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# A Role for Branched-Chain Amino Acids in the Pathophysiology of Diabetes: Using Data to Guide Discovery

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**Featured Article:** Wang TJ, Larson MG, Vasan RS, Cheng S, Rhee EP, McCabe E, et al. Metabolite profiles and the risk of developing diabetes. Nature Med 2011;17:448–53.[2]

Over the past 10 years, the ability to quantify human biologic activity has grown exponentially. We now have the technology for high-throughput genomics, transcriptomics, proteomics, and metabolomics and the ability to interrogate the human biochemical system at every level *(1)*. Indeed, hundreds of chemicals can now be measured in hundreds of individuals at reasonable cost. These advances have fueled analyses that drive medicine forward in a new way: instead of a hypothesis motivating the generation of new data, more and more discoveries are made when the data generate a new hypothesis. The article featured here exemplifies the power of this technique.

The recent advent of metabolomic profiling was seen as a great opportunity to better understand type 2 diabetes mellitus (DM). Because DM is a disease of pathologic energy management, metabolite profiling is a natural tool for understanding the underlying pathophysiology and identifying individuals at risk before the development of clinical disease. Improved quantification of this risk would allow focused prevention efforts to slow a growing national epidemic. It was with these goals in mind that we undertook a metabolomics screen in a nested case–control study of individuals from the Framingham Offspring Study. Metabolite profiles were generated using liquid chromatography–tandem mass spectrometry in 189 nondiabetics who went on to develop DM over a 12-year period, as well as in 189 propensity-matched controls. This analysis identified increased concentrations of 5 branched-chain and aromatic amino acids (BCAAs) that presaged the future onset of diabetes: isoleucine, leucine, valine, tyrosine, and phenylalanine. The concentrations of these amino acids increased predictive power beyond basic clinical features, such as body mass index, as well as established biochemical measures including blood glucose or insulin concentrations. This finding not only fit with existing data showing that BCAAs were linked to insulin sensitivity but the finding was also validated in cases and controls from the Malmö Diet and Cancer study.

Validation is of critical importance to all studies that are hypothesis-generating, and, since publication, the results of this study have been validated in various ways. The association between BCAAs and incident diabetes was also found in data from the Diabetes Prevention Program in a similar analysis *(2)*. These analyses effectively generated the hypothesis of a role for BCAAs in incident DM, but whether this relationship was causal or simply a result of reverse causality remained unclear. Recently, 2 studies have looked to strengthen the evidence for a causal relationship. Both used Mendelian randomization. In Mendelian randomization, if a biomarker has a truly causal relationship with disease, then genetic variants associated with that biomarker (and not other disease risk factors) should also predict disease. In the first study, genome-wide association for BCAAs was assessed in 16 596 individuals. The top single-nucleotide polymorphisms from this analysis were then assessed for their ability to differentiate 47 877 cases of DM from 267 694 controls. Indeed, several single-nucleotide polymorphisms were identified *(3)*. The second study generated evidence for a causal relationship between insulin resistance and concentration of BCAAs *(4)*. Finally, in a very recent small study of 25 participants, 1 month of treatment with empagliflozin, an SGTL2 inhibitor for treatment of diabetes, resulted in an increase in short-chain acylcarnitines that are derived from catabolism of BCAAs *(5)*. Taken together, these various data support a potential causal role for BCAAs mediating the progression from insulin resistance to clinical DM and identify them as a useful biomarker of insulin resistance, disease risk, and treatment effect *(6)*.

The study presented in our featured article set an example for the use of bioinformatics and systems biology to gain a deeper understanding of disease states and demonstrated an end-to-end pipeline for identification of novel biochemical pathways of disease. With high-

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[2] This article has been cited >950 times since publication.

throughput "-omic" characterization of large cohorts, hypothesis-generating associations between metabolites and disease phenotypes can be identified and validated. These associations can then be screened for probable causality using genome-wide association and Mendelian randomization studies, in parallel with mechanistic studies manipulating pathway-specific enzymes or metabolites in model systems. The pairwise linking of metabolite, genetics, and disease holds promise to rapidly identify numerous candidates for targeted experiments with a high likelihood of success and can set us on the path to the next generation of treatments.

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