CARDIOMETABOLIC PROFILE IN DRIED BLOOD SPOT

The Problem

The incidence of cardiovascular disease (CVD), obesity and type 2 diabetes mellitus (DM2) is rising at an alarming rate. CVD is the leading cause of mortality for both men and women in the United States; obesity, insulin resistance and DM2 significantly predispose individuals to developing CVD, yet these conditions are potentially avoidable. If we are to make an impact on the serious health and economic consequences of these diseases, we need to identify risk early enough for people to make lifestyle modifications or seek medical help, and avoid becoming a part of the rising statistics.

What is CardioMetabolic Risk?

Cardiometabolic risk has been defined as "the cluster of modifiable risk factors and markers that identify individuals at increased risk for cardiovascular disease (myocardial infarction, stroke, peripheral arterial disease) and type 2 diabetes[1]." The National Cholesterol Education Program (NCEP)'s Adult Treatment Panel III (ATP III) has identified the metabolic syndrome/insulin resistance syndrome as a major risk factor for DM2 and CVD[2,3]. NCEP-ATP III criteria for identifying metabolic syndrome include:

- hypertension/elevated blood pressure
- abdominal obesity
- atherogenic dyslipidemia (low HDL cholesterol, elevated triglycerides, elevated LDL cholesterol)
- prothrombotic/pro-inflammatory state
- insulin resistance/glucose intolerance

Advantages of a Simple Blood Spot Test to Assess CardioMetabolic Risk

- A simple, almost painless finger stick provides the few drops of blood required, which are collected on the filter paper provided
- Convenient sample collection at home - no phlebotomist required
- Easy shipment of samples by regular mail for analysis - samples are stable for several weeks at room temperature
- Dried blood spots carry little infection risk - infectious agents, such as HIV, are inactivated when dry
- Excellent correlation with conventional venipuncture serum/plasma assays[4]

Which Biomarkers are Included in the CardioMetabolic Profile?

High Sensitivity C-Reactive Protein (hs-CRP)

C-reactive protein (CRP) is an established marker of inflammation and has recently been suggested to be an important contributor to pro-inflammatory and pro-thrombotic elements of CVD risk. Extremely high CRP levels are seen in acute inflammatory states, but the small elevations that are indicative of the pro-inflammatory and pro-thrombotic states implicated in the metabolic syndrome require high sensitivity assays, and are thus referred to as hs-CRP levels. These high sensitivity assays have recently been developed for use with blood spots[5,6,7].

- Overweight, obese, insulin resistant and diabetic individuals typically have elevated CRP levels[8]

866.600.1636 | info@zrtlab.com | zrtlab.com

ZRT LABORATORY

Dried Blood Spot Testing.
Minimally-invasive home test kit.

- Studies have shown correlations between elevated CRP and increased risk of future heart attacks, ischemic stroke, and peripheral arterial disease[9-12]

- Elevated CRP levels have been found to predict the development of DM2[13]

- Increased CRP levels, which correlate inversely with insulin sensitivity, have been found in individuals with polycystic ovarian syndrome and may be a marker of early cardiovascular risk in these patients[14,15]

- Lifestyle changes such as aerobic exercise, weight loss and smoking cessation lower CRP[10,16]

- Medications like aspirin and statins can lower CRP levels[12,17]

- Levels below 3.0 mg/L are considered to be normal; 3.1 to 10 mg/L is elevated, in the context of CVD risk, and above 10 mg/L is very high, more likely indicating an acute inflammatory event due to infection or trauma

Fasting Insulin

Dried blood spot technology has effectively been used for measurement of insulin levels[18-20]. The requirement to measure fasting insulin makes convenient blood spot collection at home especially advantageous.

- High fasting insulin levels are a good indicator of insulin resistance, which occurs when the cellular response to the presence of insulin is impaired, resulting in a reduced ability of tissues to take up glucose for energy production. Chronically high insulin levels are seen as the body attempts to normalize blood sugar levels

- High fasting insulin indicates the presence of insulin resistance, whether or not the patient shows glucose intolerance

- The normal range for fasting insulin is 1 – 15 μIU/mL, but levels between 2 and 6 μIU/mL are optimal

Hemoglobin A1c (HbA1c)

HbA1c is a measure of red blood cell hemoglobin glycation, indicating mean glycemia over the previous three months, which is the lifespan of circulating red blood cells. It can therefore indicate impaired glucose tolerance even when occasional fasting plasma glucose measurements are normal[21]. Recent research has confirmed the stability of HbA1c in dried blood spot samples stored at room temperature for up to a month[22].

- The American Diabetes Association's recommendation is to measure HbA1c every 3-6 months; normal levels are 4 - 6%

- Levels of HbA1c above 6% in diabetics are associated with an increased risk of developing complications such as eye, kidney, and heart disease, nerve damage, and stroke, therefore treatment should aim to keep levels below 7%[23]

- HbA1c levels above 6% can predict CVD and DM2 in high risk individuals[24-26]

Fasting Triglycerides

Hypertriglyeridemia, a triglyceride level >150 mg/dL, is an established indicator of atherogenic dyslipidemia and is often found in untreated DM2 and obesity.

- Studies have shown that levels above 200 mg/dL indicate an increased risk of heart disease and stroke[27]

- Some studies have shown that fasting triglyceride levels lower than 100 mg/dL should be considered as a more optimal cutoff in coronary heart disease risk assessment[28]

- The NCEP-ATP III defines levels of 150 mg/dL or above as one of the diagnostic criteria for metabolic syndrome[2]

Total Cholesterol, LDL Cholesterol, VLDL Cholesterol, and HDL Cholesterol

Abnormalities in the lipid profile, including high total cholesterol, high LDL cholesterol, high VLDL cholesterol, and low HDL cholesterol, are a significant component of coronary heart disease risk because of their contribution to the development of atherosclerosis. As with other cardiometabolic risk factors, they are more significant when other cardiometabolic parameters are already abnormal, or in patients who already have diabetes or CVD.

Reduced HDL cholesterol constitutes one of the established criteria for the diagnosis of metabolic syndrome[3], and has long been regarded as a powerful

predictor of CVD in both diabetics and non-diabetics[29]. Currently, the LDL cholesterol/HDL cholesterol ratio is regarded as a reliable tool for the evaluation of CVD risk: the higher the ratio, the greater the risk of CVD[30]. In a large cohort from the Framingham Study, the total cholesterol/HDL cholesterol ratio and the LDL cholesterol/HDL cholesterol ratio were associated with increased coronary heart disease risk, and the HDL cholesterol level was associated with reduced risk, in both men and women[31].

While absolute values of each are still considered by the NCEP and the American Heart Association as the optimal diagnostic indicators, an LDL/HDL ratio below 3 and a total cholesterol/HDL ratio below 4 are currently accepted by doctors and researchers as optimal for health.

A recent analysis of clinical trials using lipid modifying drugs in people already at risk showed that artificially increasing HDL cholesterol levels with drug therapy did not translate to a reduced risk of coronary heart disease; however, for every 10% reduction in LDL cholesterol with drug therapy, there was a 10% relative reduction in coronary heart disease events[32].

Very low density lipoprotein (VLDL) cholesterol is a reliable marker of remnant lipoproteins, which play a significant role in atherogenesis. VLDL plus LDL cholesterol is referred to as "non-HDL cholesterol" or "atherogenic cholesterol" and gives a more complete picture of total risk than LDL cholesterol alone, especially in patients with a triglyceride level >200 mg/dL[2].

The current NCEP-ATP III recommendations[2] for cholesterol levels (in mg/dL) are:

Total cholesterol:

    <200 desirable   200 - 239 borderline high

    >240 high

HDL cholesterol:

    >40 optimal

LDL cholesterol:

    <100 optimal   100 - 129 near optimal

    130 -159 borderline high

    160 - 189 high   >190 very high

VLDL cholesterol:

    <30 optimal

The American Diabetes Association and American College of Cardiology Foundation, in a recent consensus statement on lipoprotein management, recommended the following cutoffs for LDL cholesterol in patients at high risk[33]:

Highest risk patients, including those with known CVD or diabetes plus one or more additional major CVD risk factors:

LDL Cholesterol:

    <70 mg/dL

High-risk patients, including those without diabetes or CVD but having 2 or more additional major CVD risk factors:

LDL Cholesterol:

    <100 mg/dL

Clinical Utility

The blood spot CardioMetabolic Profile allows early detection of major indicators associated with metabolic/insulin resistance syndrome. Used as a screening profile this can help clinicians make the most appropriate treatment recommendations to reduce the overall risk of DM2 and CVD. Regular testing can also be used for risk assessment and monitoring patients with DM2. Screening, along with clinical assessment, can be of reliable predictive value for determining overall cardiometabolic risk.

References

1. Watson K. Managing cardiometabolic risk: an evolving approach to patient care. Crit Pathw Cardiol. 2007; 6:5-14.

2. National Cholesterol Education Program (NCEP) Expert Panel on Detection, Evaluation, and Treatment of High Blood Cholesterol in Adults (Adult Treatment Panel III). Third Report of the National Cholesterol Education Program (NCEP) Expert Panel on Detection, Evaluation, and Treatment of High Blood Cholesterol in Adults (Adult Treatment Panel III) final report. Circulation 2002;106:3143-421.

3. Grundy SM, Brewer HB Jr, Cleeman JI, Smith SC Jr, Lenfant C; American Heart Association; National Heart, Lung, and Blood Institute. Definition of metabolic syndrome: Report of the National Heart, Lung, and Blood Institute/ American Heart Association conference on scientific issues related to definition. Circulation 2004;109:433-8.

4. Kapur S, Kapur S, Zava D. Cardiometabolic risk factors assessed by a finger stick dried blood spot method. J Diabetes Sci Technol 2008; 2:236-241.

5. McDade TW, Burhop J, Dohnal J. High sensitivity enzyme immunoassay for C-reactive protein in dried blood spots. Clin Chem 2004; 50:652-4.

6. Cordon SM, Elborn JS, Hiller EJ, Shale DJ. C-reactive protein measured in dried blood spots from patients with cystic fibrosis. J Immunol Methods 1991;143:69-72.

7. Beesley R, Al Serouri A, Filteau SM. Measurement of C-reactive protein in dried blood spots on filter paper. Trans R Soc Trop Med Hyg. 2000;94:348-349.

8. Marques-Vidal P, Mazoyer E, Bongard V, Gourdy P, Ruidavets JB, Drouet L, Ferrieres J. Prevalence of insulin resistance syndrome in southwestern France and its relationship with inflammatory and hemostatic markers. Diabetes Care 2002;25:1371-7.

9. Shankar A, Li J, Nieto FJ, Klein BE, Klein R. Association between C-reactive protein level and peripheral arterial disease among US adults without cardiovascular disease, diabetes, or hypertension. Am Heart J 2007;154:495-501.

10. Church TS, Barlow CE, Earnest CP, Kampert JB, Priest EL, Blair SN. Associations between cardiorespiratory fi tness and C-reactive protein in men. Arterioscler Thromb Vasc Biol 2002;22:1869-76.

11. Madsen T, Skou HA, Hansen VE, Fog L, Christensen JH, Toft E, Schmidt EB. C-reactive protein, dietary n-3 fatty acids, and the extent of coronary artery disease. Am.J Cardiol 2001;88:1139-42.

12. Ridker PM, Cushman M, Stampfer MJ, Tracy RP, Hennekens CH. Infl ammation, aspirin, and the risk of cardiovascular disease in apparently healthy men. New Engl J Med 1997;336:973-9.

13. Pradhan AD, Manson JE, Rifai N, Buring JE, Ridker PM. C-reactive protein, interleukin 6, and risk of developing type 2 diabetes mellitus. JAMA 2001;286:327-34.

14. Boulman N, Levy Y, Leiba R, Shachar S, Linn R, Zinder O, Blumenfeld Z. Increased C-reactive protein levels in the polycystic ovary syndrome: a marker of cardiovascular disease. J Clin Endocrinol Metab 2004;89:2160-65.

15. Tarkun I, Arslan BC, Canturk Z, Turemen E, Sahin T, Duman C. Endothelial dysfunction in young women with polycystic ovary syndrome: relationship with insulin resistance and low-grade chronic inflammation. J Clin Endocrinol Metab 2004;89:5592-6.

16. Hastie CE, Haw S, Pell JP. Impact of smoking cessation and lifetime exposure on C-reactive protein. Nicotine Tob Res 2008;10:637-42.

17. Ridker PM, Rifai N, Clearfi eld M, Downs JR, Weis SE, Miles JS, Gotto AM Jr. Measurement of C-reactive protein for the targeting of statin therapy in the primary prevention of acute coronary events. New Engl J Med 2001;344:1959-65.

18. Shields BM, Knight B, Shakespeare L, Babrah J, Powell RJ, Clark PM, Hattersley AT. Determinants of insulin concentrations in healthy 1-week-old babies in the community: applications of a blood spot assay. Early Hum Dev 2006;82:143-8.

19. Butter NL, Hattersley AT, Clark PM. Development of a blood spot assay for insulin. Clin Chim Acta. 2001;310:141-150.

20. Dowlati B, Dunhardt PA, Smith MM, Shaheb S, Stuart CA. Quantification of insulin in dried blood spots. J Lab Clin Med 1998;131:370-4.

21. Geberhiwot T, Haddon A, Labib M. HbA1c predicts the likelihood of having impaired glucose tolerance in high-risk patients with normal fasting plasma glucose. Ann Clin Biochem. 2005;42:193-5.

22. Buxton OM, Malarick K, Wang W, Seeman T. Changes in dried blood spot Hb A1c with varied postcollection conditions. Clin Chem. 2009;55:1034-6.

23. Saudek CD, Kalyani RR, Derr RL. Assessment of glycemia in diabetes mellitus: hemoglobin A1c. J Assoc Physicians India 2005;53:299-305.

24. Grant T, Soriano Y, Marantz PR, Nelson I, Williams E, Ramirez D, Burg J, Nordin C. Community-based screening for cardiovascular disease and diabetes using HbA1c. Am J Prev Med 2004;26:271-5.

25. Perry RC, Shankar RR, Fineberg N, McGill J, Baron AD; Early Diabetes Intervention Program (EDIP). HbA1c measurement improves the detection of type 2 diabetes in high-risk individuals with nondiagnostic levels of fasting plasma glucose: the Early Diabetes Intervention Program (EDIP). Diabetes Care 2001;24:465-71.

26. Singer DE, Nathan DM, Anderson KM, Wilson PW, Evans JC. Association of HbA1c with prevalent cardiovascular disease in the original cohort of the Framingham Heart Study. Diabetes 1992;41:202-8.

27. Tirosh A, Rudich A, Shochat T et al. Changes in triglyceride levels and risk for coronary heart disease in young men. Ann Intern Med 2007;147:377-85.

28. Ahmad I, Zhan M, Miller M. High prevalence of C-reactive protein elevation with normal triglycerides (100-149 mg/dL): are triglyceride levels below 100 mg/dL more optimal in coronary heart disease risk assessment? Am J Med Sci 2005;329:173-7.

29. Boden WE. High-density lipoprotein cholesterol as an independent risk factor in cardiovascular disease: assessing the data from Framingham to the Veterans Affairs High--Density Lipoprotein Intervention Trial. Am J Cardiol. 2000;86:19L-22L.

30. Fernandez ML, Webb D. The LDL to HDL cholesterol ratio as a valuable tool to evaluate coronary heart disease risk. J Am Coll Nutr. 2008;27:1-5.

31. Ingelsson E, Schaefer EJ, Contois JH, McNamara JR, Sullivan L, Keyes MJ, Pencina MJ, Schoonmaker C, Wilson PW, D'Agostino RB, Vasan RS. Clinical utility of different lipid measures for prediction of coronary heart disease in men and women. JAMA. 2007;298(7):776-85.

32. Briel M, Ferreira-Gonzalez I, You JJ, Karanicolas PJ, Akl EA, Wu P, et al. Association between change in high density lipoprotein cholesterol and cardiovascular disease morbidity and mortality: systematic review and meta-regression analysis. BMJ. 2009;338:b92.

33. Brunzell JD, Davidson M, Furberg CD, Goldberg RB, Howard BV, Stein JH, Witztum JL; American Diabetes Association; American College of Cardiology Foundation. Lipoprotein management in patients with cardiometabolic risk: consensus statement from the American Diabetes Association and the American College of Cardiology Foundation. Diabetes Care. 2008 ;31:811-22.