Tests for Screening and Diagnosis of Type 2 Diabetes

Mary E. Cox, MD, and David Edelman, MD

Although type 2 diabetes is common and tests to screen for and diagnose it are widely available, the disease remains under-diagnosed.1 Approximately 25% of people with a new diabetes diagnosis already have microvascular disease, suggesting that they have had the disease for 4–7 years by the time of diagnosis.2,3

In these patients, it is speculated that with earlier disease identification and intensive treatment of hyperglycemia, risk for microvascular and cardiovascular complications can be reduced.4–6 However, the spectrum of severity does not provide any finite threshold at which complications arise and medical treatment should begin. At the level of the earliest glucose abnormality (by current threshold recommendations), there is clearly disease at the cellular level, and complications can be seen even in those with nominally normal glucose levels.7–10 Despite this, the evidence for aggressive use of glucose-lowering medication is uncertain when the baseline glucose levels are not frankly abnormal.

Accordingly, screening recommendations by different governing bodies are in conflict. The American Diabetes Association (ADA) recommends screening of all patients > 45 years of age or who have risk factors, whereas other organizations such as the U.S. Preventative Services Task Force recommend more limited, targeted screening.11,12 Additionally, there are differences of opinion as to which diagnostic test represents the “gold standard.” The purpose of this review is to discuss available tests for type 2 diabetes, explain the evidence supporting different screening strategies, and describe the test characteristics of different diagnostic approaches.

Overview of Available Tests

Proposed tests for diabetes screening are numerous and vary from history- and anthropometric-based questionnaires to proteomics-based risk assessment.12–15 Although some of these tests might prove to be useful, the current preferred tests are limited to two groups: serum glucose–based tests and glycated proteins. Serum glucose–based tests include fasting plasma glucose (FPG), random plasma glucose (RPG), and the oral glucose tolerance test (OGTT). The most well-studied and useful glycated protein is A1C.

The 1997 ADA recommendations for diagnosis of diabetes focus on the FPG, whereas the World Health Organization (WHO) focuses on the OGTT. However, practicing physicians frequently employ other measures in addition to those recommended, including urinary glucose, RPG, and A1C. In one survey of primary care physicians and mid-level providers, 89% of providers reported using FPG for screening in some cases, 58% used RPG, and 42% used A1C. For confirmation of a diabetes diagnosis, 80% used A1C, and 64% used FPG. Only 7% of providers reported that they regularly use the OGTT to diagnose impaired glucose tolerance (IGT).16 A survey conducted by Ealovega et al.17 found that 95% of opportunistic screening was done by RPG, 3% by FPG, 2% by A1C, and < 1% by OGTT.

In addition to identification of the appropriate diagnostic test, another practical consideration is determination of the diabetes-defining threshold. Some studies have evaluated cut-points that are two standard deviations above normal, and others have used points that represent a natural break between normal and hyperglycemic peaks in populations with a high incidence of diabetes. However, the theoretical clinical ideal would be to estimate the point above which treatment specific to diabetic patients would signifi-
significantly lower the rate of diabetes complications.

Complication-based diagnosis studies have focused primarily on microvascular complications because they are specific to diabetes and easy to measure. There is an emerging body of literature about macrovascular complications, and there have also been studies looking at testing thresholds and mortality risk. These threshold determinations are discussed in the respective testing sections below.

Current Guidelines for Screening and Diagnosis

In its 2009 position statement, “Standards of Medical Care in Diabetes,” the ADA recommended screening with FPG to detect pre-diabetes or diabetes in nonpregnant adult patients who are >45 years of age or who are <45 years of age, have a BMI ≥25 kg/m², and have an additional risk factor for diabetes (Table 1). Repeat testing should be carried out at 3-year intervals.\textsuperscript{11}

The tests recommended for screening are the same as those for making the diagnosis, with the result that a positive screen is equivalent to a diagnosis of pre-diabetes or diabetes. Table 2 summarizes the current screening and diagnostic criteria of the ADA. The term “pre-diabetes” has been assigned to those considered to be at higher risk for developing diabetes. Pre-diabetes is diagnosed by having one or both of the following: 1) an FPG of 100–125 mg/dl, which is also referred to as impaired fasting glucose (IFG) or 2) a 2-hour, 75-g OGTT, with 2-hour plasma glucose levels of 140–199 mg/dl, which is also described as IGT. To get a diagnosis of diabetes, patients must satisfy one of the following criteria: 1) symptoms of diabetes (polyuria, polydipsia, and unexplained weight loss) AND an RPG ≥200 mg/dl, 2) an FPG ≥126 mg/dl, or 3) a 2-hour plasma glucose level ≥200 mg/dl during a 75-g OGTT.\textsuperscript{18}

In July 2009, the International Expert Committee recommended the

### Table 1. 2007 ADA Standards of Medical Care for Diabetes: Risk Factors that Indicate Screening for Diabetes

<table>
<thead>
<tr>
<th>Nonpregnant Adults</th>
<th>Pregnant Women</th>
<th>Children/Adolescents</th>
</tr>
</thead>
<tbody>
<tr>
<td>• Habitually physically inactive</td>
<td>• Age ≥ 25 years</td>
<td>• Family history of type 2 diabetes in first- or second-degree relatives</td>
</tr>
<tr>
<td>• First-degree relative with diabetes</td>
<td>• Prepregnancy BMI ≥ 25 kg/m²</td>
<td>• High-risk ethnic group*</td>
</tr>
<tr>
<td>• Member of a high-risk ethnic population*</td>
<td>• Member of a high-risk ethnic group*</td>
<td>• Signs of insulin resistance or conditions associated with insulin resistance (acanthosis nigricans, hypertension, dyslipidemia, or polycystic ovarian syndrome)</td>
</tr>
<tr>
<td>• Delivered a baby weighing &gt; 9 lb or diagnosed with gestational diabetes mellitus</td>
<td>• Known diabetes in a first-degree relative</td>
<td>• Maternal history of diabetes or gestational diabetes</td>
</tr>
<tr>
<td>• Hypertensive</td>
<td>• History of abnormal glucose tolerance</td>
<td></td>
</tr>
<tr>
<td>• HDL cholesterol &lt; 35 mg/dl and/or triglyceride level &gt; 250 mg/dl</td>
<td>• History of poor obstetric outcome</td>
<td></td>
</tr>
<tr>
<td>• Polycystic ovarian syndrome</td>
<td></td>
<td></td>
</tr>
<tr>
<td>• On previous testing had IFG or IGT</td>
<td></td>
<td></td>
</tr>
<tr>
<td>• Other clinical conditions associated with insulin resistance (i.e., acanthosis nigricans)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>• History of vascular disease</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

*\textit{African American, Hispanic, Native American, Asian, or Pacific Islander.}

### Table 2. ADA Diagnostic Criteria for Pre-Diabetes and Diabetes

<table>
<thead>
<tr>
<th>Test</th>
<th>IFG</th>
<th>IGT</th>
<th>Diabetes</th>
<th>Gestational Diabetes*</th>
</tr>
</thead>
<tbody>
<tr>
<td>FPG</td>
<td>100–125 mg/dl</td>
<td>Not defined</td>
<td>≥ 126 mg/dl</td>
<td>≥ 95 mg/dl</td>
</tr>
<tr>
<td>RPG</td>
<td>Not defined</td>
<td>Not defined</td>
<td>≥ 200 mg/dl</td>
<td>Not defined</td>
</tr>
<tr>
<td>75-g OGTT 2-hour plasma glucose</td>
<td>Not defined</td>
<td>140–199 mg/dl</td>
<td>≥ 200 mg/dl</td>
<td>Not defined</td>
</tr>
<tr>
<td>100-g OGTT</td>
<td>Not defined</td>
<td>Not defined</td>
<td>Not defined</td>
<td>1-hour: ≥ 180 mg/dl; 2-hour: ≥ 155 mg/dl; 3-hour: ≥ 150 mg/dl</td>
</tr>
<tr>
<td>A1C</td>
<td>Not defined**</td>
<td>Not defined**</td>
<td>≥ 6.5%</td>
<td>Not defined</td>
</tr>
</tbody>
</table>

*Must have two or more of the glucose-based abnormalities for diagnosis. **High risk, not specifically IGT or IFG, defined as A1C ≥ 6 and < 6.5%.
additional diagnostic criteria of an A1C result $\geq 6.5\%$ for diabetes. This committee suggested that use of the term “pre-diabetes” may be phased out but identified the range of A1C levels $\geq 6.0\%$ and $< 6.5\%$ to identify those at high risk for developing diabetes. The “high-risk” determination is qualified by the caveat that preventive measures can be initiated even in patients with lower A1C levels if other risk factors are present.19

Properties of each of the available tests are summarized in Table 3.

### Plasma Glucose–Specific Tests
Glucose-specific tests are often favored because they measure the pathophysiological outcome of diabetes (i.e., the amount of excess glucose in the blood). Additionally, they are inexpensive and are relatively easy to obtain during a clinical office visit. Here, we review the properties of FPG, RPG, and OGTT tests and the evidence for their use.

#### The FPG test
The FPG test is a simple plasma glucose measurement obtained after at least 8 hours of fasting (usually an overnight fast). It is an attractive option for screening and diagnosis because it is easy, inexpensive, and relatively risk-free. It has been the ADA test of choice for diagnosis of both pre-diabetes and diabetes. When compared directly, FPG has better intra-individual reproducibility than 2-hour post-load plasma glucose, with intra-individual coefficients of variation of 6.4–11.4\% for FPG versus 14.3–16.7\% for 2-hour plasma glucose.20

Practical downsides to the FPG are that it requires patients to fast, which can be imperfectly done, and testing may require an additional office visit for patients with afternoon appointments. Additionally, processing of the blood sample must be prompt (< 2 hours after collection), or the results can be falsely low.21 Finally, although the intra-individual stability is fair, FPG should be confirmed on a second occasion or with a second test to avoid false results.14

In 2003, expert committees lowered the FPG concentration diagnostic for diabetes from 140 to 126 mg/dl because of concern that

### Table 3. Summary of Diagnostic Test Characteristics for Use in Nonpregnant Adults

<table>
<thead>
<tr>
<th>Test</th>
<th>Pros</th>
<th>Cons</th>
<th>Recommendations for Use</th>
<th>Use in Screening</th>
</tr>
</thead>
<tbody>
<tr>
<td>Urinary glucose</td>
<td>Does not require blood sample; rapid processing time; inexpensive</td>
<td>Unable to measure glucose above the renal threshold; not fully quantitative</td>
<td>Not recommended</td>
<td>Not recommended</td>
</tr>
<tr>
<td>FPG</td>
<td>Single plasma glucose level; highly correlated with presence of complications; inexpensive</td>
<td>Patient must be fasting; potential for processing error; point measurement can be affected by short-term lifestyle changes</td>
<td>Diabetes diagnosis with FPG $\geq 126$ mg/dl; pre-diabetes diagnosis with FPG 100–126 mg/dl</td>
<td>Informal recommendation for follow-up testing with FPG $\geq 100$ mg/dl</td>
</tr>
<tr>
<td>RPG</td>
<td>Single glucose level; component of routine lab testing; inexpensive</td>
<td>Potential for processing error; point measurement can be affected by multiple factors (time since prior meal, short-term lifestyle changes, etc.)</td>
<td>Diabetes diagnosis with RPG $\geq 200$ mg/dl and symptoms of polyuria, polydipsia, and unintentional weight loss</td>
<td>Informal recommendation for follow-up test with RPG $\geq 130$</td>
</tr>
<tr>
<td>OGTT</td>
<td>Most sensitive test for IGT</td>
<td>Impractical in clinical setting, lower reproducibility than other diagnostic tests</td>
<td>Diagnosis of IGT with 2-hour plasma glucose $\geq 200$ mg/dl</td>
<td>Diagnostic criteria apply</td>
</tr>
<tr>
<td>Capillary glucose</td>
<td>Rapid test; does not require phlebotomy</td>
<td>Not standardized</td>
<td>Not recommended for diagnostic testing</td>
<td>Confirm any hyperglycemia with central laboratory glucose level or A1C</td>
</tr>
<tr>
<td>A1C</td>
<td>Gold standard for measuring glucose control; easy to obtain; does not require fasting; point-of-care testing available</td>
<td>Potential for nonglycemic causes of error; insensitive for IGT</td>
<td>A1C $\geq 6.5%$ diagnostic for diabetes; should be confirmed with a second A1C</td>
<td>High-risk group with A1C $\geq 6.0%$ and $&lt; 6.5%$. Patients with A1C $&lt; 6%$ and other risk factors are still eligible for preventive measures</td>
</tr>
</tbody>
</table>

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**Volume 27, Number 4, 2009 • CLINICAL DIABETES**
the previous level was insensitive for diagnosis of diabetes that manifests as postprandial hyperglycemia. At the same time, the FPG concentration for diagnosis of IFG was decreased to its current range of 100–125 mg/dl.22

Despite the new, lowered threshold for diagnosing diabetes, FPG continues to have only modest sensitivity. A Korean study23 evaluating the diabetes threshold found that an FPG ≥ 126 mg/dl detected only 55.7% of diabetic patients based on diagnosis by OGTT, with 100% specificity. FPG of > 110 mg/dl improved sensitivity of 85.2% but decreased specificity to 88.5% (area under the curve [AUC]: 0.944); this was the investigators’ proposed threshold for diabetes. A study of young African-American patients with pre-diabetes defined by the old ADA criteria found insensitivity of FPG for diagnosis of impaired glucose tolerance as compared to OGTT. FPG of 110 mg/dl detected only 27.4% of cases, whereas a complete OGTT detected 87.1%. The new FPG threshold did not perform much better, identifying only 28.9% of the impaired glucose tolerance cases.24

FPG is highly correlated with diabetes complications, particularly retinopathy. The Atherosclerosis Risk in Communities study examined patients not diagnosed with diabetes, divided into quartiles for FPG. The highest quartile of FPG, with levels > 113 mg/dl, had significantly higher rates of diabetic retinopathy compared to those with FPG ≤ 113 mg/dl.18 In the Mauritius study of a multiethnic population, an 18 mg/dl increase in FPG corresponded to an average odds ratio of 1.34 for development of retinopathy. However, the relationship was not totally linear; incident retinopathy was very low, with FPG results ≥ 108 mg/dl; steadily increased through 130 mg/dl; and then rose dramatically, with FPG results ≥ 135 mg/dl.25

The sensitivity of current FPG thresholds for detecting complication risk remains controversial. A study conducted with data from the Baltimore Longitudinal Study of Aging (BLSA) found that risk for mortality was increased for FPG levels > 110 mg/dl. The relative risk for mortality in the group having FPG levels of 126–139 mg/dl was 2.02. The authors systematically reviewed FPG and mortality data from other studies and found that the FPG range of 110–125 mg/dl is a zone of intermediate risk for mortality; relative risk in the BLSA data was 1.41. Data did not consistently support increased risk for mortality in the group having FPG levels of 100–110 mg/dl, with the relative risk for mortality in this study of 1.03.26

There is good evidence that FPG is a reliable predictor of diabetes complications at the current threshold for diagnosis, and studies examining FPG have underlined much of the current knowledge about the pathology of diabetes. However, clinicians should be aware that data supporting the threshold for pre-diabetes and its relationship to complications are not as clear. Additionally, studies evaluating glucose-lowering therapy, as well as the ADA guidelines for diabetes management, have focused on A1C rather than FPG as a measure of glucose control. Therefore, when making a diagnosis of diabetes or pre-diabetes with FPG, it is probably useful to also examine a baseline A1C to inform subsequent medical decision-making.

The RPG test
Advantages of the RPG (or “casual” plasma glucose) measurement are that it is easily obtained on the day of an office visit, does not require fasting, and is frequently included in a basic metabolic panel ordered for other purposes. It shares some of the practical downsides of the FPG in that it requires prompt processing and possibly an additional office visit for confirmatory testing.

The commonly held RPG threshold is ≥ 200 mg/dl, along with symptoms of polyuria, polydipsia, and unexplained weight loss to indicate a second test for confirmation of diagnosis. An RPG of 140–199 mg/dl is suggestive of pre-diabetes.18 Based on diagnosis by OGTT, an RPG ≥ 200 mg/dl is insensitive but has a specificity approaching 100%,27 which, in the setting of symptoms, is unlikely to lead to a false-positive diagnosis.

Despite the relatively established diagnostic threshold, it is not so obvious how to interpret RPG levels that are noted opportunistically on routine metabolic panels. In one large study comparing RPG with OGTT for screening, an RPG cut-off of 125 mg/dl was recommended to be cost-effective as an “index of concern.” At this level, RPG exhibited 93% specificity and 41% sensitivity. For identification of pre-diabetes, the specificity was still high, at 94%, but sensitivity was only 23%.28 A recent expert panel recommended a similar cut-off point, an RPG ≥ 130 mg/dl, which has a more balanced sensitivity (63%) and specificity (87%), based on diagnosis by OGTT.27

Impairing the overall utility of the RPG as a testing tool is the absence of data comparing it directly to rates of diabetes-specific complications. For this reason, its best clinical use is probably its presently recommended use; that is, as a rapid, any-time test with high specificity in symptomatic patients.

The OGTT
Oral glucose tolerance testing was introduced in 1922 and has been one of the diagnostic tests of choice for the
past 80 years. It is currently considered the gold standard for diabetes diagnosis, probably because of its longstanding use. It is recommended by WHO for diagnosis and is listed as an option in the ADA recommendations, but its use in the clinic remains controversial. It is the only way to formally diagnose IGT, which represents the fundamental pathophysiological defect in type 2 diabetes (i.e., the inability to respond to insulin release).

Regarding the diagnosis of diabetes, OGTT identifies about 2% more individuals than does FPG. OGTT has poor reproducibility compared to other glucose-based tests or A1C. OGTT also has obvious practical downsides, which are the required 8-hour fast before testing, commitment of nursing staff, the length of the test itself, and the necessity of an additional office visit.

The risk of diabetic microvascular complications has been the basis for determination of the threshold for 2-hour post-load plasma glucose. In a study of Pima Indians, Rushforth et al. examined the association between FPG and 2-hour plasma glucose and the presence of diabetic retinopathy and nephropathy. They determined that the optimal level for diagnosis, based on sensitivity and specificity, was an FPG of 136 mg/dl and a 2-hour plasma glucose level of 250 mg/dl. A second study found that for the current 2-hour plasma glucose cut-off of 200 mg/dl, sensitivity was 87.5% and specificity was 75.8% for presence of diabetic retinopathy. Finally, the Diabetes Epidemiology: Collaborative Analysis of Diagnostic Criteria in Europe study, conducted by a European diabetes epidemiology group, reviewed 13 prospective European cohort studies for risk of death according to the various glucose categories. This group showed that IGT in the absence of IFG (defined as 2-hour plasma glucose measurement of 140–199 mg/dl with FPG < 110 mg/dl) was associated with an increased risk of death. The hazard ratio was 1.8 in men and 2.6 in women.

The additional risk from postprandial hyperglycemia demonstrates that IGT is a clinically important entity, but it is controversial whether the OGTT is a reliable method of obtaining this diagnosis. Additionally, it is inconvenient, and physicians do not regularly order it.

**Capillary blood glucose meters**

Capillary glucose measurement is a popular method for determination of point glucose measurements at the time of office visits and is recommended for self-monitoring by patients. However, because of meter imprecision and the substantial differences among meters, their usefulness in screening and diagnosis is limited. Any glucose abnormalities detected with a capillary glucose meter should be confirmed with laboratory testing.

**A1C Testing**

A1C testing was first proposed as a measure of blood glucose control in 1976 and has developed into the standardized measure that is now broadly used for both research and clinical purposes. Its major practical advantages are that it can be obtained in both fasting and nonfasting states, and it represents average glucose control over a period of months rather than a single point value.

Although it had been widely accepted since the mid-1990s as the gold standard for therapy assessment and prognostication, it was only in June 2009 that the test was endorsed by the ADA as a first-line test for screening and diagnosis. At approximately the same time, the International Expert Committee released the formal recommendation of an A1C level ≥ 6.5% for diabetes diagnosis. The time lapse between acceptance of use for monitoring and acceptance of use for diagnosis was largely because of concern about the lack of standardization of the assay, which has been resolved through the National Glycohemoglobin Standardization Program, which started in 1996. Additionally, there was concern about errors caused by nonglycemic factors such as hemoglobinopathies; however, these are infrequent, and use of glucose-specific tests can confirm the diagnosis of diabetes in such cases.

Numerous studies have been done to determine the sensitivity and specificity of A1C testing using definitions based on FPG, 2-hour plasma glucose, and prevalence and incidence of complications. Two large analyses using the Third National Health and Nutrition Examination Survey (NHANES III) sample characterized A1C with respect to FPG. The first analysis found that 61% of patients with an FPG in the range of 110–125 mg/dl had normal A1C results (i.e., within the normal range of the assay used in the individual studies), along with 18.6% of patients with FPG between 126 and 139 mg/dl. Very few abnormal A1C results were seen in patients with an FPG < 110 mg/dl. In the second analysis, A1C was examined based on its standard deviation from the normal mean (5.13%) and a diagnosis of diabetes based on FPG ≥ 126 mg/dl. At a level of 1 SD above the mean (A1C of 5.6%), the sensitivity and specificity to detect diabetes were 83.4 and 84.4%, respectively. At 2 SD above the mean (A1C of 6.1%), the sensitivity and specificity were 63.2 and 97.4%, respectively. At 3 SD (A1C of 6.5%) and 4 SD (A1C of 7.0%) above the mean, specificity approached 100%, but sensitivity dropped to 42.8 and 28.3%, respec-
tively. Sensitivity to detect IFG was low (13.4%).

Buell et al. conducted a similar analysis with the 1999–2004 NHANES population. In these subjects, AIC measurement of 5.8% had the highest combination of sensitivity (86%) and specificity (92%) for diabetes diagnosis based on an FPG of 126 mg/dl. Bennett et al. conducted a systematic review of nine studies that measured both FPG and A1C, and, at the AIC cut-off of 6.1%, sensitivity was 78–81% and specificity was 79–84% to diagnose diabetes based on an FPG of 126 mg/dl.

To examine the utility of AIC in detection of IFG, which cannot be characterized by FPG, investigators compared AIC to 2-hour plasma glucose measurements. The Early Diabetes Intervention Program examined patients with pre-diabetes by FPG (100–125 mg/dl) but diabetes by 2-hour plasma glucose, and in these patients, detection of an AIC level > 6.1% increased the sensitivity of the FPG screen from 45 to 61%. In a group of Pima Indians, A1C had a sensitivity of 84.7%. A meta-analysis combined data from 10 observational studies of type 2 diabetes and found the pooled relative risk for cardiovascular disease was 1.18 per 1% increase in AIC in type 2 diabetes. In a recent review of data from the NHANES III, Saydah et al. found that higher levels of GHb across diabetic and nondiabetic patients were associated with increased risk of mortality from all causes (relative hazard 2.59), heart disease (3.38), and cancer (2.64). For adults with diagnosed diabetes (using total GHb as the test), having a GHb ≥ 8 vs. < 6% had a relative hazard of 1.68 for all-cause mortality and 2.48 for heart disease mortality.

As with the glucose-based tests, there is no finite threshold of AIC at which normality ends and diabetes begins. The International Expert Committee has elected to recommend a cut point for diabetes diagnosis that emphasizes specificity, commenting that this “balanced the stigma and cost of mistakenly identifying individuals as diabetic against the minimal clinical consequences of delaying the diagnosis in someone with an AIC level < 6.5%.”

Capillary blood A1C testing

Capillary blood A1C measurement, also called “point-of-care” (POC) AIC testing, is becoming a popular method for office-based monitoring of glucose control. In a study of 597 subjects (79% female and 96% African American), rapid POC A1C measurement resulted in more frequent intensification of the diabetes regimen when A1C was ≥ 7%. In the same study, in the 275 patients with two follow-up visits, AIC fell significantly in the rapid-test group (from 8.4 to 8.1%) but not in the routine group (from 8.1 to 8.0%).

In a study of correlation between a specific POC A1C method (the DCA 2000) and a standardized laboratory value from the Diabetes Control and Complications Trial, the two were found to be similar, although the DCA 2000 measured slightly higher values. Newer POC instruments are now available, and although more studies are needed to confirm reliability with standardized assays, the POC method seems promising for convenient monitoring of glucose control.

Conclusions

Type 2 diabetes is a prevalent disease with morbidity and mortality, and diagnosis is essential so that appropriate treatment can be provided. Office-based testing is recommended and can be conveniently undertaken with glucose-based tests, along with AIC testing in appropriate patients.

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