Pharmaceutical Applications of Metabolomics

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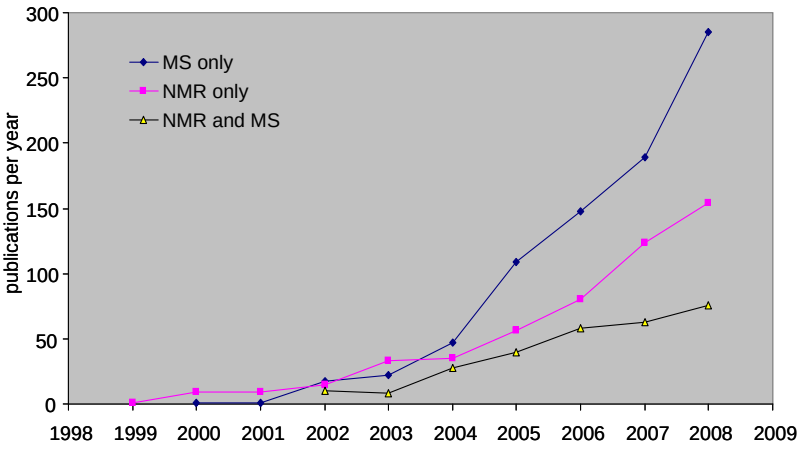
The metabolome, or the total compliment of small molecules in a living system that includes endogenous and introduced species, reflects the overall global biochemical state of an organism. Changes in the functional genome, transcriptome and proteome are closely tied to changes in the metabolome. Metabolomics (or metabonomics) is the comprehensive measurement of the metabolome and how it changes in response to external stressors. In Pharmaceutical R&D, this information can be used deduce the relationship between a perturbation (such as disease or pharmacological intervention to disease) and the affected biochemical pathways, yielding mechanistic information and biomarkers that report upon the perturbation. These biomarkers can in turn inform and accelerate the discovery of safe and efficacious drugs.

The modern era of what is now known as metabolomics began in the early 1970's with the use of pattern recognition for the analysis of complicated gas chromatography (GC) and GC mass spectrometry (GC-MS) in the context of human disease. Since these early reports, our understanding of biology, analytical instrumentation and methods to analyze analytical data have improved dramatically. The recent exponential growth in the number of literature reports on metabonomics applications speaks to the current interest level in this technology. If these applications are organized by analytical discipline, mass spectrometry clearly emerges as the most popular analytical platform (Figure on left).



There are three main approaches within the field of metabonomics which really should be thought of more as a continuum: fingerprinting, non-targeted metabonomics and targeted metabonomics, each of which has applications in many places within the pharmaceutical research paradigm, Figure 2. These approaches differ philosophically, in their ease of execution and information content, but each has practical utility and unique analytical workflows. Furthermore, each requires a high quality, high information content data set that can be mined for biochemical signatures that reflect the temporal state of an organism's metabolome.

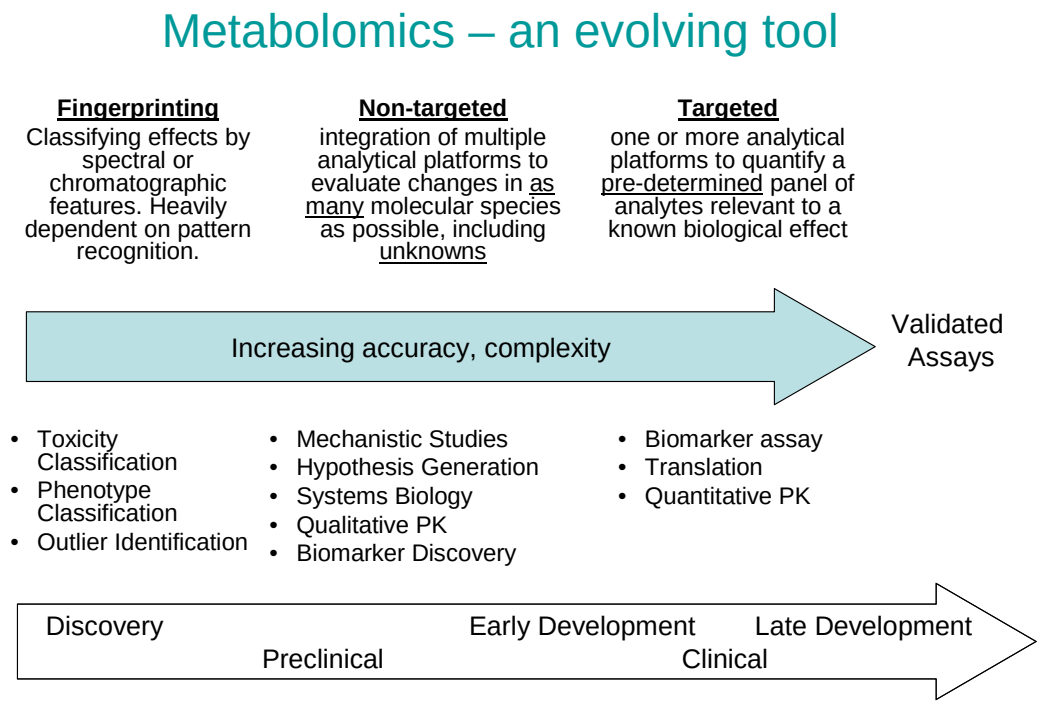


Figure 2. Metabonomics approaches

LC-MS, GC-MS and NMR are all suitable for each of these approaches and are complimentary to each other. Mass spectrometry, through isotopic distribution, fragmentation and adduct formation, produces many ions for each metabolite. Thus, a typical LC-MS data set may contain tens of thousands peaks (not including noise) that represent only hundreds of compounds. An important theme in the application of mass spectrometry to metabonomics is to properly evaluate these individual ions in the context of individual molecular species. While NMR has unique advantages such as ease of sample preparation and data collection and linear response with high dynamic range, it suffers from poor sensitivity and overlapping peaks complicates quantitative analysis. LC-MS provides several orders of magnitude higher sensitivity, which is essential for a comprehensive metabolome characterization. GC-MS often rivals the sensitivity of LC-MS, and although this technique requires extensive sample preparation, the orthogonal information that this method can provide is extremely valuable.

Fasting is a common practice prior to sample collection for clinical chemistry in both animal and clinical settings. The act of fasting itself has profound effects on biochemistry and these are reflected in the metabolome. These must be understood to separate fasting-induced changes from potential pharmacological or toxicological effects in a given study. In order to investigate how fasting impacts typical metabolomic studies, we have carried out experiments in rats and humans Fatty acids and bile acids respond to fasting differently in both species and I will use this observation to exemplify the utility of non-targeted and targeted metabolomics in putting fasting into context.