Grand Challenge concept - "Enabling precision metabolomics in medicine and society"

Personalized medicine has been currently focused on the analysis of the genome and expressed genes (the transcriptome), made possible by massive improvements in the technology of nucleic acid analysis over the past 20 years. However, we (and the various animal models we use for research) are made up of a very wide array of chemical substances, besides genes and nucleic acids, that impact health, disease and development - these include

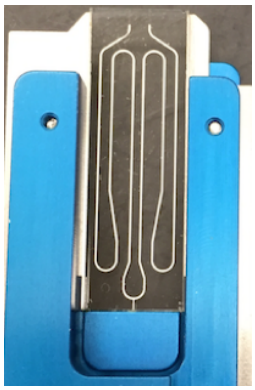
- biochemical metabolites produced by enzymatic activity, some of which are the building blocks of genes and nucleic acids
- metabolites coming from other genomes in the foods we eat to overcome the low gene diversity of humans (it was the use of fire, cuisine and agriculture that enabled the rise of humanity)
- metabolites produced by bacterial "friends" that we live with
- chemicals generated in the post-industrial world we live in
    - Health-related compounds
        - therapeutics, over-the-counter medications, and dietary supplements (also involving other genomes)
    - Industrial contaminants and toxins
        - emerging contaminants such as pharmaceuticals, cyanotoxins, personal care products, nanoparticles, flame retardants, etc.

Exposure to all of these compounds contributes to who "we" are throughout our lives. Some alter the expression of genes, e.g., endocrine disruptors, others are absolutely essential for our well-being (vitamins). As people move around the world (and outer space) to different environs, our metabolic requirements change. In the 18th century Captain Scott and fellow explorers of the South Pacific discovered the importance of vitamin C; today, dark-skinned immigrants to Northern Europe and North America suffer from osteoporosis because the low sun intensity reduces production of vitamin D in their skin.

National Phenome Centers (for metabolomics) have been established in the UK, Australia and Singapore. NIH created a Common Fund Program in Metabolomics in 2012 with six regional centers, and has recently invested $170M in the Molecular Transducers of Physical Activity Consortium (MoTrPAC), a national research consortium designed to discover and perform preliminary characterization of the range of molecular transducers (the "molecular map") that underlie the effects of physical activity in humans. The analysis of the "metabolome" (sum of all the low molecular weight compounds in the body and its biofluids) is carried out by NMR and liquid chromatography-mass spectrometry (LC-MS). However, both these analytical methods involve an initial large capital expense ($1 M+ for NMR and $0.4-0.6 M (or more) for each mass spectrometer plus $50-100 K for chromatography equipment), and are still lacking in their capability to uncover many of the chemical substances humans are exposed to.

This proposal is aimed at improving metabolomics analysis to produce a cost-effective, precise and rapid procedure that could be widely used and applied to the health care of people in Birmingham, Jefferson County, Alabama, the rest of the USA and the world. In addition, it also applicable to the welfare of domesticated animals and pets, and in agriculture. To do so requires the inputs and expertise of several groups both at UAB and elsewhere.

Unlike the genome and transcriptome, the metabolome is incredibly chemically diverse. At this time, it is typically separated into two parts, water-soluble and lipid-soluble fractions. We propose to develop high-throughput engineering approaches to (A) segregate metabolites according to their binding to chemically modified glass surfaces that separate metabolome members into classes based on different degrees of hydrophobicity or charge, and/or (B) separate broad metabolite classes in a microfluidics system involving capture and transfer to a mass analyzer. In A, metabolites would be eluted from the glass surface by lasers. In both A and B, mass interrogation would be carried out using ultra-high resolution mass spectrometry coupled with ion mobility molecular shape analyzers.



One layer CHIP system (two etched-glass plates in a sandwich)

An additional approach to B would be to miniaturize the chromatographic system that separates the metabolome into its component parts. Existing analysis has shrunk column diameters and the particles within them to the low micron range. Furthermore, machine etching of thin glass plates to create (CHIP) columns formed by joining two plates together (see Figure to the left) has improved retention time reproducibility such that it becomes another parameter in the identification of a particular metabolite.

A major limitation of the CHIP units is the packing (particles coated covalently with a thin layer of a hydrophobic liquid phase). Making the particles smaller improves separation of metabolites, but geometrically increases the back pressure. A solution to this is to remove the particles altogether and instead "attach" the stationary phase to the wall of the column. The lowered back pressure allows the columns to be much longer and to allow for greatly improved chromatographic resolution and/or allow high-speed chromatography (as occurred in open tubular gas chromatography).

To pull off a grand improvement in the analysis of the metabolome will take the application of the expertise of multiple experts. Discussions have previously occurred with Don Arnold, founder of Eksigent (now owned by SCIEX) who made the chipLC shown in the figure, and Iwan Alexander, Dean of the School of Engineering, concerning 3D-printing of micro-network open tubular LC chips. Additional discussions have occurred with Dean Jones, Professor of Medicine at Emory University, pioneer of the Million Metabolome project, and Facundo Fernandez, Chair of Bioanalytical Chemistry at Georgia Tech whose expertise is in ambient-MS and ion mobility methodologies that allow for direct analysis of the metabolome. Discussions with Dean and Facundo have indicated that commercial partners (established instrument companies and start-ups with novel approaches applicable to this challenge) should be incorporated into the task team. Facundo is already working with Waters Corp. in the development of a high-performance "racetrack" ion-mobility analyzer. This not only could become part of the Grand Challenge program, but also would be a conduit to commercialization of the technologies that would be developed. Waters has been a heavy investor in the UK National Phenome Center at Imperial College since 2012 and has made a case study available regarding their University-Industry interactions. In summary, metabolomics is poised to do for medicine and society what DNA sequencing has accomplished over the past 20 years. We expect that with advances in engineering design, *Moore's law* will apply to metabolomics, too.

Participants in this Grand Challenge Proposal

Leader - Stephen Barnes, PhD, Professor of Pharmacology & Toxicology, UAB
(MCLM 452 - 934-7117 - sbarnes@uab.edu)

Co-leaders - J. Iwan D. Alexander, PhD, Dean, School of Engineering, UAB
Role: engineering design of microfluidic systems
(HOEN 101 - 934-8400 - ialex@uab.edu)

Dean Jones, PhD, Professor of Medicine, Emory University
Role: integration of Million metabolome
(404-727-5970 - dpjones@emory.edu)

Facundo Fernandez, PhD, Chair of Bioanalytical Chemistry, Georgia Tech
Role: Surface-based chemistry of analytical systems and novel analyzers
(404-385-4432 - facundo.fernandez@chemistry.gatech.edu

Others Jake Y. Chen, PhD, Professor of Genetics, UAB
Role: Bioinformatics analysis of metabolomics data
(1900 University Blvd - 934-9411 - jakechen@uab.edu)

Karan Uppal, PhD, Assistant Professor of Medicine, Emory University
Role: Integration of metabolomics and genetics data
(404-727-5027 - kuppal2@emory.edu)

If this proposal moves forward to the next phase of development, we would add representatives of clinical medicine at UAB as well as members of the instrument companies such as Waters, Agilent, SCIEX and Bruker Daltonics who are interested in forming an academia-industry partnership. We would also want to add partners in high-end computing since metabolomics will generate large datasets to be stored and analyzed.