Evaluation of diabetic marker Hba1c and anemia in the context of kidney disease

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Each year, more than 100,000 people in the United States are diagnosed with kidney failure, the final stage of kidney disease.[1] The most common cause is diabetes, accounting for nearly 44 percent of new cases.[1] Often, a consequence of kidney disease is anemia. This occurs when kidneys fail to generate enough erythropoietin hormone to trigger adequate red blood cell production. For decades, clinicians have successfully used the hemoglobin A1c (HbA1c or A1C) assay to monitor long-term blood glucose control for patients with chronic diabetes.[2,3] More recently, researchers have studied the HbA1c assay's use as a potential diagnostic marker for diabetes complications such as kidney disease.[4]

The HbA1c test measures average plasma glucose—hemoglobin in a red blood cell that was combined with glucose over the previous eight to 12 weeks.[5] The higher the HbA1c value, the greater the risk that the diabetes patient will develop kidney disease, and perhaps, anemia, a common consequence of renal disease. However, a chemically modified derivative of hemoglobin called carbamylated hemoglobin (CHb) can affect the accuracy of the HbA1c test results. Studies have shown that the formation of CHb due to abnormal urea concentration is linked to both the severity and the duration of renal failure.[6] Research findings have inspired conflicting viewpoints on the efficacy of HbA1c test results in the presence of CHb and on the level of CHb it takes to affect results. This article explores the links between diabetes and renal failure. It discusses what research has discovered about the effect of CHb on HbA1c testing. Finally, it shows how testing technology has improved to ensure HbA1c testing accuracy.

Kidney disease and diabetes

Nearly 24 million people in the U.S. have diabetes,[7] and nearly 180,000 people are living with kidney failure as a result of diabetes.[1] Poor blood glucose control is among the key factors that can contribute to the onset of diabetic kidney disease. Other factors include family history, race (African-Americans, Hispanics, and some Native American tribal members are more prone), obesity, and poor blood pressure control. The risk of developing chronic kidney disease (CKD) is similar in patients with type 1 or type 2 diabetes, with about 30 percent of either group developing some form of kidney disease.[7] In diabetic patients with end-stage renal disease, erythrocyte lifespan tends to be decreased. This may result in part from iron deficiency anemia, frequent transfusions, or other effects of kidney disease on erythrocyte survival.

Benefits of the HbA1c test

The A1C assay has been used for the management of chronic diabetes for decades and more recently has been recommended for its application as an aid to diagnosis and an aid to identify patients who may be a risk for developing diabetes.[8] The HbA1c test measures long-term blood glucose levels. Unlike the usually self-administered finger stick blood test used to monitor current blood sugar levels, the HbA1c test is a reflection of glycemic control over the previous two to three months. The HbA1c values provide clinicians with an overall picture of the average blood sugar levels over that period. When A1C levels are normal or near normal, the risk for complications from diabetes, including kidney and cardiovascular disease, ophthalmic problems, and nerve damage, decreases. When A1C levels rise, the risk of kidney disease increases. The test can be performed at any time of the day and does not require special preparation such as fasting.

In a recent seminar on A1C organized by the American Association for Clinical Chemistry in June 2016, it was reported that HbA1c is not a measure of renal function; however, persistent elevations in HbA1c do predict an increased risk for microvascular complications that cause diabetic nephropathy.[9]

Methods for measuring HbA1c

Several methods are available for measuring HbA1c:

- Boronate affinity chromatography: Glycohemoglobin binds affinity resin while non-glycated hemoglobins pass through the column. The glycated hemoglobin peak represents HbA1c, and the non-glycated peak represents all non-HbA1c hemoglobins.
- Ion-exchange high performance liquid chromatography (HPLC): Ion-exchange HPLC methods separate the hemoglobin species based on charge by applying a differential in ionic concentrations of buffers.
- Capillary electrophoresis (CE): CE methods separate the hemoglobin species based on charge by applying voltage.
- Immunoassays: Immunoassays employ an antibody targeted against the glycated N-terminus of the B-chain.
- Enzymatic methods: Measure A1c using enzymes that specifically cleave n-terminal valine.

CHb and HbA1c test results

Several factors can affect the accuracy of HbA1c measurement. One of them is CHb. CHb is found in high levels in patients with renal failure. It is formed by non-enzymatic condensation of cyanate with the N-terminal valine of hemoglobin, and is increased due to abnormal urea concentration, which is dissociated in vivo to yield cyanate ions.[5]

Studies have shown that formation of CHb depends upon both the severity and the duration of renal failure. CHb can play a role in the adequacy of hemodialysis treatment and may be useful in the clinical evaluation of renal failure patients.[10,11] There have been several studies to find out if CHb affects HbA1c values.[12,13]

Differentiating CHb from HbA1c

Some methods such as ion-exchange HPLC enable separation and detection of peaks other than HbA1c, including CHb. Methods such as immunoassays, boronate affinity chromatography, and enzymatic methodologies lack the ability to detect hemoglobins other than HbA1c.

Several methods have been improved to have better resolution between HbA1c and other hemoglobin adducts. Newer ion-exchange HPLC methods in particular have improved the resolution between HbA1c and other hemoglobin adducts so that peaks such as CHb are clearly demarcated from the HbA1c peak in the chromatogram.[14] Methods such as capillary electrophoresis also provide information on hemoglobins other than HbA1c.

Some studies on the effects of CHb from varying degrees of renal conditions have indicated that the CHb in the methods used showed no apparent effect on A1c values at high thresholds of CHb levels. That said, there is an ongoing debate about the most useful way to monitor glycemic control in chronic renal failure (CRF) patients.[15,16]

CKD and anemia

In patients diagnosed with both diabetes and CKD, there is a high risk for anemia, especially among those who require dialysis. Anemia is almost universal in end-stage renal disease (ESRD), because of insufficient production of erythropoietin by non-functioning

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kidneys. To screen and manage patients at risk for anemia, practitioners can use the comprehensive reticulocyte panel as part of expanded CBC results. The additional data includes absolute reticulocyte counts, reticulocyte percentage, immature reticulocyte fraction (IRF), and reticulocyte hemoglobin equivalent (RET-He). RET-He is a measurement of the hemoglobin content of the developing reticulocyte population. Unlike traditional iron tests such as serum iron, ferritin, and TSAT, which are affected by inflammation, RET-He is not sensitive to interference from underlying inflammatory conditions. Pathophysiological mechanisms of anemia can be elucidated and used to justify specific therapy choices. Reticulocyte counts represent the erythropoietic contributions, while reticulocyte hemoglobin content represents iron-dependent hemoglobinization.

The RET-He is included in key guidelines that drive the national and global anemia management of kidney disease. These guiding principles include Kidney Disease Outcome Quality Initiative Guidelines (KDOQI) published in 2006; Kidney Disease: Improving Global Outcomes Guidelines (KDIGO) published in 2012; and most recently NICE (National Institute for Health and Care Excellence) 2015 recommendations. The use of RET-He in challenging patients is extremely helpful because it can alert practitioners to the presence of iron deficiency anemia when traditional tests are inconclusive. It can also help assess the effectiveness of treatment earlier than traditional parameters.[17]

Lab testing and automation

Automation is a key focus by laboratories today. Laboratories implement or explore automation to reduce errors, improve clinical data turnaround time, and reduce overall operational costs. Testing solutions that provide clinical data to assess and manage this patient population are available as stand-alone work stations or as a single workstation.

Most A1C methods provide accurate results in CRF patients; methods such as ion-exchange HPLC can provide the ability to detect the presence of CHb that is elevated in CRF patients. Automation can provide both HbA1c (using ion-exchange HPLC) and RET-He through the use of a single sample from the patient. In addition, the single workstation automation system includes decision logic software to maximize clinical data turnaround time. Decision logic software allows filtering abnormal results (those that need further attention) from normal/auto-verified (those that will be automatically released to LIS and directly to patient chart) using instrument-specific rule set.

Thus, patients who exhibit peak abnormalities are quickly and automatically filtered (through the activation of rules) and identified, so that the laboratorian can focus and follow up promptly. Laboratories that have applied this type of automation can realize benefits with workstation elimination, reduction in hands-on time with testing, and improvement in overall turnaround time.

Clinicians have options that can help to diagnose and manage disease states. The HbA1c assay, used successfully by clinicians for diabetes control, is a valuable tool in diagnosis of diabetes. The higher a diabetes patient's measured HbA1c value, the greater the risk of developing diabetes-related kidney disease. An accurate A1C result is important, especially in the presence of CHb that is elevated in kidney disease patients. It is important that clinician and laboratory be aware of methods that provide an accurate A1C in the presence of CHb for proper patient care.[18] The risk of developing anemia in kidney disease patients is great, and proper assessment and management of this condition is also important. A comprehensive retic panel with RET-He provides better reliability versus traditional anemia testing panels, enabling clinicians to determine the best course in overall care for these patients.[19] RET-He, measured at the cellular level, reflects change within three to five days of therapy, and is not impacted by inflammatory processes. National and global kidney organizations have recognized the value of RET-He in anemia management and have included the parameter in key guidelines.[19-21] Early screening for iron deficiency anemia and efficient disease management can promote patient outcomes and reduce healthcare costs.

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