EDUCATIONAL COMMENTARY – HEMOGLOBIN A1C

Educational commentary is provided through our affiliation with the American Society for Clinical Pathology (ASCP). To obtain FREE CME/CMLE credits click on Earn CE Credits under Continuing Education on the left side of the screen.

LEARNING OBJECTIVES

On completion of this exercise, the participant should be able to

- explain the American Diabetes Association criteria for the diagnosis and monitoring of diabetes mellitus using glycated hemoglobin (HbA1c) levels.
- explain the correlation between HbA1c levels and the plasma glucose concentration.
- identify or classify methods available to measure HbA1c.
- discuss factors affecting HbA1c levels, including interferences and variant hemoglobins.
- interpret test results for HbA1c levels.

Introduction

Until 2010, the American Diabetes Association (ADA) criteria for the diagnosis of diabetes mellitus included any of the following test results (verified by repeated analysis): a fasting plasma glucose level of 126 mg/dL or greater (reference range: 60-99 mg/dL; to convert to mmol/L, multiply by 0.0555); a random plasma glucose level of 200 mg/dL or greater, in conjunction with symptoms of hyperglycemia (polyuria, polydipsia, and unexplained weight loss); or a 2-hour plasma glucose of 200 mg/dL or greater during an oral glucose tolerance test using a 75-g glucose load. In 2010, the ADA added a fourth possible criterion, a glycated hemoglobin (HbA1c) concentration of 6.5% or greater.¹ The adoption of the fourth criterion was based on the 2009 recommendation of an International Expert Committee. Reasons for the recommendation included epidemiologic evidence that HbA1c levels were related to the risk for retinopathy, the convenience of using a nonfasting sample, and that levels of HbA1c are more stable than plasma glucose levels during periods of stress and illness. The plasma glucose criteria remain valid, and there is no perfect correlation between any 2 of the criteria. Data from the National Health and Nutrition Examination Survey (NHANES) indicate that using the HbA1c cutpoint of 6.5% or greater, rather than the fasting plasma glucose cutpoint of 126 mg/dL or greater, would identify one-third fewer cases of undiagnosed diabetes mellitus.²

Hemoglobin A1c

In normal hematologic function, the composition of hemoglobin is approximately 95% to 97% HbA and 3% to 5% Hb A2 and HbF (fetal). Glucose reacts irreversibly and nonenzymatically with the amino-terminal valine residues of the β chains of HbA to form HbA1c. Glycated hemoglobin levels are expressed as the amount of HbA1c as a percentage of total hemoglobin. Several factors affect the amount of HbA1c
that is formed; two major factors are the production and destruction of red blood cells and the plasma glucose concentration during the life span of these cells. Glycated hemoglobin concentrations represent the average glucose levels during the preceding 8 to 12 weeks in persons with normal hematologic function.

**Diagnosis of Diabetes Mellitus**

**Table I** summarizes the ADA criteria for the diagnosis of diabetes mellitus. HbA1c levels may be used for the diagnosis of diabetes mellitus and prediabetes. According to the ADA criteria, the HbA1c test should be performed by a laboratory that is NGSP (formerly, National Glycohemoglobin Standardization Program) certified and standardized to the assay used in the Diabetes Control and Complications Trial. The NGSP’s purpose is to standardize HbA1c results to those of the two studies, the Diabetes Control and Complications Trial and the United Kingdom Prospective Diabetes Study, that established the direct relationship between HbA1c levels and outcome risks in patients with diabetes mellitus. When an HbA1c concentration is 6.5% or greater, the test should be repeated before a diagnosis of diabetes mellitus is made, unless the patient is exhibiting classic symptoms of hyperglycemia or is in a hyperglycemic crisis. Repeating the HbA1c test is preferable to using a different diagnostic test, such as fasting plasma glucose; however, another test can be performed. If two different tests are used and the results agree (both above the cutpoint), the diagnosis is confirmed. If the results from two different tests are discordant (one above and one below the cutpoint), the test with results above the cutpoint should be repeated and the diagnosis should be based on the confirmed test results.

**Table I. American Diabetes Association Criteria for Diagnosis of Diabetes Mellitus**

<table>
<thead>
<tr>
<th>Stage</th>
<th>Plasma Glucose and HbA1c Tests</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Fasting Glucose</td>
</tr>
<tr>
<td>Normal</td>
<td>&lt; 100 mg/dL</td>
</tr>
<tr>
<td>Diabetes mellitus</td>
<td>≥ 126 mg/dL</td>
</tr>
<tr>
<td>Prediabetes</td>
<td>100-125 mg/dL (IFG)</td>
</tr>
</tbody>
</table>

Abbreviations: HbA1c, glycated hemoglobin; IFG, impaired fasting glucose; and IGT, impaired glucose tolerance.

*a Using plasma glucose and HbA1c tests

*b Polyuria, polydipsia, and unexpected weight loss

Adapted from the American Diabetes Association Position Statement, Standards of Medical Care in Diabetes—2013. [http://care.diabetesjournals.org/content/36/Supplement_1/S11.full.pdf+html](http://care.diabetesjournals.org/content/36/Supplement_1/S11.full.pdf+html)
EDUCATIONAL COMMENTARY – HEMOGLOBIN A_{1C} (cont.)

Monitoring Diabetes Mellitus

The two primary tools for monitoring the effectiveness of treatment for diabetes mellitus are self-monitoring of blood glucose, and HbA_{1c} testing. The HbA_{1c} level reflects the glycemic control status of the previous two to three months and has been correlated to the risk for complications of diabetes mellitus. An HbA_{1c} level of less than 7% is a reasonable goal for treatment of diabetes mellitus, but goals of 6.5% or 8% may be considered for selected patients. The frequency of testing should depend on the patient’s clinical situation: ADA recommendations include a minimum of twice a year for patients who have stable glycemic control and quarterly for patients who are not meeting glycemic goals or whose treatment has changed.\(^1\)

Because patients understand and are familiar with glucose values obtained by self-monitoring, the ADA and other organizations recommend that an average glucose value (also called the estimated average glucose or eAG) be reported with the HbA_{1c} level. A formula for calculating the average glucose value from the HbA_{1c} level was developed using data (approximately 2700 glucose measurements during a 3-month period in 507 adults with type 1, type 2, and no diabetes) collected as part of the HbA_{1c}-Derived Average Glucose trial.\(^4\) Table II shows the correlation of selected levels of HbA_{1c} and average glucose using the following formula: eAG = 28.7 \times HbA_{1c} – 46.7. A calculator for converting HbA_{1c} level to average glucose value is available at http://professional.diabetes.org/eAG. If access to the formula or online calculator is unavailable, approximate eAG can be calculated by remembering that an HbA_{1c} concentration of 6% equals 126 mg/dL of glucose, and for every 1% change in HbA_{1c} concentration, the level of eAG changes by 28 mg/dL. Glucose values obtained using this calculation are typically within 2 mg/dL of the value obtained using the formula.

Table II. Correlation of HbA_{1c} and eAG\(^a\)

<table>
<thead>
<tr>
<th>HbA_{1c}, %</th>
<th>eAG, mg/dL</th>
</tr>
</thead>
<tbody>
<tr>
<td>6</td>
<td>126</td>
</tr>
<tr>
<td>7</td>
<td>154</td>
</tr>
<tr>
<td>8</td>
<td>183</td>
</tr>
<tr>
<td>9</td>
<td>212</td>
</tr>
<tr>
<td>10</td>
<td>240</td>
</tr>
</tbody>
</table>

Abbreviations: eAG, estimated average glucose; HbA_{1c}, glycated hemoglobin.
\(^a\) Using the formula eAG = 28.7 \times HbA_{1c} – 46.7

Adapted from http://professional.diabetes.org/eAG.
HbA1c Testing Methods

Most HbA1c testing methods separate glycated from nonglycated components based on either charge or structural differences. Examples of methods based on charge difference include ion-exchange chromatography, agar gel electrophoresis, and high-performance liquid chromatography. Boronate affinity chromatography and immunoassays are examples of methods based on structural differences. Methods may vary in the extent that they measure compounds other than HbA1c, but standardization allows values obtained by different methods to be compared and universal cutpoints to be adopted. Laboratory personnel should be aware of any possible limitations of the HbA1c method used in their laboratory.

Interpretation of Test Results

Table I presents interpretive information, including HbA1c levels (verified by repeated analysis), for the diagnosis of diabetes mellitus and prediabetes. In persons without diabetes, the HbA1c range is approximately 4% to 6%. Because studies have shown increased risk for diabetes mellitus at HbA1c levels as low as 5.5%, the ADA has termed the range of 5.7% to 6.4% as prediabetes (sometimes referred to as impaired glucose homeostasis). The ADA recommends that patients diagnosed as having prediabetes be referred to a weight loss and exercise program; metformin therapy may also be considered. These patients should undergo testing at least annually for the development of diabetes mellitus.

Factors that influence the production or destruction of red blood cells will affect the interpretation of HbA1c levels. Hemoglobin A1c results would be falsely lowered in patients with hemolytic anemia and those recovering from acute blood loss regardless of testing method. Some studies have concluded that HbA1c levels are increased in iron-deficiency anemia, but other studies have found no effect. Other conditions and compounds reported to affect HbA1c levels include vitamins C and E, hypertriglyceridemia, hyperbilirubinemia, uremia, chronic alcoholism, and opiate addiction.

The presence of hemoglobin variants (e.g., HbS, HbC, HbD, and HbE) or levels of HbF greater than 10% can affect HbA1c results, but this is method dependent. Information compiled by the NGSP on the effects of variant hemoglobins on the most-used HbA1c methods can be obtained at http://www.ngsp.org/interf.asp. Laboratory managers should consider potential interferences and the prevalence of variant hemoglobins among their particular patient populations when selecting a method or interpreting results.

When using HbA1c for patient monitoring, the performance characteristics of the method are important in determining if changes in HbA1c levels indicate a clinically significant change in glycemic control.
EDUCATIONAL COMMENTARY – HEMOGLOBIN A1C (cont.)

Treatment guidelines from the ADA, the European Association for the Study of Diabetes, and the National Institute for Clinical Excellence in the United Kingdom recommend using a decrease in the HbA1c concentration by 0.5% to evaluate the effectiveness of new treatment regimens. This criterion correlates to a within-laboratory coefficient of variation of 2% or lower. Most accredited laboratories are now able to attain this due to method standardization and increased emphasis on progressively lowering proficiency testing acceptability criteria. Authors of one review concluded that when HbA1c is measured in an accredited laboratory, physicians can be reasonably certain that a difference of 0.5% or greater between successive patient samples represents a statistically significant change in glycemic control.

Summary

Glycated hemoglobin is a stable molecule formed when hemoglobin in red blood cells is exposed to glucose during the life span of these cells. It represents the average glucose concentration of the previous two to three months. Levels of 5.7% to 6.4% are diagnostic for prediabetes and indicate an increased risk for diabetes mellitus, and HbA1c levels of 6.5% or greater are diagnostic for diabetes mellitus.

The recommended goal for HbA1c levels when monitoring treatment of diabetes mellitus is typically less than 7%, but it may be modified for selected patients. A calculated eAG concentration should be included with each HbA1c report. The potential interferences and response to hemoglobin variants for the method used by the laboratory should be considered when interpreting HbA1c levels.

References


EDUCATIONAL COMMENTARY – HEMOGLOBIN A₁C (cont.)


© ASCP 2013