% (95%CI: 87.4%, 98.7%).**

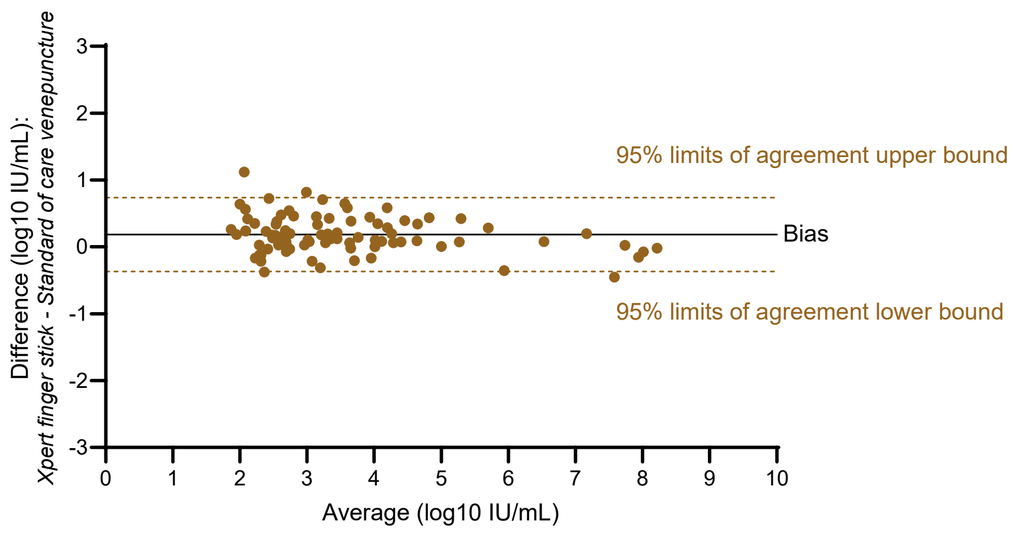

Figure 2: Bland–Altman bias plot of differences between HBV viral loads measured by the Xpert® and standard-of-care assays

Bias=0.18 log; SD=0.28 log; 95% limits of agreement=-0.37 to 0.74

The plot indicated that the viral load detected by the Xpert® were a mean of 0.18 log higher than those detected by the standard-of-care, while 95% of the differences between viral load detected by two assays were between −0.37 and 0.74 log.

Table 3: HBV viral load (IU/mL) of 13 participants with non-concordant results by two assays

ID	Standard-of-care viral load	Xpert® viral load	HBV clinical care status
61202-035	198	<100	Treatment
61202-041	115	<100	Monitoring only
61210-012	115	<100	Initial assessment
61210-028	63	230	Initial assessment
61216-007	93	160	Treatment
61216-027	48	210	Monitoring only
61216-042	80	210	Treatment
61216-043	55	100	Monitoring only
61216-045	72	110	Treatment
61216-048	92	160	Monitoring only
61216-056	32	420	Treatment
61202-034	<20	260	Monitoring only
61216-005	<20	150	Treatment

Limitation:
The sample dilution needed in this pilot study prevented a comparison of agreement with the standard-of-care assay using registered lower limits of quantitation and required using a minimum threshold of 100 IU/mL to equalise the comparison.

Conclusion

Minimal difference was observed in HBV viral load detection between Xpert® using fingerstick capillary blood and standard-of-care assays using venous plasma.

Among participants with discordant results, the difference was not sufficient to impact clinical decisions.

These results support development of a dedicated Xpert® HBV Viral load fingerstick assay to simplify HBV clinical care, including for treatment decision making in pregnancy and in remote and resource-limited settings.

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Contacts:
For more information about the study, please contact: Behzad Hajarizadeh (bhajarizadeh@kirby.unsw.edu.au), Tanya Applegate (tapplegate@kirby.unsw.edu.au), or Gail Matthews (gmatthews@kirby.unsw.edu.au)