Top Ten Things to Know
Potential Impact and Study Considerations of Metabolomics in Cardiovascular Health and Disease

1. Metabolomics, among the most recently emerged “omics” fields, involves the systematic measure and study of small molecule metabolites across biological systems using biosamples that include plasma, urine, saliva, and tissues.

2. Metabolomics serves as a reflection of gene and protein functional activity and, in doing so, captures information that is proximal to a given disease phenotype while spanning the spectrum from genetic sequence to cellular physiology.

3. Where the genetic sequence is largely static from birth, metabolomic measures are relatively dynamic, representing cellular activity as well as the effects of extrinsic exposures including dietary intake, physical fitness, microbiota variation, and toxicant exposures. The ability to integrate measures of both intrinsic and extrinsic origin, as well as their interplay, may effectively bridge investigations of both gene and environment and will be critical for understanding complex multi-faceted human diseases such as cardiovascular disease (CVD).

4. Several methods are available for detecting molecular markers in biological samples including nuclear magnetic resonance (NMR), mass spectrometry (MS), liquid chromatography (LC), and gas chromatography (GC). These methods can be coupled together or with other measurement methods to detect metabolomics markers such as circulating free amino acids or fatty acids, branched chain amino acids (BCAA), and trimethylamine-N-oxide (TMAO) that may be associated with cardiovascular risk factors.

5. Although broad scale metabolomics assays have not been approved by the FDA for clinical purposes, mass spectrometry is routinely used under FDA approval to measure a variety of select compounds or metabolites in clinical laboratories. Metabolomics data are often expressed as a percent increase/decrease relative to a reference population, so translating metabolomics measures from the research laboratory to clinical practice will require quality control systems for relative quantitation.

6. Metabolomics studies often require maintenance, processing, analysis, and reporting of large data sets.

7. Accounting for diet, comorbidities, and other confounders in metabolomics studies of CVD will require a variety of approaches:
   - **Data sets** – metabolomics data acquired from large, well-phenotyped cohorts are required to establish the matrix of correlations between metabolite levels and clinical variables known to be associated with cardiometabolic disease (e.g. age, race, sex, body size, insulin resistance, blood pressure, and renal function).
   - **Associated factors** – more in-depth studies of individuals over time are required to clarify how fasting versus fed status, dietary patterns, circadian cycles, other time-dependent factors, and random variation can impact circulating metabolites.
• **Clinical assessments** – although less important from a strict biomarker sense, studies that utilize invasive catheterization of select vascular beds are important to establish the potential organ-specificity of select peripheral venous metabolite signatures.

• **Genetic associations** – understanding the genetic determinants of plasma metabolite levels and cross-referencing with prior genetics studies of CVD offer the potential to implicate select metabolite biomarkers in causal pathways.

8. An integrated approach to understanding and targeting the links between genetic predisposition and risk exposures is especially needed for complex and multifactorial disease entities such as CVD. The ability to efficiently and effectively use metabolomics tools to conduct molecular phenotyping could serve to substantially advance the goals of precision medicine.

9. The success of the field of metabolomics to yield a next generation of important discoveries in cardiovascular science will depend on key challenges including:
   - Bioinformatics approaches for handling high-throughput untargeted metabolomics data.
   - Strategies for identifying the biochemical structure and functional role of novel metabolites.
   - Methods for determining the true relevance of metabolites observed in association with clinical outcomes.

10. Using metabolomics to integrate genomic and metabolomic data could further our understanding of the pathophysiology of diseases and allow clinicians and researchers to better explain why an individual develops CVD in response to a given exposure history such as tobacco or chronic stress.