

# Global standardization of glycated hemoglobin measurement: the position of the IFCC Working Group<sup>1),2)</sup>

**International Federation of Clinical Chemistry and Laboratory Medicine (IFCC)  
IFCC Scientific Division**

Working Group on Standardization of HbA1c  
(WG-HbA1c)

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## Abstract

The measurement of glycated hemoglobin is central in the monitoring of glycemic control in patients with diabetes. There are at least 30 different laboratory assays commercially available to measure the proportion of HbA1c in blood. In 1995 the IFCC established a Working Group (IFCC WG-HbA1c) to achieve international standardization of HbA1c measurement. The main achievements can be summarized as follows: a) a reference measurement procedure has been established with purified primary calibrators; b) a network of reference laboratories has been developed worldwide; and c) work has begun on implementation of traceability to the IFCC reference system. The IFCC WG-HbA1c recognizes the recommendation of the IFCC-IUPAC Committee on Nomenclature, Properties and Units that the analyte measured by the IFCC reference measurement procedure has been defined as  $\beta$ N1-deoxyfructosyl-hemoglobin and that the recommended measurement units are mmol/mol. The IFCC WG-HbA1c recommends maintaining the use of the name HbA1c in clinical practice.

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**Keywords:** diabetes; glycated hemoglobin; HbA1c; network; reference methods; standardization.

## Background

The measurement of glycated hemoglobin (HbA1c;  $\beta$ N1-deoxyfructosyl-hemoglobin) is frequently used in diabetes management to monitor mid- to long-term glycemic control and to assess the risk of development of diabetic complications in patients with diabetes (1, 2). A level "A" recommendation in the 2002 guidelines by the US National Academy of Clinical Biochemistry (NACB) emphasizes these issues, also stating that treatment goals have to be based on the results of retrospective clinical trials, such as the Diabetes Control and Complications Trial (DCCT) and UK Prospective Diabetes Study (UKPDS) (3, 4).

At present at least 30 different laboratory methods are commercially available to measure the proportion of HbA1c in blood. A recent review on this topic has

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been published by John (5). There are a number of published reports relating to between-laboratory and between-method agreement for HbA1c, much of this information coming from National External Quality Assessment Schemes (EQAS) (6–12). In the United States, as well as in several other countries, participation in proficiency testing is mandatory. Based on one large proficiency survey (College of American Pathologists GH2 survey), more than 99% of the laboratories in the US use a method that is aligned to the National Glycohemoglobin Standardization Program (NGSP) (13). In addition to the Designated Comparison Method (DCM) developed by the NGSP, other DCMs have been developed in Sweden and in Japan. It is beyond the scope of the present paper to analyze the performance of different methods. However, data obtained from the above studies demonstrate that standardization of HbA1c methods between laboratories is still an important issue. Indeed, interlaboratory variability of 5%–7% (expressed as CV) has been shown for HbA1c values between 6% and 10% HbA1c (% of total Hb). Poor between-laboratory agreement can also be found when laboratories are using the same manufacturer's method, although some manufacturers display between-laboratory agreement as low as 3%.

### The IFCC program for HbA1c standardization

In 1995 the IFCC established a Working Group (IFCC WG-HbA1c) to achieve international standardization of HbA1c measurement (14). The activities achieved by this WG so far can be summarized as follows:

- a) Highly purified HbA1c and HbA0 materials have been produced (15), and these have been made available to the 14 laboratories of the IFCC network (see below). These primary reference materials will be available in 2007 through the Institute for Reference Materials and Measurements (IRMM) (identification codes 466 and 467, respectively).
- b) A reference measurement procedure for HbA1c has been developed (16). This method is based on the proteolytic digestion of red cell hemoglobins followed by quantitative peptide mapping by HPLC-mass spectrometry or HPLC-capillary electrophoresis. It has been voted on by the National Societies affiliated to the IFCC and published as an "approved IFCC reference measurement procedure" (17).
- c) A network of reference measurement laboratories has been implemented. Two experiments are performed every year, in which materials are distributed to the laboratories for comparison purposes, and also to assign HbA1c values to candidate calibrators and controls. These studies have been performed since 1999 and have also been important in refining the reference measurement procedure, which is regularly updated as soon as new technical information becomes available (18). The network has developed a set of

rules for the certification of reference values and for the calculation of the uncertainties of the calibrators (19, 20).

- d) Several comparison studies have been performed between the IFCC reference measurement laboratories and the existing DCMs. These studies found stable relationships between the IFCC and different DCM systems and the corresponding regression equations (the "master equations") were published (21).
- e) Secondary reference materials have been produced in the form of panels of fresh and frozen whole blood and distributed to the manufacturers and to laboratories performing DCMs to anchor their methods to the IFCC reference system.

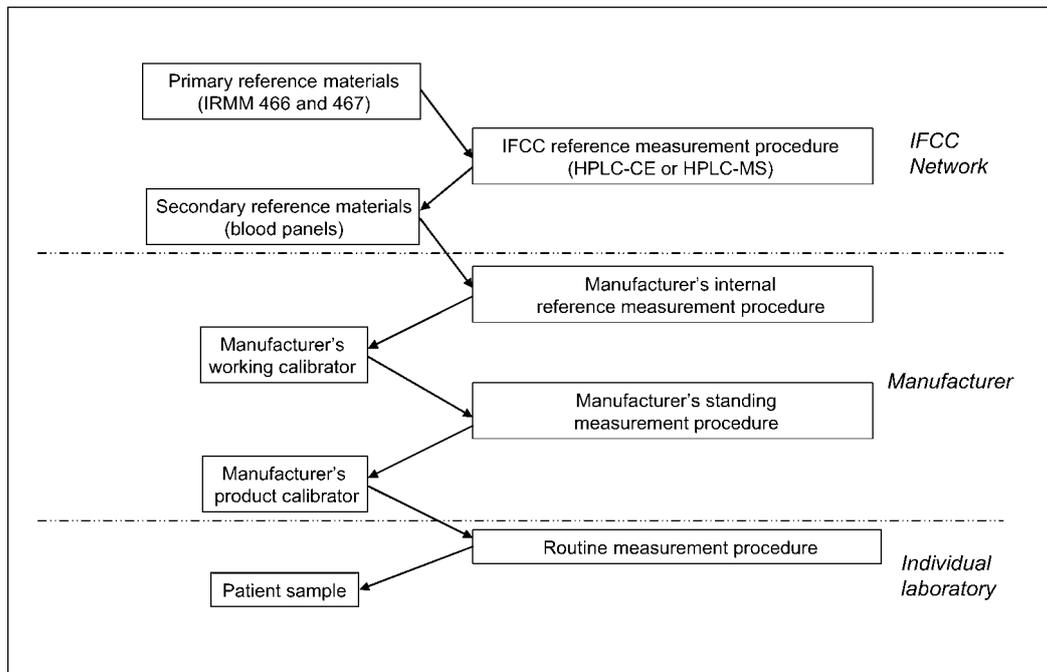
Figure 1 displays the IFCC reference system and the traceability chain for HbA1c.

### Current issues

Traceability to the IFCC reference system for HbA1c has not been implemented because concern has been expressed about the impact that changes in HbA1c values may have on patient care (22–24). Criticisms are related to the fact that the IFCC reference measurement procedure gives HbA1c values numerically lower (–1.3% to –1.9% across the pathophysiological range) than those obtained by the DCMs and NGSP-aligned methods. This finding has generated significant debate on how HbA1c should be reported.

From the beginning of 2004, another group dealing with the topic of harmonizing HbA1c assays has been established among three clinical societies: the American Diabetes Association (ADA), the European Association for the Study of Diabetes (EASD) and the International Diabetes Federation (IDF). The decisions that this team of experts has reached so far have been published (24) and can be summarized as follows:

1. Adopt the IFCC reference measurement procedure as the new global standard for calibration of HbA1c assays by manufacturers.
2. Use the new IFCC methodology to anchor an "international certification process" within the existing international laboratory networks. The ADA/EASD/IDF group did not elaborate on this statement. Currently the IFCC network of reference laboratories is anchoring the other DCM networks (the NGSP network in the US and the networks in Sweden and in Japan). This is actually carried out through the exercises of the network, in which the WG-HbA1c has been able to monitor the stability of the master equations previously published (21).
3. Manufacturers/laboratories should not change the HbA1c values reported until further work has been completed, i.e., DCCT/UKPDS numbers and derived decision limits will continue to be used. Further work refers to the studies designed to investigate the relationship between HbA1c and mean blood glucose (MBG).



**Figure 1** IFCC reference measurement system and traceability chain for HbA1c.

- Following completion of the ongoing clinical trials, if the relationship between HbA1c and MBG is sufficiently defined and constant in different populations worldwide, HbA1c could be reported as MBG.

Until now, the IFCC WG-HbA1c has not taken an official position on definitive implementation of traceability to the IFCC reference system for HbA1c, nor has it expressed an opinion on possible endorsement of the document published by the ADA/EASD/IDF Working Group (24). The present document, therefore, serves to express the IFCC WG-HbA1c position on this issue.

### Name and units for the IFCC standardized HbA1c test

Recently, a recommendation by the IFCC-IUPAC Committee on Nomenclature, Properties and Units (C-NPU) has been prepared that relates to the systematic name and units for HbA1c as measured by the IFCC reference measurement procedure. This IFCC-IUPAC document has been approved by the National Societies affiliated to the IFCC and is published in this issue of the journal as an IFCC recommendation (25).

Briefly, the C-NPU proposes that the term for indicating the fraction of the  $\beta$ -chains of hemoglobin that has a stable hexose adduct on the N-terminal amino acid valine may be expressed as "Hemoglobin beta chain(Blood)—N-(1-deoxyfructos-1-yl)hemoglobin beta chain". In the IFCC-IUPAC document it is recommended that this term be used to describe the measurand of the IFCC reference measurement procedure, and that this can be shortened in "everyday speech" to DOF-Hb. The IFCC WG-HbA1c agrees with the use of

this nomenclature when used to describe the analyte measured by the IFCC reference measurement procedure. The IFCC WG-HbA1c believes that this term cannot be used to describe the fraction measured by routine clinical methods; these methods, even though traceable to the IFCC reference measurement procedure, do not specifically measure the fraction reflecting glycation at the N-terminal valine on the  $\beta$ -chains of the hemoglobin molecule. In addition, the IFCC WG-HbA1c does not agree to the use of DOF-Hb in clinical practice and recommends that the abbreviation "HbA1c" remains as long as the measurands of routine clinical methods remain unchanged. With regard to the measurement units and numerical value expressed in IFCC numbers, to avoid confusion among healthcare personnel and patients, the measurement unit "millimole per mole" will be chosen instead of "percent" (%).

The IFCC WG-HbA1c does not support the concept of reporting HbA1c only as "mean blood glucose" (24).

### How to move to the IFCC values for HbA1c

When introducing a new analytical system or a new method of reporting results, it is important that this is done in a planned way. This is especially true if the change may impact patient care. It is crucial if HbA1c values are changed that there is thorough planning and preparation of literature that informs clinicians, patients and laboratories about the new way of reporting HbA1c results. It will be necessary to prepare documents so that the crucial information collected up to now (for instance, data from large clinical trials such as the DCCT and UKPDS) is not lost when expressing HbA1c in the new IFCC standardized val-

ues. It is difficult to estimate in advance how long this phase will be, but probably at least 1 year is needed.

It also seems advisable to consider the results of the EASD/ADA/IDF study evaluating the relationship between HbA1c and MBG. When this trial is complete, it will be necessary to define acceptability limits for this relationship for implementing estimation of MBG from the measurement of HbA1c. To this end, the IFCC WG-HbA1c is available to collaborate with the above-mentioned Societies in such work. If the correlation between HbA1c and MBG is sufficiently close to the selected limits, then reporting of an estimated MBG (eMBG), using a mathematical formula based on the IFCC standardized HbA1c value, together with the HbA1c value itself, will be possible. The final results of the study are expected in December 2007.

## References

- Sacks DB, Brunns DE, Goldstein DE, Maclaren NK, McDonald JM, Parrott M. Guidelines and recommendations for laboratory analysis and management of diabetes mellitus. *Clin Chem* 2002;48:436–72.
- American Diabetes Association. Standards of medical care in diabetes. *Diabetes Care* 2004;27(Suppl 1):S15–35.
- DCCT Research Group. The effect of intensive treatment of diabetes on the development and progression of long-term complications in insulin-dependent diabetes mellitus. *N Engl J Med* 1993;329:977–86.
- UK Prospective Diabetes Study (UKPDS) Group. Intensive blood glucose control with sulphonylureas or insulin compared with conventional treatment and risk of complications in patients with type 2 diabetes (UKPDS 33). *Lancet* 1998;352:837–53.
- John WG. Haemoglobin A<sub>1c</sub>: analysis and standardisation. *Clin Chem Lab Med* 2003;41:1199–212.
- <http://www.ukneqas.org.uk/>. Accessed June 21, 2007.
- <http://www.euroreflab.com/>. Accessed June 21, 2007.
- Valdiguie P, de Graeve J, Corberand JX, Fernet P. 20 years of quality control in clinical laboratories. *Ann Biol Clin* 2000;58:659–61 ([www.ctcb.com](http://www.ctcb.com). Accessed June 21, 2007).
- <http://www.equalis.se/>. Accessed June 21, 2007.
- <http://www.instand-ev.de/>. Accessed June 21, 2007.
- <http://www.glicata.org/>. Accessed June 21, 2007.
- <http://www.rcpaqap.com.au>. Accessed June 21, 2007.
- <http://www.ngsp.org>. Accessed June 21, 2007.
- Hoelzel W, Miedema K. Development of a reference system for the international standardisation of HbA<sub>1c</sub>/glycohemoglobin determinations. *J Int Fed Clin Chem* 1996;9:62–7.
- Finke A, Kobold U, Hoelzel W, Weycamp C, Jeppsson JO, Miedema K. Preparation of a candidate primary reference material for the international standardisation of HbA<sub>1c</sub> determinations. *Clin Chem Lab Med* 1998;36:299–308.
- Kobold U, Jeppsson JO, Dülffer Th, Finke A, Hoelzel W, Miedema K. Candidate reference method for HbA<sub>1c</sub> based on peptide mapping. *Clin Chem* 1997;43:1944–51.
- Jeppsson JO, Kobold U, Barr J, Finke A, Hoelzel W, Hoshino T, et al. Approved IFCC reference method for the measurement of HbA<sub>1c</sub> in human blood. *Clin Chem Lab Med* 2002;40:78–89.
- <http://www.ifcchba1c.net/>. Accessed June 21, 2007.
- Konnert A, Berding C, Arends S, Parvin C, Rohlfing CL, Wiedmeyer HM, et al. Statistical rules for laboratory networks. *J Test Eval* 2006;34:1–7.
- Konnert A, Arends S, Schubert S, Berding C, Weykamp C, Siebelder C. Uncertainty calculation for calibrators of the IFCC HbA<sub>1c</sub> standardization network. *Accred Qual Assur* 2006;11:316–28.
- Hoelzel W, Weykamp C, Jeppsson JO, Miedema K, Barr JR, Goodall I, et al. IFCC reference system for measurement of hemoglobin A<sub>1c</sub> in human blood and the national standardization schemes in the United States, Japan, and Sweden: a method-comparison study. *Clin Chem* 2004;50:166–74.
- Hanas R. Psychological impact of changing the scale of reported HbA<sub>1c</sub> results affects metabolic control. *Diabetes Care* 2002;25:2110–1.
- Home P. Standardisation of glycated haemoglobin. *Br Med J* 2004;329:1196–7.
- Sacks DB for the ADA/EASD/IDF Working group for the HbA<sub>1c</sub> Assay. Global harmonization of hemoglobin A<sub>1c</sub>. *Clin Chem* 2005;51:681–3.
- Nordin G, Dybkaer R. Recommendation for term and measurement unit for “HbA1c”. *Clin Chem Lab Med* 2007;45:1081–2.