Comparing HbA1C by POC and HPLC

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ABSTRACT

OBJECTIVE: Point-of-care (POC) Hemoglobin A1C (HbA1C) testing is frequently used to assess glycemic control in diabetes management. Studies are lacking on the comparison of POC with high performance liquid chromatography (HPLC) when the POC HbA1C is ≥ 14%.

METHODS: Retrospective chart review of children with T1DM at Rhode Island Hospital from 2007–2013. Primary objective was to delineate the range of HPLC HbA1C values when the POC is ≥ 14% and characterize these patients.

PRIMARY RESULTS: There were 72 patients, 5–21 years old, with corresponding POC and HPLC tests. Nineteen children, mean age 16.1 years, had a POC HbA1C ≥ 14%. Their mean HPLC value was 14.1% (95% CI [13.4, 14.8]), with range 11.1–16.3 and standard deviation 1.4%.

CONCLUSION: There is wide variation when POC HbA1C values are ≥ 14%. We suggest routine central HbA1C testing when the POC is ≥ 14% for proper counseling and follow-up of glycemic control. Tracking relative changes in HbA1C at subsequent clinic visits is important as it allows clinicians to gauge whether or not interventions are effective. Additionally, knowledge that their HbA1C is trending down may provide positive reinforcement to adolescents.

KEYWORDS: pediatrics, diabetes, Type 1, HbA1C, point-of-care

ABBREVIATIONS: POC = point-of-care, HbA1C = Hemoglobin A1C, HPLC = high performance liquid chromatography, T1DM = type 1 diabetes, NGSP = National Glycohemoglobin Standardization program, ADA = American Diabetes Association, CV = coefficient of variation

INTRODUCTION

The Diabetes Control and Complications Trial (DCCT) showed that good glycemic control is crucial to delay the progression of diabetic retinopathy, neuropathy and nephropathy.[1] Hemoglobin A1c (HbA1C), a reflection of average blood glucose over the preceding 2–3 months[2,3], is a good predictor of the risk of developing diabetes-related complications.[1] Measurement of HbA1C was initiated in the 1970s.[4] There are different ways to assess HbA1C, including immunoassays and ion-exchange or affinity chromatography. [5] Chromatography assays are based on either charge differences or structure differences, as glycation of hemoglobin adds an extra negative charge.[4,6] Most of the commercially available platforms, of which there are 15–20, use either type of chromatography.[4] HbA1C lab tests are standardized by the National Glycohemoglobin Standardization Program (NGSP).

Point-of-care (POC) HbA1C has been utilized for over two decades, and while it is not standardized for diagnosing diabetes it is comparable to standard central laboratory testing.[7] While POC hemoglobin A1C correlates with values obtained by central lab testing there is little data on the accuracy of POC testing when the HbA1C ≥ 14%. An accurate HbA1C is vital for monitoring glycemic control and appropriate counseling.[8,9]

The Diabetes Research in Children Network (DirecNet) study found that the DCA 2000 POC HbA1C values had good correlation with central laboratory values ($r=0.94$, $p < 0.001$), albeit higher with a mean difference of 0.2% (95% confidence interval, 0.14–23%, $p < 0.001$).[7] The POC HbA1C range in the DirecNet study was 7-11%.[7] In our clinic we have a significant number of patients with poor glycemic control and POC HbA1Cs ≥ 14%. The primary goal of this study was to compare POC testing to central lab testing when POC testing is ≥ 14% and to characterize those patients. Our secondary objective was to compare both methods of testing when the A1C is 9–13.9%.

METHODS

We performed a retrospective chart review of all children treated for diabetes at Rhode Island Hospital between the years 2007–2013. Of the 1002 patients with POC tests, 72 children with T1DM aged 5–21 years old had a corresponding central lab value done on the same day. Twenty-seven children had a POC HbA1C < 9%, and these patients were excluded from the study, as this HbA1C range has been previously studied and our focus was on patients with poor glycemic control. Twenty-four had a POC HbA1C between 9–13.9% and 19 patients had a POC HbA1C ≥14%. The POC test does not report numbers higher than 14%.

During routine clinic visits, HbA1C was measured on a fingerstick blood sample via the DCA 2000 (until April 2010) or DCA Vantage methods (Seimens, Tarrytown, NY, USA). Both of these methods use latex immuno-agglutination inhibition methodology and are certified by the NGSP. The predecessor to the DCA Vantage, the DCA 2000, has been shown to correlate well to HPLC methods ($r=0.939$, $p < 0.000110$ and $r=0.94$, $p < 0.00111$).[6] The whole blood sample, obtained by venipuncture, was analyzed via the Tosoh Automated Glycohemoglobin Analyzer HLC-723G8 (Tosoh Bioscience Inc, South San Francisco, CA, USA). This method utilizes non-porous ion exchange, high performance liquid chromatography (HPLC). It is also certified by the NGSP.

For patients with POC HbA1Cs ≥ 14%, we collected data on age, duration of diabetes, pubertal status (Tanner stage), and insulin regimen. Their duration of diabetes was rounded to the nearest year. Pubertal status was determined using the Tanner Method of pubertal staging.[10,11] Those on a basal-bolus insulin regimen were taking multiple daily injections with both rapid-acting insulin and one dose of Glargine or Detemir as their basal insulin. Those on a split-mixed regimen were taking NPH with a rapid-acting insulin, divided in 2–3 injections daily.

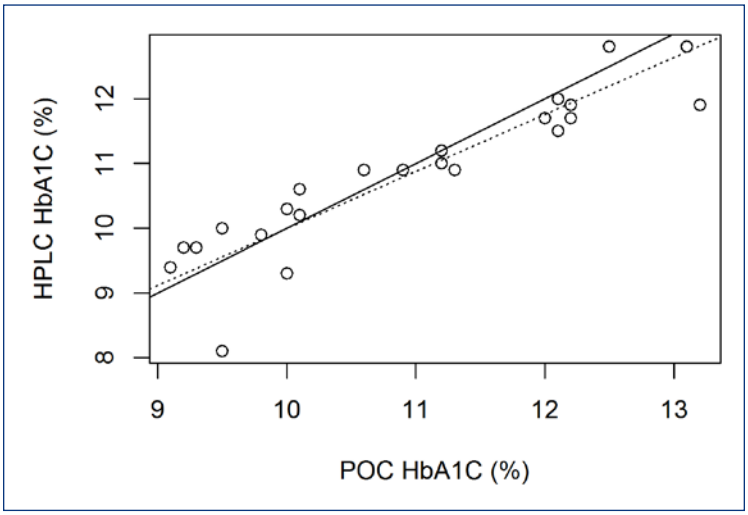
Each patient whose POC HbA1C was in the range of 9–13.9% was included in a concordance analysis of POC and central lab HbA1C. Because both biomarkers contain error, Deming regression and Bland-Altman plots were used to evaluate concordance between POC testing and HbA1C done by HPLC. A Bland-Altman plot was used to evaluate agreement between the two clinical measurements, given that both measurements contain error.[12] This plot was used as it visually illustrates the possibility of systematic bias by plotting the mean of the two measurements (x-axis) by their difference (y-axis). All data analyses were performed using the base and MethComp packages with R 3.0.0 (R Foundation for Statistical Computing, Vienna, Austria).[13]

Table 1. Clinical characteristics of patients with POC HbA1C ≥ 14%

Patient	Age (years)	Sex	HPLC HbA1C (%)	Duration of DM (years)	Tanner Stage	Insulin Regimen
1	16.7	F	14.9	7	5	split-mixed
2	17.3	M	13.7	7	5	basal-bolus
3	17.7	M	14.4	16	5	split-mixed
4	14.8	M	13	1	4	basal-bolus
5	14.8	M	14.1	2	3	split-mixed
6	17.9	M	15.9	11	5	basal-bolus
7	17.3	M	12.5	2	5	basal-bolus
8	15.2	M	11.1	3	5	basal-bolus
9	14.6	F	14	5	4	split-mixed
10	11.2	M	15.2	3	1	split-mixed
11	14.6	F	13.5	2	4	basal-bolus
12	18.9	F	14.2	12	5	split-mixed
13	15.6	F	15.6	4	5	basal-bolus
14	16.7	M	16.3	11	5	split-mixed
15	16.3	F	13.2	11	5	split-mixed
16	16.7	F	13.8	8	5	split-mixed
17	15.5	F	16.2	5	5	basal-bolus
18	16.9	M	14.6	3	5	split-mixed
19	17.5	M	12	4	4	split-mixed

Abbreviations: POC = point of care; HbA1C = hemoglobin A1C, HPLC = high performance liquid chromatography, DM = diabetes mellitus.

Figure 1. Deming Regression Comparing POC (*point-of-care)* HbA1C (*hemoglobin A1C)* and HPLC (*high performance liquid chromatography)* HbA1C.



RESULTS

There were 24 patients (12 female, 12 male) with POC values in the range of 9.1–13.2%; mean age was 14.2 years (range 4.6–21.5 years). There were 19 patients with POC values ≥ 14%, with mean age 16.1 years (range 11.2-18.9 years) and male predominance (11 male, 8 female) (**Table 1**). None of the patients in our cohort had a known hemoglobinopathy affecting A1C measurements.

As illustrated in **Figure 1**, the Deming regression indicates good concordance between the two markers when the POC values were < 14%. In addition, the 95% confidence interval of the intercept contains 0 (i.e., [-1.68, 3.35]), 0 being perfect agreement (the observed intercept was 1.19), and the 95% confidence interval of the slope contains 1 (i.e., [0.69, 1.13]), where a slope of 1 reflects perfect agreement (the observed

Figure 2. Bland-Altman Plot Comparing POC (*point-of-care)* HbA1C (*hemoglobin A1C)* and HPLC (*high performance liquid chromatography)* HbA1C.

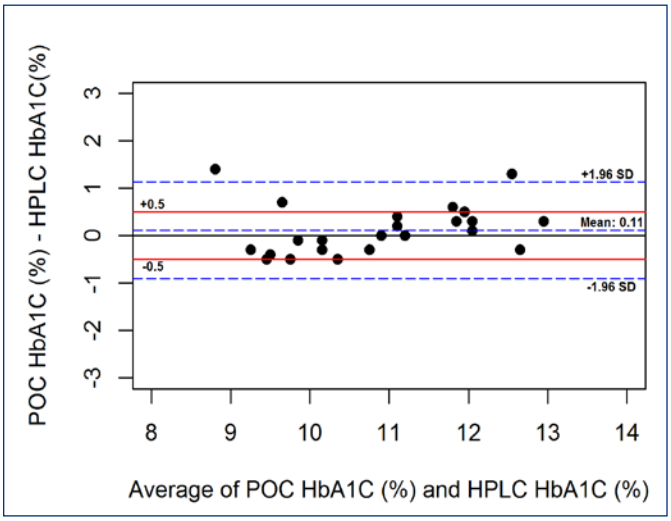
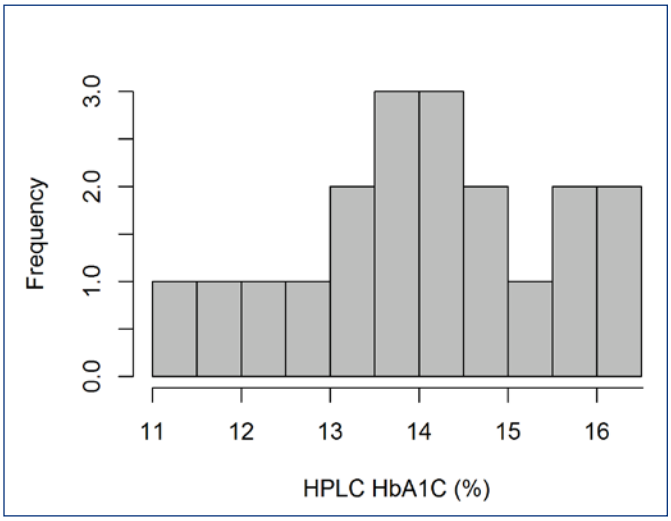


Figure 3. Histogram of HPLC (*high performance liquid chromatography)* HbA1C (*Hemoglobin A1C)* Values for POC (*point-of-care)* HbA1C Values ≥ 14%.



slope was .88), both indicating concordance of the two markers. The Bland-Altman plot indicates no major systematic trend in the difference between the two markers while also revealing only a small systematic bias between the two measures: HbA1C done by HPLC appears to be slightly lower on average compared to POC HbA1C, with a mean difference of .1125 (95% CI [-0.11, 0.33]). In addition, four observations fell outside the 0.5% bounds (a difference of 0.5% is considered clinically significant);[14] of these, only two observations fell outside of the 1.96 standard deviation bounds, all with HPLC values underestimating POC values (**Figure 2**). Any possible difference between machines could not be assessed given the low sample size.

For the patients with POC HbA1C values ≥ 14% the mean central lab value was 14.1% (95% CI [13.4, 14.8]), with a range of 11.1–16.3 and a standard deviation of 1.4% (**Figure 3**). The patients with POC HbA1Cs ≥ 14% (**Table 1**) had diabetes for an average duration of ~6 years (range 1–16 years). Thirteen patients were Tanner 5, four were Tanner 4, one was Tanner 3, and one patient was Tanner 1. Eleven patients were on a split-mixed insulin regimen (9 of those using a pen with pre-mixed insulin) and 8 patients were using a basal-bolus regimen.

DISCUSSION

Our study shows that POC testing is concordant with central lab testing when the A1C is 9–13.9%, with POC values being slightly higher, similar to the findings by DirecNet study.[7] When the POC value is ≥ 14%, however, the corresponding central lab value varies greatly. The mean HbA1C value done via central lab testing is 14.1%, but the range is wide, from 11.–16.3%.

This wide variation makes detecting relative changes in HbA1C at subsequent clinic visits problematic. It is undeniable that when one's POC A1C is ≥ 14%, the patient is in poor glycemic control whether the actual A1C is 11% or 16%. In fact, the new American Diabetes Association (ADA) guidelines recommend target A1C < 7.5% across all pediatric age groups.[15] However, being able to track relative changes in HbA1C at subsequent clinic visits is important as it allows clinicians to gauge whether or not interventions are effective. Additionally, knowledge that their HbA1C is trending down may provide positive reinforcement to adolescents. We therefore suggest routine central HbA1C testing when the POC is ≥ 14% for proper counseling and monitoring of glycemic control.

Studies in adults with Type 1 or Type 2 diabetes have shown that when the HbA1C is immediately available, there is improvement of glycemic control that persists at 12 months.[9] The same results have not been found in children, however. Agus et al performed a randomized controlled trial of children less than 18 years of age, and found that having the HbA1C available for immediate feedback did not lead to persistent improvement of glycemic control.[8] The study did find that using a POC machine is helpful in reducing the amount of patient-clinician communication required in between visits, thus POC testing is becoming standard practice in many diabetes centers.

It is important to note that while there are many commercially available POC HbA1C machines, not all of them have been found to meet generally accepted analytic performance criteria.[16] Lenters-Westra et al tested eight different machines and found that only the DCA Vantage (the machine used in this study) and the Afinion met the acceptance criteria of having a total CV (coefficient of variation)

of < 3% (in the clinically relevant range). However, there are still differences among the lot numbers of cartridges.[16,17]

The majority of patients with POC HbA1Cs ≥ 14% were male adolescents in mid-late puberty with long-standing diabetes duration. They were typically on a split-mixed insulin regimen, which is not surprising given that poorly controlled T1DM patients are often switched to twice daily injections to improve compliance. The SEARCH study found that in children with type 1 diabetes, there was a correlation between poor glycemic control and longer duration of diabetes.[18] This correlation is partially explained by the progressive loss of beta cell function with increasing diabetes duration. It is also well known that adolescents with T1DM pose particular challenges to maintaining good glycemic control.[19] Teenagers often do not adhere to their diabetes care regimen due to avoidance of standing out from their peers and increased incidence of depression.

Interventions found to help adolescents achieve improved glycemic control include involving family members, motivational interviewing (by a trained professional), ensuring regular diabetes clinic appointments (preventing loss-to-follow-up), and improving patient/provider communication by employing technology.[19] Use of an insulin pump has also been shown to be associated with lower HbA1C levels.[20]

Our study is limited by the sample size. In our retrospective chart review we found that not many patients had both a POC HbA1C and one done by central laboratory testing, limiting our sample size. Additionally, characterization of those with POC HbA1C values ≥ 14% was only done in those with both values available.

CONCLUSION

The results from this study provide evidence for good concordance between HbA1C done by HPLC and POC HbA1C values < 14% and wide variation for POC HbA1C values ≥ 14%. We therefore suggest routine central HbA1C testing when the POC is ≥ 14% for proper counseling and follow-up of glycemic control.

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