VOLUME - VI
ISSUE - XXXIX
MAY / JUN 2010

The Crux

BIMONTHLY FORUM FOR THE LABORATARIANS

CONTENTS

Editorial

This is perhaps for the first time that a complete issue is being dedicated to one disorder. The issue is delving around TYPE 2 DIABETES MELLITUS. Type 2 diabetes mellitus is a disorder characterized by hyperglycemia and associated with microvascular (ie, retinal, renal, possibly neuropathic), macrovascular (ie, coronary, peripheral vascular), and neuropathic (ie, autonomic, peripheral) complications. Unlike patients with type 1 diabetes mellitus, patients with type 2 are not absolutely dependent upon insulin for life, even though many of them are ultimately treated with insulin. Over the years, diagnostic criteria have changed as our knowledge about the disease process improved. Now a days Classic symptoms of diabetes mellitus (polyuria, polydipsia, polyphagia and weight loss and random plasma glucose $\geq$200 mg/dL OR any two fasting plasma glucose levels $\geq$ 126 mg/dL OR Two hour post glucose load (75g) plasma glucose $\geq$200 mg/dL, and confirmed by repeat test OR A1c level of 6.5 or more are considered diagnostic. New terminologies have evolved like MODY i.e., Maturity Onset Diabetes of the Young. The pathophysiology of Type 2 diabetes is 1) increased hepatic glucose production, 2) decreased insulin secretion, or 3) decreased peripheral glucose uptake. Increased cardiovascular risk appears to begin prior to the development of frank hyperglycemia, presumably because of the effects of insulin resistance. Stern in 1996 and Haffner and D'Agostino in 1999 developed the "ticking clock" hypothesis of complications, asserting that the clock starts ticking for microvascular risk at the onset of hyperglycemia, while the clock starts ticking for macrovascular risk at some antecedent point, presumably with the onset of insulin resistance. Type 2 diabetes mellitus is less common in non-Western countries where the diet contains fewer calories and caloric expenditure on a daily basis is higher. However, as people in these countries adopt Western lifestyles, weight gain and type 2 diabetes mellitus are becoming virtually epidemic. Diabetes mellitus is one of the leading causes of morbidity and mortality worldwide because of its role in the development of optic, renal, neuropathic, and cardiovascular disease. The prevalence of type 2 diabetes mellitus varies widely among various racial and ethnic groups. Type 2 diabetes mellitus is becoming virtually a pandemic. The risk of retinopathy and nephropathy appears to be greater in coloured people world over. Complete clinico-diagnostic approach as related to Type 2 diabetes mellitus is presented in the subsequent pages under the DISEASE DIAGNOSIS section.

The INTERPRETATION segment outlines all latest specific changes in the 2010 Clinical Practice Recommendations as relevant to Type 2 Diabetes Mellitus. Clinical Implications along with Clinical Context are summarized for professional use.

One of the most sensitive tests for evaluating diabetes in retrospective context is HbA1c, all trouble shooting aspects as related to Turbidimetric estimation of HbA1c are covered under the TROUBLESHOOTING portion.

Lastly, BOUQUET can never be forgotten. It is very much there but with different fragrances and hues.

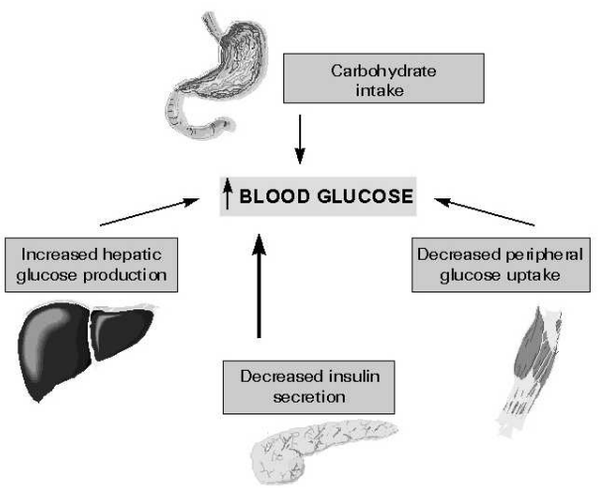
DISEASE DIAGNOSIS

TYPE 2, DIABETES MELLITUS

Background

Type 2 diabetes mellitus is a group of disorders characterized by hyperglycemia and associated with microvascular (ie, retinal, renal, possibly neuropathic), macrovascular (ie, coronary, peripheral vascular), and neuropathic (ie, autonomic, peripheral) complications. Unlike patients with type 1 diabetes mellitus, patients with type 2 are not absolutely dependent upon insulin for life, even though many of them are ultimately treated with insulin.

Pathophysiology: Hyperglycemia results from lack of endogenous insulin, which is either absolute, as in type 1 diabetes mellitus, or relative, as in type 2 diabetes mellitus. Relative insulin deficiency usually occurs because of resistance to the actions of insulin in muscle, fat, and the liver and an inadequate response by the pancreatic beta cell. Insulin resistance, which has been attributed to elevated levels of free fatty acids in plasma, leads to decreased glucose transport in muscle, elevated hepatic glucose production, and increased breakdown of fat. The genetics of type 2 diabetes are complex and not completely understood, but presumably this disease is related to multiple genes (with the exception of maturity-onset diabetes of the young [MODY]). Evidence supports inherited components for pancreatic beta-cell failure and insulin resistance. Considerable debate exists regarding the primary defect in type 2 diabetes mellitus. Most patients have insulin resistance and some degree of insulin deficiency. However, insulin resistance per se is not the sine qua non for type 2 diabetes mellitus because many people with insulin resistance (particularly those who are obese) do not develop glucose intolerance. Therefore, insulin deficiency is necessary for the development of hyperglycemia. Insulin concentrations may be high, yet inappropriately low for the level of glycemia. MODY is associated with autosomal dominant inheritance and is characterized by onset in at least 1 family member younger than 25 years, absence of autoantibodies, correction of fasting hyperglycemia without insulin for at least 2 years, and absence of ketosis. At least 6 genetically different types of MODY have been described. Some patients ultimately require insulin to control glycemia. Variants in 2 of the genes associated with MODY (HNF-1alpha and, to a lesser extent, HNF-4alpha) have been shown to predict future type 2 diabetes. Presumably, the defects of type 2 diabetes mellitus occur when a diabetogenic lifestyle (ie, excessive caloric intake, inadequate caloric expenditure, obesity) is superimposed upon a susceptible genotype. The body mass index at which excess weight increases risk for diabetes varies with different racial groups. For example, compared with persons of European ancestry, persons of Asian ancestry are at increased risk for diabetes at lower levels of overweight. In addition, an in utero environment resulting in low birth weight may predispose some individuals to develop type 2 diabetes mellitus. A simplified scheme for the pathophysiology of abnormal glucose metabolism in type 2 diabetes mellitus is depicted in the image below.



Simplified scheme for the pathophysiology of type 2 diabetes mellitus.

Hyperglycemia appears to be the determinant of microvascular and metabolic complications. However, glycemia is much less related to macrovascular disease. Insulin resistance with concomitant lipid (ie, small dense low-density lipoprotein [LDL] particles, low high-density lipoprotein-cholesterol [HDL-C] levels, elevated triglyceride-rich remnant lipoproteins) and thrombotic (ie, elevated type-1 plasminogen activator inhibitor [PAI-1], elevated fibrinogen) abnormalities, as well as conventional atherosclerotic risk factors (eg, family history, smoking, hypertension, elevated low-density lipoprotein-cholesterol [LDL-C], low HDL-C), determine cardiovascular risk. Increased cardiovascular risk appears to begin prior to the development of frank hyperglycemia, presumably because of the effects of insulin resistance. Stern in 1996 and Haffner and D'Agostino in 1999 developed the "ticking clock" hypothesis of complications, asserting that the clock starts ticking for microvascular risk at the onset of hyperglycemia, while the clock starts ticking for macrovascular risk at some antecedent point, presumably with the onset of insulin resistance.

Frequency: International: Type 2 diabetes mellitus is less common in non-Western countries where the diet contains fewer calories and caloric expenditure on a daily basis is higher. However, as people in these countries adopt Western lifestyles, weight gain and type 2 diabetes mellitus are becoming virtually epidemic.

Mortality/Morbidity: Diabetes mellitus is one of the leading causes of morbidity and mortality worldwide because of its role in the development of optic, renal, neuropathic, and cardiovascular disease. These complications, particularly cardiovascular disease (~50-75% of medical expenditures), are the major sources of expenses for patients with diabetes mellitus. Approximately two thirds of people with diabetes die from heart disease or stroke. Men with diabetes face a 2-fold increased risk for coronary heart disease, and women have a 3-to-4-fold increased risk. Approximately 20% of Medicare funds are spent on these patients. Diabetes mellitus is the leading cause of blindness in working-age adults on a worldwide canvas; diabetic retinopathy accounts for lakhs of newly blind persons every year. The National Eye Institutes estimate that laser surgery and appropriate follow-up care can reduce the risk of blindness from diabetic retinopathy by 90%.10 Diabetes mellitus is the leading cause of end-stage renal disease (ESRD), accounting for 44% of new cases. Diabetes mellitus is the leading cause of nontraumatic lower limb amputations world overs, with a 15- to 40-fold increase in risk over that of the nondiabetic population. Iannually lakhs of nontraumatic lower limb amputations are performed related to neuropathy and vasculopathy.

Race: The prevalence of type 2 diabetes mellitus varies widely among various racial and ethnic groups. Type 2 diabetes mellitus is becoming virtually a pandemic. The risk of retinopathy and nephropathy appears to be greater in coloured people world over.

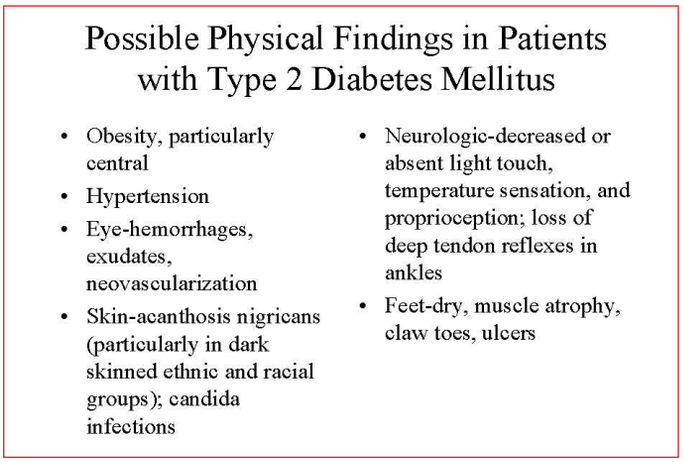
Sex: Type 2 diabetes mellitus is slightly more common in older women than men.

Age: While type 2 diabetes mellitus traditionally has been thought to affect individuals older than 40 years, it is being recognized increasingly in younger persons, particularly in highly susceptible racial and ethnic groups and the obese. In some areas, more type 2 than type 1 diabetes mellitus is being diagnosed in prepubertal children, teenagers, and young adults. Virtually all cases of diabetes mellitus in older individuals are type 2.

Clinical: History: While a diagnosis of diabetes mellitus is readily entertained when a patient presents with classic symptoms (ie, polyuria, polydipsia, polyphagia, weight loss), most patients with type 2 diabetes mellitus are asymptomatic for years. Other symptoms that might suggest hyperglycemia include blurred vision, lower extremity paresthesias, or yeast infections, particularly balanitis in men. However, the asymptomatic state does not mean that hyperglycemia is not affecting the individual. The possible presence of diabetes mellitus should be considered in obese patients, patients with a first-degree relative with type 2 diabetes mellitus, members of high-risk ethnic groups, women with a previous delivery of a large infant (>9 lb) or with a history of gestational diabetes mellitus, patients with hypertension, or patients with high triglycerides (>250 mg/dL) or low HDL-C (<35 mg/dL). While the physicians do not recommend routine screening for diabetes, targeted screening may be useful. For example, in one study of patients admitted to the hospital with acute coronary syndrome, none of whom were known to have diabetes, admission and fasting plasma glucose testing identified diabetes in 27%. Because polycystic

ovary disease is an insulin-resistant state, screening these women may be warranted. Whether at-risk persons should be screened for prediabetes is unclear at present. The therapy would generally be lifestyle changes to facilitate weight loss and improve cardiovascular fitness, and in virtually all cases, this would be the recommendation for such patients without a measured glucose value.

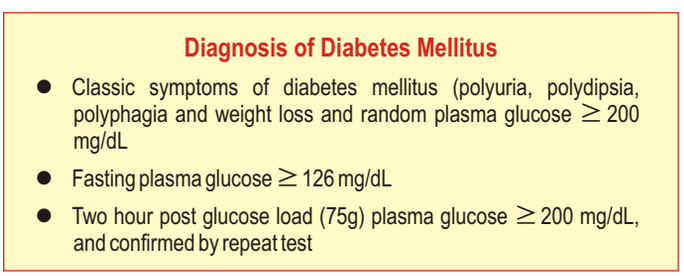
Physical: Early in the course of diabetes mellitus, the physical examination findings are likely to be unrevealing. However, ultimately, end-organ damage may be observed. Potential findings are listed in the image below.



Possible physical examination findings in patients with type 2 diabetes mellitus.

Causes: Superimposition of caloric excess (typically, high intake and low expenditure) on a susceptible genotype appears to cause type 2 diabetes mellitus. A large, population-based, prospective study has shown that an energy-dense diet may be a risk factor for the development of diabetes that is independent of baseline obesity. Diabetes mellitus may be caused by other conditions. Secondary diabetes may occur in patients taking glucocorticoids or when patients have conditions that antagonize the actions of insulin (eg, Cushing syndrome, acromegaly, pheochromocytoma).

Workup: Laboratory Studies: The American Diabetes Association criteria for the diagnosis of diabetes are listed in the image below. Most commonly, the diagnosis is made when the health care provider discovers either fasting plasma glucose (FPG) greater than or equal to 126 mg/dL on 2 occasions or random glucose greater than or equal to 200 mg/dL and classic symptoms of diabetes mellitus (ie, polyuria, polydipsia, polyphagia, weight loss).



Diagnostic criteria (American Diabetes Association) for diabetes mellitus type 2.

Plasma glucose is determined using blood drawn into a gray-top (sodium fluoride) tube, which inhibits red blood cell glycolysis immediately. A serum glucose measurement (commonly obtained on chemistry panels, using a red- or speckled-top tube) may be significantly lower than will a plasma glucose measurement. Capillary whole blood measurements are not recommended for the diagnosis of diabetes mellitus. The noted values for fasting glucose measurements are based on the level of glycemia at which retinopathy, a fairly pathognomonic diabetic complication, appears. (However, evidence suggests that retinopathy may occur even in prediabetes.) Fasting glucose measurements are not as predictive for indicating macrovascular risk as post-glucose load values. However, there are no formal recommendations for using glucose tolerance tests for this purpose. These criteria are a better predictor of increased macrovascular risk than the American Diabetes Association's current intermediate category of impaired fasting glucose (IFG) or prediabetes. Presumably, patients with IFG are at increased risk for development of diabetes mellitus, but their risk for macrovascular disease does not appear to be the same as for patients with IGT (which is about the same as patients with frank type 2 diabetes mellitus). FPG <140 mg/dL, and at 2 hours after a 75-g glucose load. Plasma glucose >140 mg/dL to <200 mg/dL with 1 intervening plasma glucose value >200 mg/dL.

Hemoglobin A1c (HbA1c or A1c), or glycosylated hemoglobin (GHb), measurements have previously not been considered useful for the diagnosis of diabetes mellitus because of a lack of international standardization and an insensitivity for the detection of milder forms of glucose intolerance. Changes in standardization that could affect the actual values that individual laboratories generate have also been a concern. In a 2009 report, however, an international expert committee appointed by the ADA, the European Association for the Study of Diabetes, and the International Diabetes Association recommended HbA1c assay for diagnosing type 1 and type 2 diabetes mellitus. (In the case of type 1 diabetes mellitus, however, the committee recommended using the test only when the condition is suspected but the classical symptoms of type 1 diabetes mellitus—polyuria, polydipsia, polyphagia, a random glucose level of 200 mg/dL, and unexplained weight loss—are absent.) The committee cited the following advantages of HbA1c testing over glucose measurement: Captures long-term glucose exposure, Has less biologic variability, Does not require fasting or timed samples, Is currently used to guide management decisions. The committee's recommendation for a diagnosis of diabetes mellitus is an HbA1c level of 6.5% or higher, with confirmation from repeat testing (unless clinical symptoms are present and the glucose level is >200 mg/dL). Glucose measurement should remain the choice for diagnosing pregnant women or if HbA1c assay is unavailable. HbA1c measurements are the criterion standard for monitoring long-term glycemic control and reflect glycemia for the previous 3 months. Whether HbA1c or GHb assays are superior for measuring glycemic control is debatable. Hemoglobinopathies can affect both measurements. Because the Diabetes Control and Complications Trial (DCCT) and the United Kingdom Prospective Diabetes Study (UKPDS), as well as the American Diabetes Association Standards of Care, refer to HbA1c measurements, this article refers to HbA1c as the standard for glycemic control. Using GHb measurements is acceptable, but these values are 1-2% higher than HbA1c concentrations. When using GHb, an available conversion factor to HbA1c for the assay utilized is helpful. Screening urine microalbumin measurements is recommended yearly in all patients with diabetes. Performing an albumin-to-creatinine ratio is probably easiest. If abnormal (ie, >30 mg/g), a quantitation on a timed urine specimen (ie, overnight, 10 hours, or 24 hours) should be performed. Normal urine albumin excretion is defined as less than 30 mg/d. Microalbuminuria is defined as 30-300 mg/d (20-200 mcg/min). Because of wide variability among patients, confirm persistent microalbuminuria on at least 2 of 3 samples over 3-6 months. Greater values can be detected by standard protein dipstick screening and are considered macroproteinuria. Unlike type 1 diabetes mellitus, in which microalbuminuria is a good indicator of early kidney damage, microalbuminuria is a common finding (even at diagnosis) in type 2 diabetes mellitus and is a risk factor for macrovascular (especially coronary heart) disease. It is a weaker predictor for future kidney disease in type 2 diabetes mellitus. Measuring concentrations of insulin or C-peptide (a fragment of proinsulin that serves as a marker for insulin secretion) rarely is necessary to diagnose type 2 diabetes mellitus or differentiate type 2 diabetes from type 1 diabetes mellitus. Insulin levels generally are high early in the course of type 2 diabetes mellitus and gradually wane over time. Stimulated C-peptide concentrations (after a standard meal challenge) are somewhat preserved until late in the course of type 2 diabetes mellitus. Absence of a C-peptide response to carbohydrate ingestion may indicate total beta cell failure. Antibodies to insulin, islet cells, or glutamic acid decarboxylase (GAD) are absent in type 2 diabetes mellitus. Latent autoimmune diabetes of adults, or LADA, is a form of slow-onset type 1 diabetes that occurs in middle-aged (usually white) adults. It can be differentiated from type 2 diabetes by measuring anti-GAD65 antibodies. Such patients may respond to insulin secretagogues for a brief period (months).

Treatment: Medical Care: The goals in caring for patients with diabetes mellitus include the elimination of symptoms; microvascular (ie, eye and kidney disease) risk reduction through control of glycemia and blood pressure (BP); macrovascular (ie, coronary, cerebrovascular, peripheral vascular) risk reduction through control of lipids and hypertension, smoking cessation, and aspirin therapy; and metabolic risk reduction through control of glycemia. Such care requires appropriate goal setting, regular complications monitoring, dietary and exercise modifications, medications, appropriate self-monitoring of blood glucose (SMBG), and laboratory assessment. Focus on glucose alone does not provide adequate treatment for patients with diabetes mellitus. Treatment involves multiple goals (ie, glycemia, lipids, BP).

Glycemic goals: Implications of the UKPDS: The UKPDS was a landmark study for the care of patients with type 2 diabetes mellitus, confirming the importance of glycemic control in reducing the risk for microvascular complications and refuting previous data implicating increased macrovascular disease risk with sulfonylureas or insulin. Major findings of the study are displayed in the images below. Significant implications include the following: Microvascular complications (predominantly the need for laser photocoagulation on retinal lesions) are reduced by 25% when median HbA1c is 7% compared with 7.9%. A continuous relationship exists between glycemia and microvascular complications, with a 35% reduction in risk for each 1% decrement in HbA1c. A glycemic threshold (above the upper limit of normal for HbA1c) below which risk for microvascular disease is eliminated does not appear to exist. Glycemic control has minimal effect on macrovascular disease risk. Excess macrovascular risk appears to be related to conventional risk factors such as dyslipidemia and hypertension. Sulfonylureas and insulin therapy do not increase macrovascular disease risk. Metformin reduces macrovascular risk in patients who are obese. Vigorous BP control reduces microvascular and macrovascular events. Beta blockers and angiotensin-converting enzyme (ACE) inhibitors appear to be equally efficacious. Glycemic goal setting and achieving glycemic goals: The DCCT and UKPDS provide ample evidence that glycemic control is paramount in reducing microvascular complications. Unless the risk outweighs the benefit, an HbA1c target of less than 7% is appropriate. Some organizations (eg, the American Association of Clinical Endocrinologists, the International Diabetes Federation) recommend a glycemic target of HbA1c less than 6.5%. The practitioners should aim for the lowest possible HbA1c that does not cause undue harm. The limiting factor is almost always risk for hypoglycemia. Unfortunately, some practitioners and their patients pursue a particular HbA1c value despite uncertain benefit (eg, patients with advanced complications) or unacceptable risk (eg, hypoglycemia unawareness, elderly patients, patients with other major systemic disease with significant risk for side effects [eg, coma, seizures, falling and breaking a hip]). Situations with an unfavorable risk-benefit ratio for intensive blood glucose lowering include advanced age, significant concomitant disease, and advanced complications. Decisions about glycemic management are generally made on the basis of HbA1c measurements performed quarterly (possibly less often in patients with adequate control through lifestyle measures alone) and the results of SMBG. If a total GHb measurement is used, the actual number is 1-2% higher, but the laboratory should provide a correlation with actual HbA1c values.

Complications monitoring: The American Diabetes Association recommends initiation of complications monitoring at the time of diagnosis of diabetes mellitus. This regimen should include yearly dilated eye examinations, yearly microalbumin checks, and foot examinations at each visit.

Self-monitoring of blood glucose: Daily SMBG is important for patients treated with insulin or insulin secretagogues to monitor for and prevent hypoglycemia and optimize the treatment regimen. The optimal frequency of SMBG for patients with type 2 diabetes is unresolved, but it should be sufficient to facilitate reaching glucose goals. Most practitioners utilize no or minimal SMBG in patients using lifestyle changes alone or agents that do not cause hypoglycemia (eg, metformin, glitazones, glucosidase inhibitors).

Laboratory monitoring: Because diabetes mellitus is a multisystem disease, focusing solely on blood sugar is inadequate. The image below lists appropriate laboratory parameters in the global assessment of patients with type 2 diabetes mellitus. Obviously, patients with abnormalities need more frequent monitoring to guide therapeutic interventions. Drug-specific monitoring is also necessary (eg, serum creatinine for metformin, serum transaminases for glitazones).

Laboratory Monitoring of Patients with Type 2 Diabetes Mellitus

Test	*HbA1c*	*FLP*	*Scr*	*Umab*	*ECG*
Type 2 DM-insulin treated	Every 3 months	Yearly	Yearly	Yearly	Baseline
Type 2 DM-no insulin	Every 3-6 months	Yearly	Yearly	Yearly	Baseline

Laboratory monitoring guidelines for patients with type 2 diabetes mellitus.

Intercurrent medical illness: Patients with intercurrent illness become more insulin resistant because of the effects of increased counter-regulatory (ie, anti-insulin) hormones. Therefore, despite decreased nutritional intake, glycemia may worsen. Patients on oral agents may need transient therapy with insulin to achieve adequate glycemic control. In patients who require insulin, scheduled doses of insulin, as opposed to sliding scale insulin, are far more effective in achieving glycemic control. If patients taking metformin have any illness that leads to dehydration or hypoperfusion, the drug should be temporarily discontinued because of a possible increased risk of lactic acidosis.

Critical illness: Standard practice in intensively ill patients has been to provide tight glycemic control through intensive insulin therapy. Research evidence, however, has called this practice into question. A meta-analysis found that in critically ill adult patients, tight glucose control is associated with an increased risk of hypoglycemia but not with significantly reduced hospital mortality. A large, international, randomized trial among adults treated in an intensive care unit (ICU) found that intensive glucose control (target, 81-108 mg/dL) resulted in higher mortality than did a blood glucose target of 180 mg/dL or less. In the case of cardiac ischemia, the beneficial effects of insulin therapy may be due to reduction in free fatty acids.

Surgery: Surgical patients may experience worsening of glycemia for reasons similar to those listed above for intercurrent medical illness. Patients on oral agents may need transient therapy with insulin to maintain blood glucose at approximately 100-180 mg/dL. In patients who require insulin, scheduled doses of insulin, as opposed to sliding scale insulin, are far more effective in controlling glucose. Intensive glucose control in surgical ICU patients appears to reduce risk of septicemia, but as with other critically ill patients, this may come at the cost of increased risk of hypoglycemia. For patients who can eat soon after surgery: The time-honored approach of administering half the usual morning dose of neutral protamine Hagedorn (NPH) insulin with 5% dextrose in the IV is acceptable, with resumption of scheduled insulin (perhaps at reduced doses) within the first 1-2 days. Patients receiving insulin glargine can often receive their usual dose if they are given intravenous glucose during surgery with appropriate intraoperative and postoperative monitoring of glucose. Oral antidiabetic agents can be restarted when the patient is stable and eating. Insulin secretagogues should be used with caution in the hospital since food intake may be interrupted by diagnostic tests and procedures. Metformin may have to be started at a lower dose and gradually titrated to full dose due to gastrointestinal side effects. Since glitazones have such a long biologic effect, their omission in the hospital is usuallyinconsequential. The role of incretins in the hospital has not yet been defined. For patients who require more prolonged periods without oral nutrition and for major surgery, such as coronary artery bypass grafting and major abdominal surgery: Constant infusion intravenous insulin is preferred. Discontinue metformin temporarily after any major surgery until the patient is clearly hemodynamically stable and normal renal function is documented. The practice of discontinuing metformin for at least 48 hours in this situation until proof of normal renal function is established is sound.

Pregnancy: The increased prevalence of type 2 diabetes mellitus in women of reproductive age has brought new prominence to the need for prepregnancy planning in this population. Insulin is the only generally accepted pharmacologic therapy for women with diabetes mellitus who are contemplating pregnancy. For women with diabetes mellitus controlled by lifestyle measures alone, conversion to insulin as soon as the pregnancy is confirmed is appropriate. For women with polycystic ovary disease who are receiving insulin sensitizer therapy and who subsequently ovulate and become pregnant, conversion to insulin is often mandatory as soon as pregnancy is confirmed. Metformin is used during pregnancy in many countries. Insulin is the only acceptable pharmacologic therapy during pregnancy for women with established diabetes mellitus.

INTERPRETATION

TYPE 2 DIABETES MELLITUS

LATEST CLINICO-DIAGNOSTIC RECOMMENDATIONS

The American Diabetes Association (ADA) revised clinical practice recommendations for diabetes diagnosis promote hemoglobin A1c (A1c) as a faster, easier diagnostic test that could help reduce the number of undiagnosed patients and better identify patients with prediabetes. The new recommendations are published December 29 in the January 2010 supplement of Diabetes Care. "We believe that use of the A1c, because it doesn't require fasting, will encourage more people to get tested for type 2 diabetes and help further reduce the number of people who are undiagnosed but living with this chronic and potentially life-threatening disease," Richard M. Bergenstal, MD, ADA president-elect of medicine & science, said in a news release. "Additionally, early detection can make an enormous difference in a person's quality of life. Unlike many chronic diseases, type 2 diabetes actually can be prevented, as long as lifestyle changes are made while blood glucose levels are still in the pre-diabetes range." The A1c test, which measures average blood glucose levels for a period of up to 3 months, was previously used only to evaluate diabetic control with time. An A1c level of approximately 5% indicates the absence of diabetes, and according to the revised evidence-based guidelines, an A1c score of 5.7% to 6.4% indicates prediabetes, and an A1c level of 6.5% or higher indicates the presence of diabetes. For optimal diabetic control, the recommended ADA target for most people with diabetes is an A1c level no greater than 7%. It is hoped that achieving this target would help prevent serious diabetes-related complications including nephropathy, neuropathy, retinopathy, and gum disease. Unlike fasting plasma glucose testing and the oral glucose tolerance test, A1c testing does not require overnight fasting. Compliance with screening may therefore be improved through use of the A1c test, which can be determined from a single nonfasting blood sample.

Recommendation Changes for 2010

Specific changes in the 2010 Clinical Practice Recommendations are as follows: A section on diabetes related to cystic fibrosis has been added to "Standards of Medical Care in Diabetes." New evidence has shown that early diagnosis of cystic fibrosis-related diabetes and aggressive treatment with insulin have narrowed the gap in mortality between patients with cystic fibrosis with and without diabetes and have eliminated the sex difference in mortality rates. New recommendations for the clinical management of cystic fibrosis-related diabetes, based on a 2009 consensus conference, will be published in 2010 in a consensus report. Revision of the section "Diagnosis of Diabetes" now includes the use of the A1c level for diabetes diagnosis, with a cutoff point of 6.5%. The section formerly named "Diagnosis of Pre-diabetes" is now named "Categories of Increased Risk for Diabetes." Categories suggesting an increased risk for future diabetes now include an A1c range of 5.7% to 6.4%, as well as impaired fasting glucose and impaired glucose tolerance levels. Revisions to the section "Detection and Diagnosis of GDM [Gestational Diabetes Mellitus]" now include a discussion of possible future changes in this diagnosis, according to international consensus. Screening recommendations for gestational diabetes are to use risk factor analysis and an oral glucose tolerance test, if appropriate. Women diagnosed with gestational diabetes should be screened for diabetes 6 to 12 weeks postpartum and should have subsequent screening for the development of diabetes or prediabetes. Extensive revisions to the section "Diabetes Self-Management Education" are based on new evidence. Goals of diabetes self-management education are to improve adherence to standard of care, to educate patients regarding appropriate glycemic targets, and to increase the percentage of patients achieving target A1c levels. Extensive revisions to the section "Antiplatelet Agents" now reflect evidence from recent trials suggesting that in moderate- or low-risk patients, aspirin is of questionable benefit for primary prevention of cardiovascular disease. The revised recommendation is to consider aspirin treatment as a primary prevention strategy in patients with diabetes who are at increased cardiovascular risk, defined as a 10-year risk greater than 10%.

Patients at increased cardiovascular risk include men older than 50 years or women older than 60 years with at least 1 additional major risk factor. Fundus photography may be used as a screening strategy for retinopathy, as described in the section "Retinopathy Screening and Treatment." However, although high-quality fundus photographs detect most clinically significant diabetic retinopathy, they should not act as a substitute for an initial and dilated comprehensive eye examination. Retinal examinations should be carried out annually or at least every 2 to 3 years among low-risk patients with normal eye examination results in the past. Extensive revisions to the section "Diabetes Care in the Hospital" now question the benefit of very tight glycemic control goals in critically ill patients, based on new evidence. Extensive revisions to the section "Strategies for Improving Diabetes Care" are based on newer evidence. Successful strategies to improve diabetes care, which have resulted in improved process measures such as measurement of A1c levels, lipid levels, and blood pressure, include the following: Delivery of diabetes self-management education. Adoption of practice guidelines developed with participation of healthcare professionals and having them readily accessible at the point of service. Use of checklists mirroring guidelines, which help improve adherence to standards of care. Systems changes, including providing automated reminders to healthcare professionals and patients and audit and feedback of process and outcome data to providers. Quality improvement programs, in which continuous quality improvement or other cycles of analysis and intervention are combined with provider performance data. Practice changes, which may include access to point-of-care A1c testing, scheduling planned diabetes visits, and clustering dedicated diabetes visits into specific times. Tracking systems with either an electronic medical record or patient registry to improve adherence to standards of care. Availability of case or (preferably) care management services using nurses, pharmacists, and other nonphysician healthcare professionals following detailed algorithms under physician supervision. "The most successful practices have an institutional priority for quality of care, involve all of the staff in their initiatives, redesign their delivery system, activate and educate their patients, and use electronic health record tools," the guidelines suggest. "It is clear that optimal diabetes management requires an organized, systematic approach and involvement of a coordinated team of dedicated health care professionals working in an environment where quality care is a priority."

The entire ADA Clinical Practice Recommendations are presented below:

Clinical Context:

Diabetes is one of the most common chronic conditions seen in practice. Yet for all of the patients known to have diabetes, many individuals with high serum glucose levels remain undiagnosed. With this in mind, the ADA has updated its recommendations for the diagnosis of type 2 diabetes, which include A1c level as a means to diagnose diabetes for the first time. Thus, the new diagnostic criteria for type 2 diabetes are as follows: An A1c level of 6.5% or more. Fasting plasma glucose level of 126 mg/dL or more. A 2-hour plasma glucose level of 200 mg/dL or more after a 75-g oral glucose tolerance test. A random plasma glucose level of 200 mg/dL or more in a patient with symptoms of hyperglycemia. In the absence of symptoms of hyperglycemia, the first 3 options listed should be confirmed with repeated testing. The potential benefits of using A1c level in the initial diagnosis of type 2 diabetes include the ability to perform the test in a nonfasting state and less perturbation of the test result because of stress and illness. Patients with an A1c level between 5.7% and 6.4% should be considered to have prediabetes and should receive appropriate counseling on therapeutic lifestyle change. Other recommendations from the ADA, particularly updates creating new guidelines, are summarized in the Study Highlights are given below.

Study Highlights:

Testing for type 2 diabetes should be considered in asymptomatic individuals with overweight or obesity, or other risk factors for diabetes. Testing may begin at age 45 years, and subsequent testing may be continued at 3-year intervals. All pregnant women, except those at low risk for gestational diabetes, should be screened with an oral glucose tolerance test at 24 to 28 weeks of gestation. Low-risk status is defined by age younger than 25 years, normal prepregnancy weight, ethnicity in a group with a low overall prevalence of diabetes, and no family or personal risk factors for diabetes. Women with gestational diabetes

should be screened for type 2 diabetes at the postpartum visit 6 to 12 weeks after delivery and then intermittently from that visit forward. The goal level for A1c remains less than 7% for adults. Patients recently diagnosed with diabetes and with a long life expectancy and no known cardiovascular disease may be considered for a lower target A1c level. All patients with diabetes should receive medical nutrition therapy, preferably from a registered dietician. Health plans should cover such a service as well as diabetes self-management education. Patients with diabetes should exercise at least 150 minutes per week at an intensity of 50% to 70% of the maximal heart rate. Exercise should include resistance training 3 times per week in the absence of contraindications to such exercise. The blood pressure goal of patients with diabetes remains less than 130 mm Hg for systolic blood pressure and less than 80 mm Hg for diastolic blood pressure. Antihypertensive regimens should include a renin-angiotensin system antagonist if possible. The fasting lipid profile should be measured at least annually, but if the goal lipid values are met (particularly low-density lipoprotein cholesterol level < 100 mg/dL), lipid assessments may be completed every 2 years. Aspirin therapy should be instituted for patients when the 10-year risk for a cardiovascular event exceeds 10%. This parameter should include men older than 50 years with diabetes and women older than 60 years with diabetes if they have 1 additional cardiovascular risk factor. Such risk factors include hypertension, dyslipidemia, smoking, albuminuria, or a family history of cardiovascular disease. Patients with type 2 diabetes should receive an initial dilated and comprehensive eye examination. Although high-quality fundus photographs detect most clinically significant diabetic retinopathy, they should not act as a substitute for a comprehensive eye examination. Retinal examinations should be carried out annually or at least every 2 to 3 years among low-risk patients with normal eye examination results in the past. Insulin therapy should be initiated in critically ill patients with diabetes when the plasma glucose level is 180 mg/dL or more. In general, the goal plasma glucose level for these patients is 140 to 180 mg/dL. The premeal glucose target for inpatients who are not critically ill is less than 140 mg/dL, and the random glucose target for these patients is less than 180 mg/dL.

Clinical Implications

Type 2 diabetes may be diagnosed if a patient is confirmed to have an A1c level of 6.5% or more, a fasting plasma glucose level of 126 mg/dL or more, a 2-hour plasma glucose level of 200 mg/dL or more after a 75-g oral glucose tolerance test, or a random plasma glucose level of 200 mg/dL or more in the presence of symptoms of hyperglycemia. Care for patients with diabetes should include treatment to decrease the A1c level to less than 7%, blood pressure values to less than 130/80 mg/dL, and low-density lipoprotein cholesterol concentrations to less than 100 mg/dL. Aspirin therapy is advised for men older than 50 years and women older than 60 years with diabetes plus 1 other cardiovascular risk factor.

BOUQUET

In Lighter Vein

One day an Irishman, who has been stranded on a desert island for over ten long years, sees an unusual speck on the horizon. "It's certainly not a ship," he thinks to himself.
As the speck gets closer and closer, he begins to rule out the possibilities of a small boat, then even a raft. Suddenly, emerging from the surf comes a drop dead gorgeous blonde woman wearing a wet suit and scuba gear.
She approaches the stunned man and says to him, "Tell me how long has it been since you've had a cigarette?"
"Ten years," replies the Irishman.
With that, she reaches over and unzips a waterproof pocket on her left sleeve and pulls out a pack of fresh cigarettes. He takes one, lights it, takes a long drag and says, "Faith and begorah! Is that good!"
"And how long has it been since you've had a sip of good Irish Whiskey?" she asks him.
Trembling, the castaway replies, "Ten years."
She reaches over, unzips her right sleeve, pulls out a flask and hands it to him. He opens the flask, takes a long swig and says, "Tis absolutely fantastic!"
At this point she starts slowly unzipping the long zipper that runs down the front of her wet suit, looks at the man and asks, "And how long has it been since you've played around?"
With tears in his eyes, the man falls to his knees and sobs, "Oh, Sweet Jesus! Don't tell me you've got golf clubs in there too."

--**A lonely frog** telephoned the Psychic Hotline and asked what his future holds.
His Personal Psychic Advisor tells him: "You are going to meet a beautiful young girl who will want to know everything about you."
The frog is thrilled, "This is great!
"Will I meet her at a party?" he croaks.
"No," says the psychic, "in biology class."

--**Two women** that are dog owners are arguing about which dog is smarter....
First Woman : "My dog is so smart, every morning he waits for the paper boy to come around and then he takes the newspaper and brings it to me.
Second Woman : "I know..."
First Woman : "How?"
Second Woman : "My dog told me."

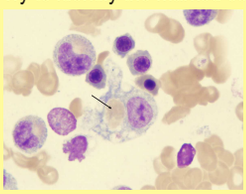
Wisdom Whispers

- "Time is - Too Slow for those who Wait; Too Swift for those who Fear; Too Long for those who Grieve; Too Short for those who Rejoice; But for those who Love; - Time is not."
- "Mediocrity can talk, but it is for genius to observe."
- "He who desires, but acts not, breeds pestilence."
- "A hungry man will listen to nothing."
- "Truth ill-timed is as bad as a lie."
- "The old ones sing, the young ones pipe."
- "Dishonest money dwindles away, but he who gathers money little by little makes it grow."
- "It's not shameful not to know, but it's shameful not to ask."
- "Misfortunes, when asleep, are not to be awakened."
- "When the will is prompt the legs are nimble."

Brain Teasers

Try to identify the marked cells in the pictures.

1.



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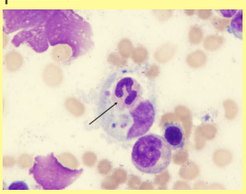

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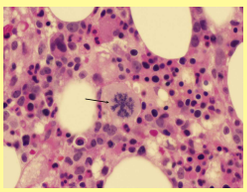
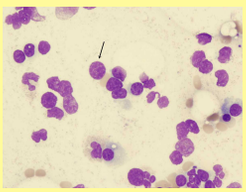


4.



Answers: Fig. 1. Histiocyte engulfing red cell, Fig. 2. Hemophagocytosis of neutrophil, Fig. 3. Mitotic division of megakaryocyte, Fig. 4. Smudge cell.

TROUBLESHOOTING

HbA1c Measurement

The measurement of HbA1c/GHb in blood has become the gold standard for the long term control of the glycemic state of the diabetic patients. The optimal therapy of diabetic patients requires carefully validated/ method independent therapeutic target values for the HbA1c levels of diabetic patients in order to reduce the long-term risk of late complications such as retinopathy, nephropathy and neuropathy as well as short term risk of life threatening hypoglycemia. The % HbA1c values can also be converted into Mean Plasma Glucose values as described in American Diabetes Association (ADA) recommendations for assays traceable to the DCCT reference method as described below:

% HbA1c	Approximate mean blood glucose		Interpretation
	Mg/dl	Mmol/L	
4	65	3.5	Non- diabetic range
5	100	5.5	
6	135	7.5	
7	170	9.5	ADA Target
8	205	11.5	Action Suggested
9	240	13.5	
10	275	15.5	
11	310	17.5	
12	345	19.5	

GHb comprises several different hemoglobin-glucose adducts including HbA1a, HbA1b and HbA1c. The different GHb assay methods available to the routine clinical laboratory can be divided into two major categories: those based on charge differences between GHb and non-GHb (these include cation-exchange chromatography, electrophoresis, and isoelectric focusing) and those based on structural characteristics of glycogroups on hemoglobin (these include affinity chromatography and immunoassay). Most methods quantify HbA1c, defined as HbA with glucose attached to the NH2-terminal valine of one or both β-chains. Other methods (boronate affinity) quantify "total glycated hemoglobin," which includes both HbA1c and other GHb adducts (e.g., glucose-lysine adducts and glucose α-chain NH2-terminal valine adducts). These factors have led to considerable variation in reference intervals and results reported by different laboratories. Proper interpretation of GHb test results is not easy and requires that health care providers understand the relationship between the test results and average blood glucose, kinetics of GHb, specific assay limitations, and patient factors (other than blood glucose levels) that can affect the results. To bring semblance in monitoring glycemic control by HbA1c it is important to understand the important aspects of different methods and interpret the results accordingly. A review of preanalytical and analytical variables affecting GHb assays has been reviewed and described below: In general, any situation that shortens erythrocyte survival or decreases mean erythrocyte age falsely lowers GHb test results regardless of the assay method. Vitamins C and E are reported to falsely lower test results, possibly by inhibiting glycation of hemoglobin. Iron-deficiency anemia is reported to increase test results. Hypertriglyceridemia, hyperbilirubinemia, uremia, chronic alcoholism, and chronic ingestion of salicylates, opiate addiction, hemoglobinopathies, and chemically modified derivatives of hemoglobin may interfere with some assay methods. Interferences from hemoglobin variants should be considered before interpreting the GHb test results. Interferences from hemoglobin variants and adducts are summarized on the National Glycohemoglobin Standardization Program (NGSP) Web site at www.ngsp.org. Laboratories should use GHb assay methods with an interassay coefficient of variation (CV) of <4% (ideally <3%). Each laboratory should also determine its own reference interval following NCCLS guidelines (Document C28A). In renal failure, carbamylated compounds abound, which also interfere with GHb estimation.

Common Analytical variables: Most common analytical variable in HbA1c/GHb measurement is the preparation of specimen Lysate for testing recommended by several methods. For preparing Lysate different methods recommend a small volume of whole blood (anywhere between 5-10µl whole blood) that has to be diluted in 500-1000 µl of hemolysing solution. A small variation in delivery of whole blood sample can significantly affect the test results. Loss in calibration of pipette can significantly affect the GHb/HbA1c test results. Homogenizing of whole blood is essential to evenly distribute the erythrocytes before aspirating specimen from the specimen tube. The time suggested for lysis of erythrocytes during lysate preparation is also very important and is the determinant in eliminating the interference of schiff's base in several assays. Most GHb methods need to measure both total hemoglobin and glycated hemoglobin. The percent HbA1c is calculated from the total and glycated hemoglobin. Error in calculation of total or glycated hemoglobin can contribute significantly in conversion to percent HbA1c.

Method specific variables: Cation exchange chromatography methods: Cation exchange chromatography can either be undertaken on mini columns or in a sophisticated, automated system. The pH and temperature conditions affect the results significantly; therefore require a sophisticated system where the conditions can be adequately controlled. In semi automated systems the room temperature varies at different times of the day and therefore can significantly influence the test results. HbA1c possesses less charge positivity and hence elutes faster from a cation exchange column. Pre-glycohemoglobin has similar mobility in this system and hence, needs to be removed before column chromatography. The kind of resin, lot to lot variation of resin , column size , buffer composition and elution time influences the test system. The presence of HbE affects the determination of GHb using cation-exchange based kits. Drugs that possess strong ionic charge like aspirin can alter GHb in ion-exchange chromatographic methods. Carbamyl-Hb has an isoelectric point similar to HbA1c and can thus interfere with charge-based methods of measuring gHb. In vitro carbamylation of Hb, to concentrations as high as 5.4% carbamyl-Hb, has been shown to produce significant spurious increases in HbA1c values in multiple cation-exchange methods. HbE1c frequently elutes as a shoulder to HbA1c in most HPLC or cation-exchange chromatographic methods. **Electrophoresis:** HbA1c can be separated from HbAo by any electrophoretic method. The most commonly used method is agargel electrophoresis where HbA1 migrates to cathodic side of HbAo. Pre-GHb migrates with GHb in this system as well and hence has to be separated in advance. Comigration of Hb variants or derivatives with either HbA or HbA1c interferes with HbA1c determinations. Comigration of HbF or carbamylated Hb with HbA1c produces spuriously increased HbA1c values. **Boron affinity methods:** Most semi-automated methods measure GHb and provide calculated HbA1c. This method is less influenced by pH, temperature and storage conditions. However, there is a batch-to-batch variation in gel characteristics, which makes application of this method difficult. A recent study demonstrated lot to lot variation in reagents of 6 of the eight HbA1c point of care instruments and proved that these do not meet the general accepted analytical performance criteria. Some of these points of care instruments used in the study were based on boron affinity method. **Immunological methods:** Immunoassays have been developed by using a HbA1c specific monoclonal antibody. These methods are more accurate and are suitable for both small and large laboratories. Most immunoassays are not affected by hemoglobin derivatives such as carbamylated or Acetylated Hb. They are also not affected by HbF, HbE, HbA2. These factors need to be considered when comparing results of immunoassays with other GHb methods. **Hb Variants:** A variety of patient-related factors and laboratory-related processes can lead to inaccurate determinations of gHb in the setting of variant Hbs. Samples should be evaluated for the presence of a Hb variant with any gHb reading >15%). In addition, any patient with a significant change in gHb coinciding with a change in laboratory gHb methods should be evaluated for the presence of variant or derivative Hb. Appropriate evaluation includes obtaining pertinent clinical history regarding hemoglobinopathies, alterations in red cell turnover, or conditions favoring the chemical modification of Hb. Manual review of cation-exchange chromatographs may identify the presence of aberrant peaks produced by variants. The gHb measurement should be repeated with a different assay method, and a Hb analysis by chromatography or electrophoresis should be performed to identify more common Hb variants. In some cases, variants may be identified only by ES-MS or by sequencing the expressed globin genes. Although most modern chromatographic and immunoassay methods are either unaffected by common heterozygous variants such as HbAS, HbAC, and HbAE or give warning flags concerning the likelihood of an underlying variant, less common variants may give no such warnings. Furthermore, all gHb methods are inadequate for the assessment of long-term glycemic control in patients homozygous for HbS, HbC, or with HbSC disease. Although technologies such as boronate affinity chromatography and ES-MS provide a means of accurately determining gHb in these individuals, results are unlikely to accurately reflect long-term glycemic control due to pathological conditions that affect the formation and turnover of gHb *in vivo*. In regions where populations have a high prevalence of variant Hbs, methods for the determination of gHb must be carefully selected to allow accurate determination of gHb in these individuals. When dealing with populations in which HbSS, HbCC, or HbSC disease are common and in which gHb determinations have limited utility, laboratories should offer alternative forms of testing, such as GSPs or GSA, to assist physicians with the determination of glycemic control in these individuals.

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Printed and published by D.G. Tripathi, Edited by Dr. R.J. Sood M.D. (path.) for and on behalf of Tulip Diagnostics Private Ltd, Gitanjali, Tulip Block, Dr. Antonio Do Rego Bagh Alto Santacruz, Bambolim Complex Post Office, Goa - 403202, INDIA.
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