An Assessment of Standardisation of HbA1c Testing Across Clinical Laboratories in India and its Impact on Diabetes Management

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Abstract

Objectives: This study is aimed at evaluating the degree of standardisation of HbA1c and glucose testing across accredited laboratories in India.

Methods: The information declared on the scope of testing by 147 medical laboratories accredited by the National Accreditation Board for Testing and Calibration Laboratories (NABL) across India was used by the authors for this study (http://www.nabl-india.org). This information on the scope of testing is available within the public domain and is accessible through the NABL website and covered laboratories accredited between 2009 and 2012. We focussed on HbA1c and glucose tests offered by laboratories and documented the way tests were named, the methodologies used and the degree of confidence in testing based on the coefficient of variation (CV). The data was independently reviewed by two medical biochemists and then subjected to analysis.

Results: Although the glucose test appeared to be ubiquitous, HbA1c assays appeared on the scope of testing in 87.1% of the laboratories. The HbA1c tests however appear to be poorly standardised across laboratories. We noted gross differences in test nomenclature, methodology and analytical performance across laboratories.

Conclusion: This is one of the first studies that has focussed on the standards of laboratory care for diabetes management in India. The study highlights the lack of standardisation in nomenclature, analytical performance and methodology of tests used for HbA1c in NABL accredited laboratories across India. Affirmative actions in terms of improved regulation, patient advocacy, further studies on impact of laboratory quality and education of physicians, healthcare providers, laboratorians may improve harmonisation and quality of patient care in diabetes in India.

Introduction

Glycated haemoglobin (HbA1c) is formed by a non-enzymatic linkage of glucose to the N-terminal valine on the beta chains of haemoglobin. The fraction of HbA1c is a function of the average glucose concentration in the RBC milieu and its lifespan making it a good biomarker to evaluate glycaemic control over a period of 6-8 weeks.

HbA1c assays made their entry into clinics as a marker for glycaemic control around the late 70’s. Variability in analytical performance and differences among laboratories were key barriers to its acceptance in clinical settings. This was however changed by an impetus for international standardisation which came after the publication of the results of the Diabetes Control and Complications Trial (DCCT) in 1993.

Efforts from various organisations internationally over the last 20 years have resulted in standardisation of calibrators and improved comparability across methods. These efforts towards standardisation and their alignment with the results of DCCT and UK prospective diabetes study (UKPDS) have made HbA1c a biomarker of choice.
for monitoring glycaemic control, adjustment of therapy and as a tool for screening and diagnosis of diabetes mellitus.\textsuperscript{5-9} On comparison with conventional glucose based assays, HbA1c demonstrates lower pre-analytical and biological variability.\textsuperscript{10} The added operational advantages such as no requirements for fasting or timed collections have further served to promote the transition to HbA1c assays over glucose assays at various centres across the world.\textsuperscript{10,11}

This transition from glucose based assays to HbA1c is not clearly evident in India. The Improve Control India study indicated that physicians utilise fasting plasma glucose (FPG) and post prandial plasma glucose (PPG) more frequently than HbA1c for clinical decisions.\textsuperscript{12} Similar disparities are visible on other studies evaluating the standard of care in diabetes mellitus in India.\textsuperscript{13} Additionally the emphasis on patient education and patient awareness with regard to the value of HbA1c in the management of diabetes was found to be low.\textsuperscript{14,15} The lack of standardisation across laboratories was reported as one of the causes for this deficiency by doctors.\textsuperscript{12,16}

**Objectives**

This study is aimed at evaluating the degree of standardisation of HbA1c and glucose testing across accredited laboratories in India.

**Methods**

In order to answer the above question, we examined the information declared on the scope of testing by 147 medical laboratories accredited by the National Accreditation Board for Testing and Calibration Laboratories (NABL) across India. The information on the scope of testing was available within the public domain and is accessible

<table>
<thead>
<tr>
<th>HbA1c</th>
<th>Glucose</th>
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<tr>
<td>146 (99.3%)</td>
<td>128 (87.1%)</td>
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<tr>
<td>127 (86.4%)</td>
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**Table 1 : Summary of data on glucose and HbA1c tests offered on scope for 148 NABL accredited laboratories**

Fig. 1 : Nomenclature of HbA1c and glucose assays across NABL accredited medical laboratories

Fig. 2 : (A) Box and whisker plots showing the mid 95 percentile dispersion of coefficients of variation for 123 and 142 laboratory-methods for HbA1c and glucose. The boxes represent the data between the 25th and 75th percentiles whereas the whiskers represent the 2.5th to 97.5th percentile within the data set. The diamonds represent the mean value for each group. (B) A scatter plot representing coefficients of variation across 116 laboratories for HbA1c and glucose assays
through the NABL website and covered laboratories accredited between 2009 and 2012 (http://www.nabl-india.org). We focussed on HbA1c and glucose tests (fasting plasma glucose, post prandial plasma glucose, oral glucose tolerance test, random plasma glucose) offered by laboratories and documented the way tests were named, the methodologies used and the degree of confidence in testing based on the coefficient of variation (CV). The data was independently reviewed by two medical biochemists and inconsistent, inaccurate or incomplete data were excluded from analysis. Data was analysed using MS excel.

Results

Data across 148 laboratories showed that the glucose tests were more commonly available when compared to the HbA1c tests (Table 1). Six (4.6%) laboratories offered HbA1c testing by more than one methodology.

It was observed that over 90% of the laboratories identified tests for glucose using relatively harmonised terminology. In contrast there appeared to be a lack of consensus on the nomenclature of HbA1c assays across laboratories (Figure 1). Thirty six (28.1%) laboratories identified the HbA1c test by more than one name on the scope of testing.

On a comparative analysis of the CV, the HbA1c assay demonstrated higher variability (SD=2.49%, n=122) across laboratories as compared to glucose (SD=1.5%, n=141) (Figure 2A). The 25th percentile for the CV across laboratories corresponded to 2.5% and 2.52% for HbA1c and glucose assays respectively. Similarly the 75th percentile corresponded to 5.3% and 4.3% across laboratories for HbA1c and glucose respectively. The CV for HbA1c and glucose displayed a weak correlation across 116 laboratories (correlation coefficient 0.478, p < 0.001) (Figure 2B).

A total of 121 laboratories declared both the methodology and the CV for the HbA1c assay. Within this set 63.6% of laboratories utilised an HPLC based methods among which 36.4% of the laboratories went on to specify further details of the methodology. High Performance Liquid Chromatography (HPLC) methods in 20.8% and 15.6% laboratories were based on ion exchange and boronate affinity chromatography respectively. Additionally, 29.8% of laboratories declared the use of assays based on spectrophotometry/immunoturbidimetry for the measurement of HbA1c. In comparison, the methods used for glucose analysis across laboratories demonstrated better harmony with 55.6%, 41.5% and 2.8% using glucose oxidase, hexokinase and both methods respectively (Figure 3).

Conclusion

Laboratory based testing for HbA1c plays a key role in the diagnosis and establishment of targets for management of diabetes mellitus. Even as glucose estimation remains a more widely available test, a large number of accredited laboratories are offering HbA1c tests in India. Laboratories need to maintain precision and valid traceability (accuracy) of HbA1c assays to ensure that results generated are suitable for clinical use.

Measurements of uncertainty in the form of CV both within and across laboratories provide an insight into analytical performance and standardisation of a diagnostic test. More than 25% of the laboratories evaluated in this study declared a CV greater than 5% for HbA1c. A CV of 5% at an HbA1c concentration of 6.2% and 7.6% implies that such a laboratory may report a value within the range 5.6-6.8% and 6.9-8.3% respectively, assuming that there is no bias in the test method. The ranges highlight the potential for poor analytical performance to misguide healthcare providers toward a wrong diagnosis, over treatment...
Several countries have tried to address these issues through regulatory processes for both assay manufacturers and laboratories aimed at standardisation and continuous monitoring of analytical performance to ensure clinically valid results. In India, there exists a regulatory framework (enforced by Central Drugs Standard Control Organisation) that monitors the introduction of an in-vitro diagnostic test into the market. However in India, the aspects of continued monitoring of performance at the level of laboratories and standardisation are largely deficient. One of the key reasons for this deficiency is the fact that medical testing laboratories in India largely operate in an unregulated environment. We opine that this may be one of the key reasons for the lack of standardisation in test nomenclature, methodology and analytical performance observed across laboratories in this study. It is also noteworthy that data for this study was obtained from NABL accredited laboratories with established quality management systems. Such laboratories form only a small fraction (< 1%) of the medical laboratories in India based on market size estimates. This leads us to an opinion that this study only shows us only the tip of the iceberg and may be understating the magnitude of the problems observed.

There is a long history of nomenclature for HbA1c which include haemoglobin A1, haemoglobin A1c, glycosylated haemoglobin, glucosylated haemoglobin, glycated haemoglobin, glycohaemoglobin and so on. Experts from the International Federation of Clinical Chemistry (IFCC), International Diabetes Federation (IDF), European Association for the Study of Diabetes and the American Diabetes Association (ADA) have agreed on HbA1c in a consensus statement. Following this lead, we believe that laboratories in India too, should adopt a standardised naming convention based on consensus. This would assist in improving patient awareness and education within a clinical setting and assist in the standardisation of management of diabetes.

We also noted large variability in the declared precision across methods and laboratories for HbA1c assays. As most laboratories in this study used globally standardised assay platforms, the large degree of variability is indicative of deficits in monitoring analytical performance at the level of the laboratory. Curiously the observed deficits in standardisation presented in this study demonstrate patterns similar to that seen in the College of American Pathologists proficiency testing surveys in the early 90’s. A comparison across methods demonstrated that laboratories using ion exchange chromatography appeared to declare lower CV as compared to other methods. Laboratories using methods based on spectrophotometry and immunoturbidometry had the maximum degree of variation in the declared data.

The observed deficits in standardisation of HbA1c testing across laboratories may serve as a barrier for effective clinical use as indicated in earlier studies. Access to data from laboratories in the public domain, as done by NABL, enables experts and healthcare policy makers to analyse impacts and take proactive measures to promote standardisation. Regulators of laboratory accreditation such as NABL must monitor external quality assessment programmes to ensure standardisation across laboratories. This may be achieved by using proficiency testing material having defined targets for accuracy and precision across laboratories, as done by the College of American Pathologists. We also noted that studies evaluating the impact of laboratory quality on healthcare outcomes are lacking in India. Such studies may help in developing a consensus on laboratory standards and provide an impetus for regulation by the government or the medical community on testing laboratories. Furthermore such studies would go a long way in educating the healthcare community to demand for improved standards in laboratory testing. The prevalence and the projected estimates of the diabetes epidemic in India, make standardisation of laboratory testing a critical concern for safe and effective healthcare delivery.

Acknowledgements

We would like to acknowledge and thank the National Accreditation Board for Testing and Calibration Laboratories (NABL) for access to the scope of testing of accredited laboratories.

References


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**Significant Activity - API**

On the World Diabetes Day, a special POSTAL COVER with API LOGO with the help of postal department regarding Diabetic Patient Education Programme was released.