AgaMatrix White Paper

Performance Evaluation of the A1Care HbA1c Analyser

An Independent UK Study

Contents

Introduction

The measurement of the glycated fraction of haemoglobin (HbA1c) has long been recommended to diagnose diabetes, in addition to monitoring glycaemic control. HbA1c reflects long-term glycaemic control over the preceding 2-3 months as opposed to glycaemic control at a single point in time e.g., fasting plasma glucose. HbA1c concentration ≥48 mmol/mol (≥6.5%) is the accepted criteria for diabetes diagnosis, with 42-47 mmol/mol (6.0-6.5%) the prediabetes range. Point of care testing (POCT) enables measurement of HbA1c in a clinical setting without the waiting time associated with laboratory testing or the high level of expertise required, however for POCT methods to be used effectively, they must be comparable to those used in a laboratory.

The A1Care analyser, manufactured by i-SENS, is a small, CE marked, whole blood, point of care HbA1c analyser. The device is also NGSP and IFCC certified.

Objectives

- To compare the performance of the A1Care POCT HbA1c analyser with a reference HPLC laboratory method (Tosoh GX).
- To compare whole blood venous and capillary HbA1c concentrations measured using the A1Care POCT HbA1c analyser.

Method

Whole blood venous EDTA samples (n=100) from subjects across a wide glycaemic range were assayed using the Tosoh GX HPLC analyser as the laboratory reference method and the A1Care HbA1c POC analyser (i-SENS). Two A1Care HbA1c POC analysers were used, and all 100 samples were measured on each analyser.

In addition, in 30 subjects a 4ml venous blood (EDTA) sample was taken. These samples were measured using the A1Care analyser and a laboratory analyser (Tosoh GX). At the same time, a finger prick sample was also taken and measured on the A1Care analyser.

Data Analysis

Intraclass correlation coefficients for agreement between the HPLC and A1Care devices were calculated to quantify how well HPLC and A1Care devices were related by HPLC and A1Care findings (McGraw & Wong, 1996). The proposed sample of 100 will have more than 80% power to identify a significant intraclass correlation (ICC) at a 0.05 level if scores derived from the 2 devices are moderately reliable and agree moderately well with each other (i.e. ICC greater than or equal to 0.5). ICC values less than 0.5 are indicative of poor reliability, values between 0.5 and 0.75 indicate moderate reliability, values between 0.75 and 0.9 indicate good reliability, and values greater than 0.90 indicate excellent reliability.

Bland–Altman plots and the associated 95% limits of agreement (LoA) were used to identify presence of any systematic and proportional bias between the index tests and the reference standard. LoA are calculated as the mean of differences between two measurements ± 1.96 x their standard deviation (Bland & Altman, 1986).

Variation between the A1Care devices was determined by Bland-Altman plots.

Intra-assay variation was determined by calculating mean±SD and percentage coefficient of variation (%CV) of the samples run in triplicate.

Inter-assay variation was determined by calculating mean±SD and %CV of repeat assaying of quality control samples over 5 consecutive assays.

Sample stability was assessed as percentage of the result on Day 1.

Results

Comparison of A1Care vs Tosoh GX

Comparison of 100 EDTA samples measured on the two A1Care analysers and the Tosoh GX HPLC analyser is shown in Fig. 1.

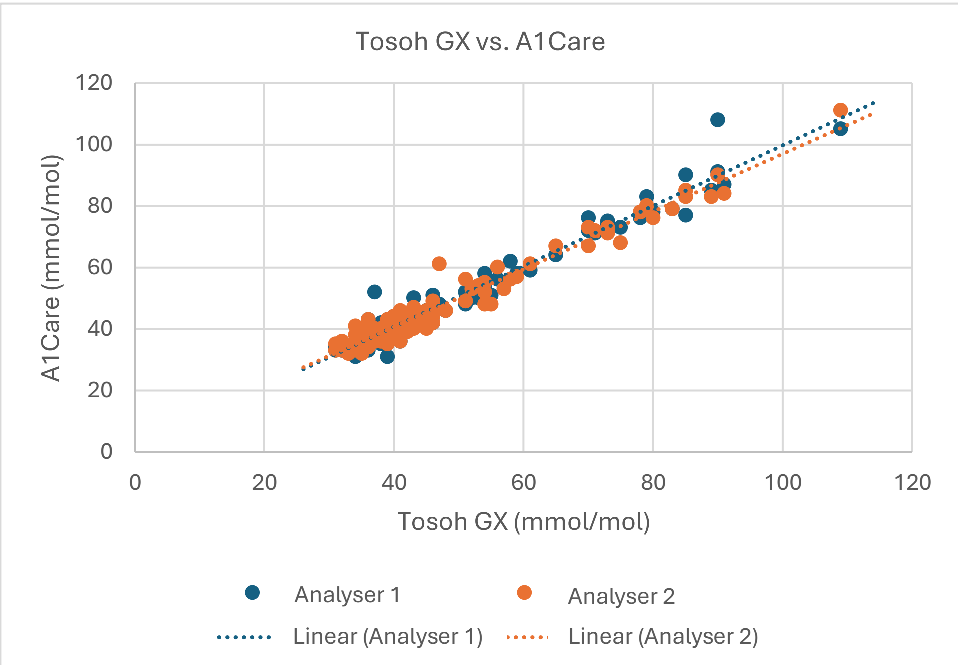

Fig. 1: Comparison of Tosoh GX vs. A1Care Analysers 1 and 2

For Analyser 1 the slope was 0.9831 and the intercept 1.3641. The coefficient of determination (R^2) was 0.9572. Intraclass correlation (95% CI) was 0.977 (0.965, 0.984) P<0.001.

For Analyser 2 the slope was 0.9377 and the intercept 3.1385. The coefficient of determination (R^2) was 0.9629. Intraclass correlation (95% CI) was 0.980 (0.971, 0.987) P<0.001.

The bias (±SD) of the individual A1Care analysers compared to the Tosoh GX reference method was 0.54 (3.63) and 0.14 (3.28) mmol/mol for A1Care analysers 1 and 2, respectively.

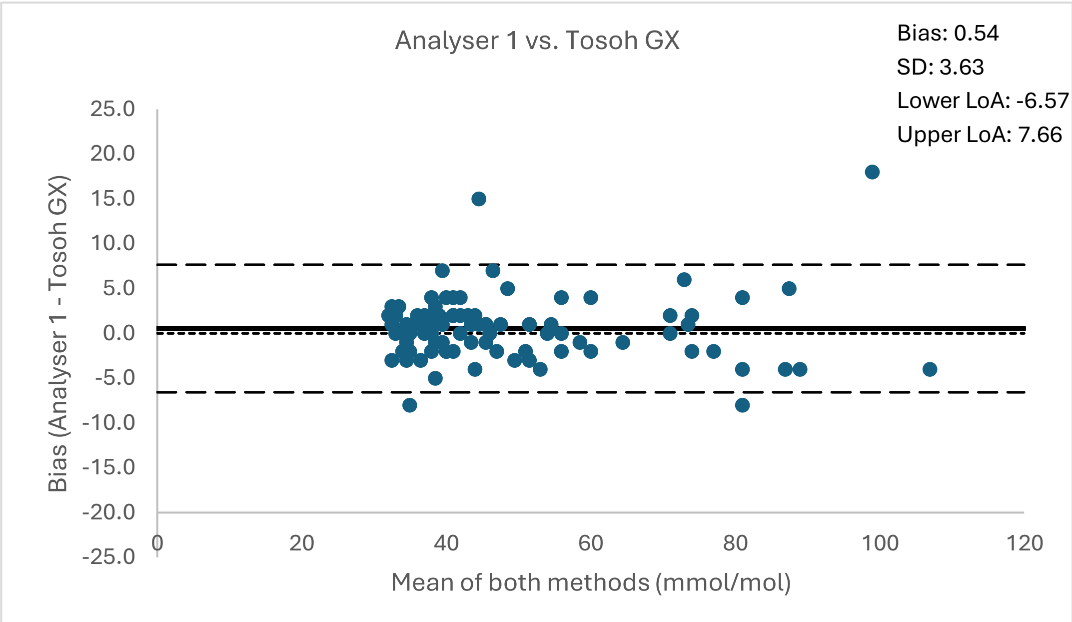

Fig. 2: Bias of Analyser 1

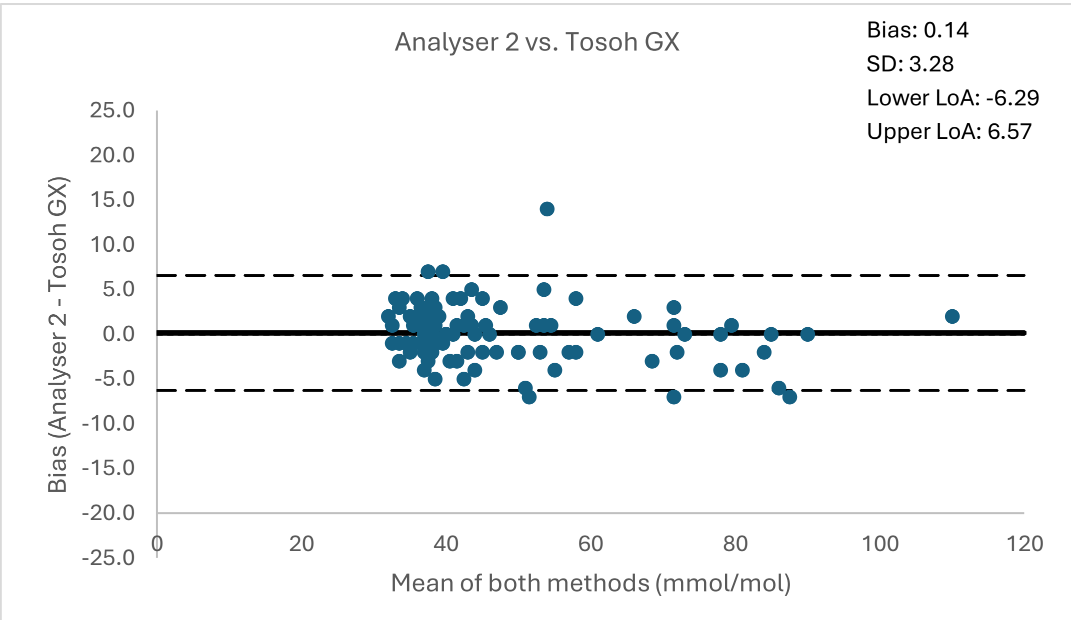

Fig. 3: Bias of Analyser 2

Comparison of the bias of the two A1Care analysers is shown in Fig. 4. The bias between the two A1Care analysers was -0.40 (4.05) mmol/L.

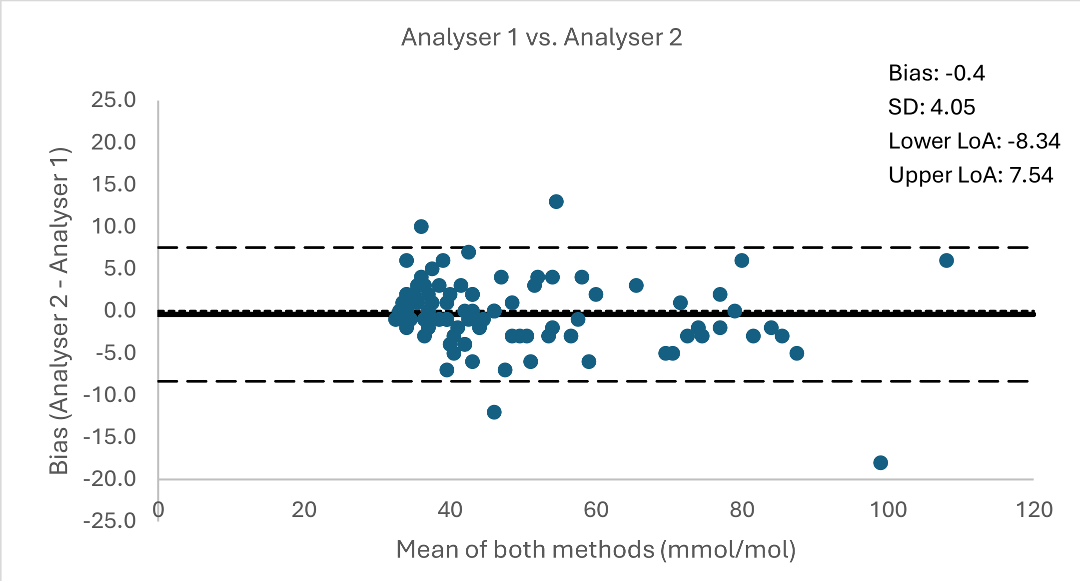

Fig. 4: Comparison of the bias of the two A1Care analysers

Results from the additional 30 subjects studied in the venous versus capillary comparison were added to the 100 EDTA samples to make a total sample size of 130 samples compared with the A1Care analyser (Analyser 1) and the Tosoh GX HPLC analyser. Comparison of original values is shown in Fig. 5, and repeated samples in Fig. 6.

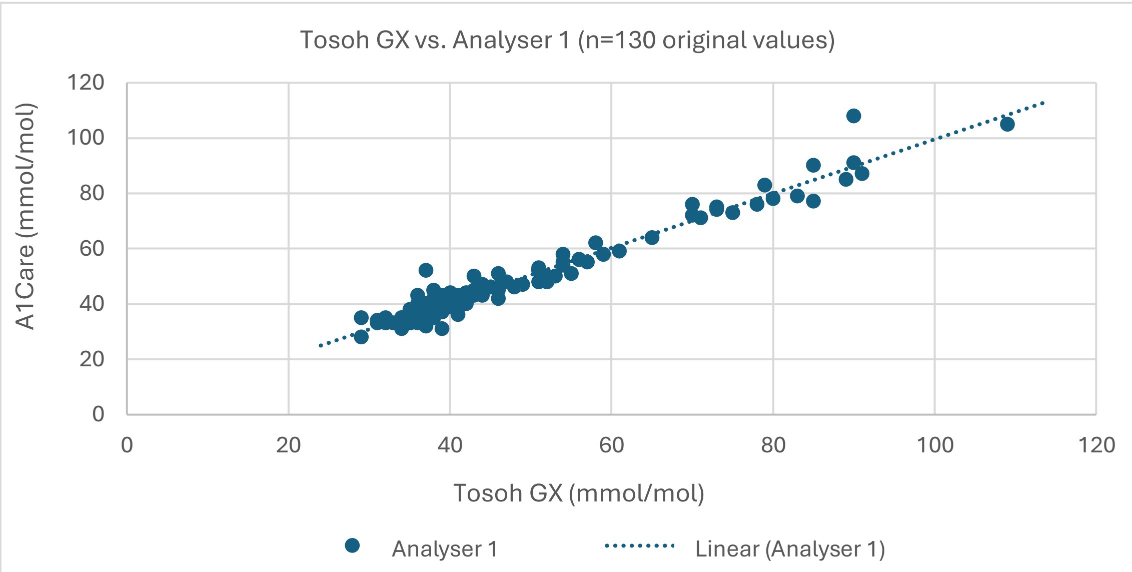

Fig. 5: Comparison of 130 samples analysed using the A1Care Analyser 1 and Tosoh GX HPLC Analyser (Original Values)

The slope was 0.980 and the intercept 1.4455. The coefficient of determination (R^2) was 0.9559. Intraclass correlation (95% CI) was 0.977 (0.968, 0.984) P<0.001.

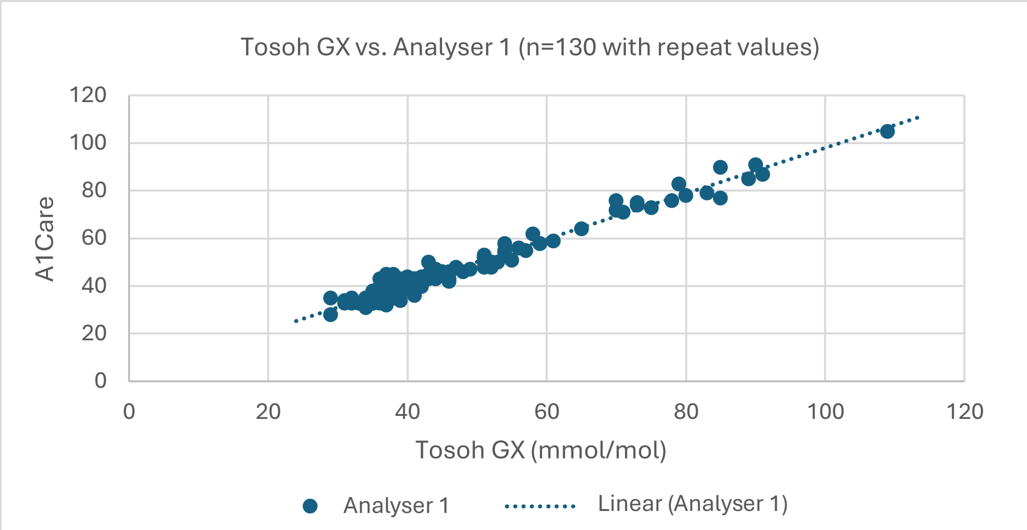

Fig. 6: Comparison of 130 samples analysed using the A1Care Analyser 1 and Tosoh GX HPLC Analyser (Repeat Values)

The slope was 0.9561 and the intercept 2.3473. The coefficient of determination (R^2) was 0.9693.

Comparison of the bias between the A1Care analyser and Tosoh GX HPLC analyser is shown in Fig. 7. The bias (±SD) between the two HbA1c analysers was -0.32 (2.73) mmol/mol.

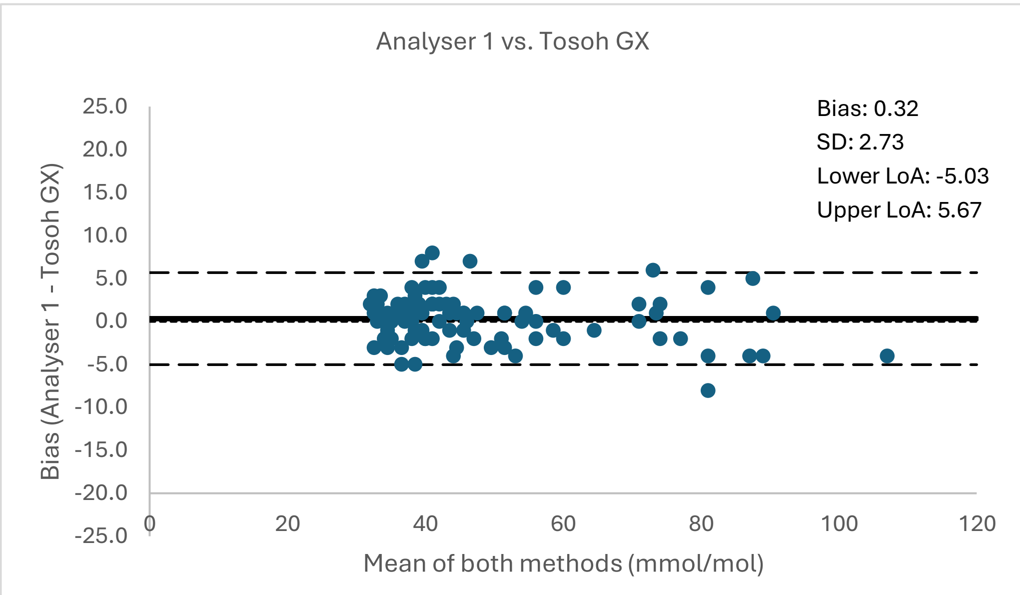

Fig. 7: Comparison of the bias between A1Care Analyser 1 and Tosoh GX HPLC analyser

Intra-Assay Variation

Intra-assay variation was determined in triplicate on two A1Care analysers by measurement of HbA1c in three blood samples (low and elevated concentrations).

Sample	Analyser 1			Analyser 2		
	Mean (mmol/mol)	SD (mmol/mol)	CV2 (%)	Mean (mmol/mol)	SD (mmol/mol)	CV2 (%)
1	33.0	1.00	3.0	31.7	0.58	1.8
2	59.0	1.00	1.7	55.3	2.08	3.8
3	105.0	4.00	3.8	104.0	2.65	2.5

Table 1: Intra-assay variation

Across a wide range of HbA1c concentrations, intra-assay variation was less than 4.0% on both analysers.

Inter-Assay Variation

Inter-assay variation was determined by repeat assaying A1Care quality control samples in 6 consecutive assays. Results are shown in Table 2.

	QC1 (Range 23.5-47.5 mmol/mol)			QC2 (Range 62.8-106.6 mmol/mol)	
	Analyser 1	Analyser 2		Analyser 1	Analyser 2
Mean (mmol/mol)	37.0	36.3		84.5	86.0
SD (mmol/mol)	3.0	2.0		2.1	1.5
CV (%)	8.2	5.4		2.5	1.8
Min (mmol/mol)	33	35		81	84
Max (mmol/mol)	41	40		87	88

Table 2: Inter-assay variation

Inter-assay variation ranged from 1.8% to 8.2%.

Sample Stability

To assess sample stability, aliquots of 3 venous blood samples (low and elevated concentrations) were measured over a period of 2 weeks (days 1, 7 and 14). Results (expressed as a percentage of the results obtained on Day 1 are shown in Tables 3 and 4.

	Sample	Day			Day 7		Day 14	
		1	7	14	Mean	SD	Mean	SD
Room Temp	**1**	100	102.4	96.5	**101.5**	5.9	**93.4**	5.9
	2	100	96.2	86.5				
	3	100	105.9	97.1				
4°C	**1**	100	104.7	104.7	**98.3**	4.2	**102.9**	4.2
	2	100	101.9	98.1				
	3	100	88.2	105.9				

Table 3:Sample stability for Analyser 1

	Sample	Day			Day 7		Day 14	
		1	7	**14**	**Mean**	**SD**	**Mean**	**SD**
Room Temp	**1**	100	100.0	92.8	**101.7**	6.3	**87.0**	6.3
	2	100	92.9	80.4				
	3	100	112.1	87.9				
4°C	**1**	100	106.0	103.6	**99.3**	15.3	**103.1**	15.3
	2	100	85.7	87.5				
	3	100	106.1	118.2				

Table 4:Sample stability for Analyser 2

Results show that samples are stable for 7 days when stored at either room temperature or at 4°C. For storage of samples longer than 7 days and up to 14 days samples would be best stored at 4°C.

Venous vs. Capillary Sampling

In 30 subjects, venous and capillary samples taken simultaneously were analysed. Comparison of results are shown in Fig. 8.

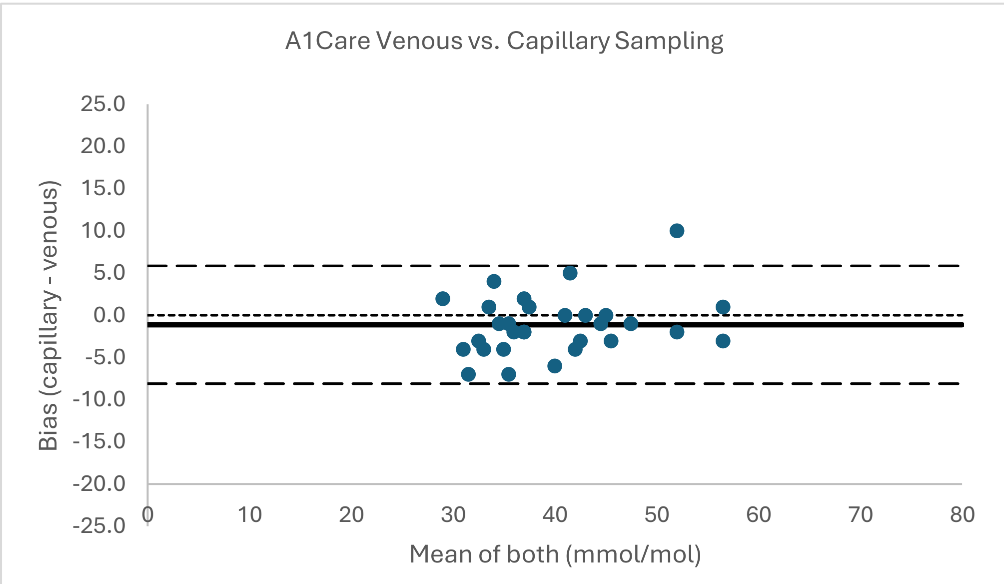

Fig. 8: Comparison of venous and capillary sampling of the A1Care analyser

Intraclass correlation (95% CI) between venous and capillary samples was 0.884 (0.769, 0.943) P<0.001. The bias (±SD) between the venous and capillary samples was -1.13 (3.56) mmol/mol.

Summary

An analysis of the performance of the A1Care point of care HbA1c analyser has been carried out. The results presented in this analysis include correlation and bias plots.

Correlation of 130 samples measured with the A1Care analyser and a laboratory HPLC HbA1c method (Tosoh GX) demonstrated excellent correlation (R^2 = 0.9693; slope 0.9561; intercept 2.3473; ICC 0.977). Bland-Altman analysis showed a bias of -0.32 (2.734) mmol/mol across the range of all HbA1c concentrations. Two A1Care analysers were also used to measure 100 patient samples and both showed excellent correlation with the HPLC HbA1c analyser (R^2 = 0.9572; slope 0.9831; intercept 1.3641; ICC 0.977 and R2 = 0.9629; slope 0.9377; intercept 3.1385; ICC 0.980 for analysers 1 and 2 respectively). Bias between the 2 analysers was small at -0.40 (4.05) mmol/mol.

Intra-assay variation determined in triplicate on two A1Care analysers showed that across a wide range of HbA1c concentrations intra-assay variation was less than 4.0% on both analysers. Inter-assay variation, determined by repeat assaying A1Care quality control samples, showed that inter-assay variation ranged from 1.8 to 8.2%.

Samples were stable for 7 days when stored at either room temperature or at 4°C, but for storage of samples up to 14 days samples would be best stored at 4°C. No difference could be observed between venous and capillary samples.

In summary, the above analysis shows that the A1Care analyser worked well with relatively few failures. Correlation with a laboratory HPLC analyser was excellent with little bias in results. Intra- and inter-assay variation in the A1Care analyser was acceptable. Results showed that there was no difference in results obtained from venous or capillary samples, and venous samples could be stored for up to one week at room temperature without affecting the results. If stored at 4°C sample results were not affected for up to 14 days.

Acknowledgements

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References

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McGraw, K. O., & Wong, S. P. (1996). Forming inferences about some intraclass correlation coefficients. Psychological Methods, 1(1), 30–46.