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:

- Identify hormonal imbalances associated with weight gain and obesity
- hypertension/elevated blood pressure
- abdominal obesity
- atherogenic dyslipidemia (low HDL cholesterol, elevated triglycerides, elevated LDL cholesterol)
- prothrombotic/pro-inflammatory state
- insulin resistance/glucose intolerance

Available Tests

CardioMetabolic Profile

Tests: Insulin, hsCRP, HbA1c, TG, CH, HDL, LDL, VLDL (blood spot)

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 type 2 diabetes and CVD. Regular testing can also be used for risk assessment and monitoring. Screening, along with clinical assessment, can be of reliable predictive value for determining overall cardiometabolic risk.

1998-2018 20 years

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]
- 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].

- Normal levels of HbA1c are <5.7%; levels of 5.7%-6.4% are indicative of prediabetes; levels of 6.5% and higher are diagnostic of diabetes (www.diabetes.org/diabetes-basics/diagnosis/)
- The American Diabetes Association's recommendation is to measure HbA1c every 3-6 months in diabetics
- Levels of HbA1c above 7% 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]

Blood Lipid Testing

Minimally-invasive home test kit

- 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

References

1. Canaris GJ, Manowitz NR, Mayer G, Ridgway EC. The Colorado thyroid disease prevalence study. Arch Intern Med 2000;160:526-34.
2. American Thyroid Association – www.thyroid.org
3. Gharib H, Tuttle RM, Baskin HJ, et al. Subclinical thyroid dysfunction: a joint statement on management from the American Association of Clinical Endocrinologists, the American Thyroid Association, and the Endocrine Society. J Clin Endocrinol Metab. 2005;90:581-5; discussion 586-7.
4. Vejbjerg P, Knudsen N, Perrild H, et al. Thyroglobulin as a marker of iodine nutrition status in the general population. Eur J Endocrinol. 2009;161:475-81.5.
5. Zava TT, Kapur S, Zava DT. Iodine and creatinine testing in urine dried on filter paper. Anal Chim Acta 2013;764:64-9.
6. Zava TT, Zava DT. Determination of iodine, bromine, selenium and arsenic by ICP-DRC-MS using urine dried on filter paper. Poster presented at the 83rd Annual Meeting of the American Thyroid Association, October 16-20, 2013, San Juan, Puerto Rico.
7. Zimmermann MB. Iodine deficiency. Endocr Rev. 2009;30:376-408.
8. WHO, UNICEF, ICCIDD, Assessment of iodine deficiency disorders and monitoring their elimination; a guide for programme managers, third ed., WHO publications, Geneva, 2007.
9. Bromism. In: Parfitt K, ed. Martindale 32nd ed. Pharmaceutical Press, 1999:1620-3.
10. Brown KM, Arthur JR. Selenium, selenoproteins and human health: a review. Public Health Nutr. 2001;4:593-9.
11. Mehdi Y, Hornick JL, Istasse L, Dufrasne I. Selenium in the environment, metabolism and involvement in body functions. Molecules. 2013;18:3292-311.
12. Bianco AC, Salvatore D, Gereben B, et al. Biochemistry, cellular and molecular biology, and physiological roles of the iodothyronine selenodeiodinases. Endocr Rev. 2002;23:38-89.
13. Kapaj S, Peterson H, Liber K, Bhattacharya P. Human health effects from chronic arsenic poisoning--a review. J Environ Sci Health A Tox Hazard Subst Environ Eng. 2006;41:2399-428.
14. Ciarrocca M, Tomei F, Caciari T, et al. Exposure to arsenic in urban and rural areas and effects on thyroid hormones. Inhal Toxicol. 2012;24:589-98.
15. Van Hulle M, Zhang C, Schotte B, et al. Identification of some arsenic species in human urine and blood after ingestion of Chinese seaweed Laminaria. J Anal At Spectrom. 2004;19:58-64.
16. Clifton JC 2nd. Mercury exposure and public health. Pediatr Clin North Am. 2007;54:237-69, viii.
17. Environmental Protection Agency. Health effects of mercury. Available at: http://www.epa.gov/hg/effects.htm
18. Khan MA, Wang F. Mercury-selenium compounds and their toxicological significance: toward a molecular understanding of the mercury-selenium antagonism. Environ Toxicol Chem. 2009;28:1567-77.
19. Branco V, Canário J, Lu J, Holmgren A, Carvalho C. Mercury and selenium interaction in vivo: effects on thioredoxin reductase and glutathione peroxidase. Free Radic Biol Med. 2012;52:781-93.
20. Park JD, Zheng W. Human exposure and health effects of inorganic and elemental mercury. J Prev Med Public Health. 2012;45:344-52.
21. ATSDR Priority List of Hazardous Substances, 2013. Available at: http://www.atsdr.cdc.gov/SPL/.
22. ATSDR Public Health Statement for Cadmium; September 2012. Available at: http://www.atsdr.cdc.gov/PHS/PHS.asp?id=46&tid=15.
23. Jancic SA, Stosic BZ. Cadmium effects on the thyroid gland. Vitam Horm. 2014;94:391-425.
24. Chen A, Kim SS, Chung E, Dietrich KN. Thyroid hormones in relation to lead, mercury, and cadmium exposure in the National Health and Nutrition Examination Survey, 2007-2008. Environ Health Perspect. 2013;121(2):181-6
25. McDermott MT, Ridgway EC. Subclinical hypothyroidism is mild thyroid failure and should be treated. J Clin Endocrinol Metab 2001;86:4585-90.
26. American Association of Clinical Endocrinologists. Medical guidelines for clinical practice for the evaluation and treatment of hyperthyroidism and hypothyroidism. AACE Thyroid Task Force. https://www.aace.com/files/hypo_hyper.pdf.
27. Banovac K, Zakarija M, McKenzie JM. Experience with routine thyroid function testing: abnormal results in "normal" populations. J Fla Med Assoc 1985;72:835-9.
28. Bjøro T, Holmen J, Krüger O, et al. Prevalence of thyroid disease, thyroid dysfunction and thyroid peroxidase antibodies in a large, unselected population. The Health Study of Nord-Trøndelag (HUNT). Eur J Endocrinol 2000;143:639-47.
29. Sakaihara M, Yamada H, Kato EH, et al. Postpartum thyroid dysfunction in women with normal thyroid function during pregnancy. Clin Endocrinol (Oxf) 2000;53:487-92.
30. Janssen OE, Mehlmauer N, Hahn S, et al. High prevalence of autoimmune thyroiditis in patients with polycystic ovary syndrome. Eur J Endocrinol. 2004;150:363-9.
31. Trokoudes KM, Skordis N, Picolos MK. Infertility and thyroid disorders. Curr Opin Obstet Gynecol. 2006;18:446-51.
32. Abalovich M, Mitelberg L, Allami C, et al. Subclinical hypothyroidism and thyroid autoimmunity in women with infertility. Gynecol Endocrinol. 2007;23:279-83.
33. Poppe K, Glinoer D, Tournaye H, Devroey P, et al. Thyroid autoimmunity and female infertility. Verh K Acad Geneeskd Belg. 2006;68:357-77.