

4. Manach C, Hubert J, Llorach R, Scalbert A. The complex links between dietary phytochemicals and human health deciphered by metabolomics. *Mol Nutr Food Res*. 2009 Oct;53(10):1303-15. Review. [PMID: 19764066]

Please note: If you know of any metabolomics research programs, software, databases, statistical methods, meetings, workshops, or training sessions that we should feature in future issues of this newsletter, please email Ian Forsythe at metabolomics.innovation@gmail.com.



2) MetabolInterviews

MetabolInterviews, a new section as of May 2012, features interviews with prominent researchers in the field of metabolomics. The aim of these interviews is to shed light on metabolomics researchers around the world and give them an opportunity to share their metabolomics story. In this issue, we feature an interview with Dr. Gary Siuzdak of the Scripps Center for Metabolomics.

Gary Siuzdak

Professor of Chemistry and Molecular Biology and Director of the Scripps Center for Metabolomics at The Scripps Research Institute in La Jolla, California



Biography

Gary Siuzdak is Professor of Chemistry and Molecular Biology and Director of the Scripps Center for Metabolomics at The Scripps Research Institute in La Jolla, California (<http://masspec.scripps.edu/>). He is also Faculty Guest at Lawrence Berkeley National Laboratory and served as Vice President of the American Society for Mass Spectrometry. His research interests include developing novel mass spectrometry-based approaches in metabolomics and nanostructure-based imaging. He has written two books, "[Mass Spectrometry for Biotechnology](#)" and the "[The Expanding Role of Mass Spectrometry in Biotechnology](#)". He is also in the process of writing a third book on Metabolism and Mass Spectrometry with Gary J. Patti (Assistant Professor at Washington University in St. Louis).

Metabolomics Interview (MN, MetaboNews; GS, Gary Siuzdak)

MN: How did you get involved in metabolomics?

GS: My first untargeted metabolomic experiments, reverse phase liquid chromatography combined with ESI QqQ mass spectrometry, were focused on identifying novel metabolites involved in sleep ([Science 1995](#)). For me, this study illuminated the need of MS/MS data for characterizing metabolites and I can trace the origins of METLIN to those initial experiments. Since then the analytical and informatic technologies have dramatically improved, and what took me almost a year to accomplish back in 1995 would only take weeks now, or, maybe, even a day.

MN: What are some of the most exciting aspects of your work in metabolomics?

GS: 2012 has highlighted both our most exciting applied and informatic results. For example untargeted metabolomics of chronic pain ([Nature Chem. Biol. 2012](#)) allowed us to identify a novel endogenous metabolite (dimethylsphingosine) that is related to a previously unexplored pathway in pain response.



From an informatics perspective we are extremely excited about our cloud-based XCMS/METLIN platform, as it is facilitating research around the globe, and that tens of thousands of scientists are making use of METLIN's metabolite and tandem mass spectrometry database ([Nature Biotechnology in press & Analytical Chemistry 2012](#)). Developing technology is one thing yet seeing so many scientists applying it to a very diverse set of problems is especially rewarding.

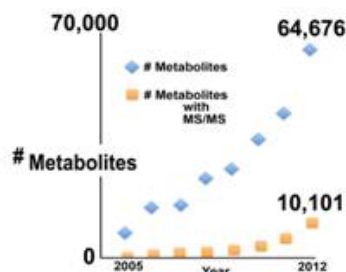


<https://xcmsonline.scripps.edu/>

MN: What key metabolomics initiatives are you pursuing at your research centre or institute? What is happening in your country in terms of metabolomics?

GS: The applications are synergistic with our analytical and informatic developments, with primary foci on cancer, aging, microbes, and microbial communities.

Related to the development of XCMS Online and METLIN data repository, we are now implementing multi-group analysis ([Analytical Chemistry 2011](#), [Nature Protocols 2012](#)), working to increase the speed of data processing, performing real-time tandem mass spectrometry searching, developing XCMS pathway analysis tools, and, of course, always increasing the size of METLIN. In fact, our partnerships with SIGMA, ChromaDex, Cayman, Agilent, Joint BioEnergy Institute (Berkeley) and the Dale Boger (Scripps) and William Gerwick (UCSD) labs have allowed the METLIN tandem mass spectrometry database to make significant leaps over the last nine years. This has been especially apparent in the last two years (below).

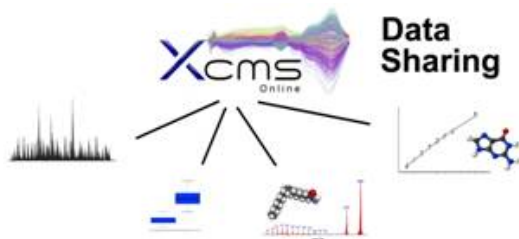


<http://metlin.scripps.edu/>

From an analytical perspective, I'm particularly intrigued by surface-based mass spectrometry technology and that is why we are putting significant effort into our next generation of nanostructure-based metabolite tissue imaging platform.

MN: How do you see your work in metabolomics being applied today or in the future?

GS: On a daily basis we observe how the cloud-based XCMS Online/METLIN platform is being applied, with over 100,000 jobs performed so far in cancer, immune response, stem cell analysis, biomarker discovery, neonatal diseases, and disease biochemistry. Beyond this, the applications also include almost every other area conceivable such as food safety/science, forensics, sports medicine, clinical analysis, drug discovery, and more. Given that the XCMS metabolomic platform is already broadly applied, we are interested in continuing to identify and enhance particular XCMS features that are being widely used. For example tandem mass spectrometry is clearly becoming indispensable for metabolite identification, and, another area which has been more of a surprise, is the number of users who are using XCMS as a data repository and as a resource for data sharing.

**MN: As you see it, what are metabolomics' greatest strengths?**

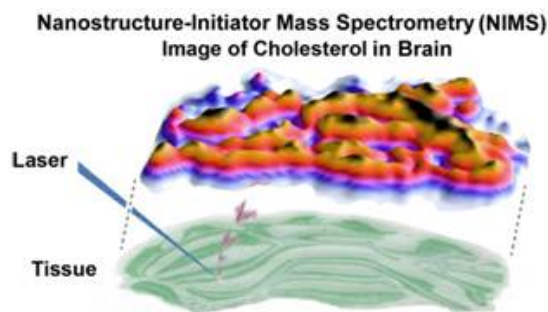
GS: The relatively low number of steps required in the sample analysis process makes performing large numbers of analyses possible with high quantitative reproducibility. For example, thousands of clinical samples are possible to process and analyze in a reasonable amount of time. Another strength is the coupling of untargeted and targeted approaches: once untargeted analyses are performed on a small sample set and specific metabolites identified, validation, and verification is relatively straightforward on a larger set of samples using QqQ targeted analyses. Robert Gerszten and Stanley Hazen have already demonstrated the large-scale analysis capabilities of metabolomics in landmark papers with the analyses of thousands of samples.

MN: What do you see as the greatest barriers for metabolomics? What improvements, technological or otherwise, need to take place for metabolomics to really take off?

GS: One barrier is sample preparation. For example quenching enzyme activity as soon as possible (and consistently) is a critical aspect of any metabolomic experiment.

Standardized methods of chromatographic techniques, ionization approaches (ESI, APCI, EI), metabolite extraction, and tandem mass spectrometry databases to identify metabolites are also areas where considerable work needs to be done.

Separately, metabolite imaging using mass spectrometry is a very powerful technology, ([Nature 2007](#) & [Neuroscience 2010](#)) as shown below with an image of intact cholesterol metabolites generated from a brain tissue slice. Yet a significant challenge for mass spectrometry-based imaging is that the current techniques, whether it is NIMS or MALDI, only allow for the observation of hundreds of metabolites. This needs to be improved to obtain a more comprehensive view of the metabolome in these images, especially in comparison to the thousands of metabolites that are observed with LC/MS-based approaches.



MN: How does the future look in terms of funding for metabolomics?

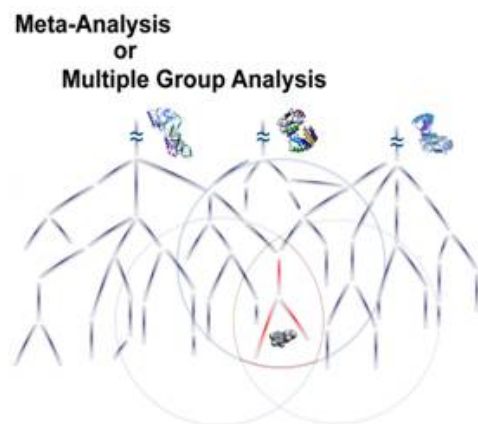
GS: Science in general is a challenging career area at the moment. Funding is uncertain and will likely be for a while. However, there are a growing number of funding bodies that are incorporating metabolomics into their repertoire including those from the NIH developed as Common Fund initiatives. Journal editors are also requesting manuscripts to include metabolomic research; this proof-of-principle will in turn improve the prospects for funding in the future. Even given the current funding uncertainty, this is one area that I'm excited about and believe that it will continue to unveil interesting biochemical insight. I would without reservation encourage young scientists to pursue it.

MN: What role can metabolomics standards play?

GS: Creativity is the driver behind innovative science, and it is that creativity that has allowed this field to mature to where it is today in terms of mass spectrometry and informatic technologies. Yet the role of standardization is still evolving; now that the field has matured, standardization is becoming even more important to achieve the ultimate goal of cross-validation between platforms and labs.

MN: Do you have any other comments that you wish to share about metabolomics?

GS: I believe the 'butterfly effect' analogy is especially relevant to metabolomics and helps explain why metabolites are "...the ultimate molecular arbiters of biological function" (J. Perkel). Very small changes on the genome or proteome can have a significant downstream effect on metabolism, allowing for a readout of the entire system. Those metabolic changes can be translated back to their genetic or protein origin using powerful experimental/bioinformatic techniques such as flux analysis, meta-analysis, or multiple group analysis (figure below, [Analytical Chemistry 2011](#), and [Nature Reviews 2012](#)) to deconvolve the information down to the most meaningful biochemical pathways.



To the young metabolomic scientists: there is no doubt that a broader understanding of metabolism will have a substantial effect on our understanding of the fundamental biochemistry behind living systems and

their perturbations. Perhaps more importantly, I would encourage those going into this area to take steps beyond searching for biomarkers, and delve deeper into the meaning that these dysregulated metabolites represent... finding a testable connection to the enzymes responsible for metabolic perturbations is the ultimate validation of your metabolomic research.



3) Biomarker Beacon

Feature article contributed by [Ian Forsythe](#), Editor, MetaboNews, Dept of Computing Science, University of Alberta, Edmonton, Canada

Metabolomics is an emerging field that is complementary to other omics sciences and that is gaining increasing interest across all disciplines. Because of metabolomics' unique advantages, it is now being applied in functional genomics, integrative and systems biology, pharmacogenomics, and biomarker discovery for drug development and therapy monitoring. More than 95% of today's biomarkers are small molecules or metabolites (MW <1500 Da), which can be used for disease testing, drug testing, toxic exposure testing, and food consumption tracking. While standard clinical assays are limited in the number and type of compounds that can be detected, metabolomics measures many more compounds. Since a single compound is not always the best biomarker (diagnostic, prognostic, or predictive), healthcare practitioners can use metabolomic information about multiple compounds to make better medical decisions. Global metabolic profiling is now being used to determine clinical biomarkers in assessing the pathophysiological health status of patients.

In the following two recent studies, metabolomic approaches were used to develop biomarker tools for the identification of biomarkers associated with: 1. Hepatocellular carcinoma in patients with liver cirrhosis, and 2. Hepatitis B virus-infected cirrhosis and alcoholic cirrhosis, respectively.

1. Resson HW, Xiao JF, Tuli L, Varghese RS, Zhou B, Tsai TH, Nezami Ranjbar MR, Zhao Y, Wang J, Di Poto C, Cheema AK, Tadesse MG, Goldman R, Shetty K. Utilization of metabolomics to identify serum biomarkers for hepatocellular carcinoma in patients with liver cirrhosis. *Anal Chim Acta*. 2012 Sep 19;743C:90-100. Epub 2012 Jul 20. [[PMID: 22882828](#)]

In this paper, the research team sought to identify serum biomarkers for hepatocellular carcinoma (HCC) in patients with liver cirrhosis. The investigators performed ultra performance liquid chromatography coupled with a hybrid quadrupole time-of-flight mass spectrometry (UPLC-QTOF MS) to compare serum metabolite levels in 78 HCC patients with 184 cirrhotic controls. The researchers found that metabolites involved in two pathways in particular, sphingolipid metabolism and phospholipid catabolism, were up-regulated in the HCC patients. The up-regulated metabolites included sphingosine-1-phosphate (S-1-P) and lysophosphatidylcholine (lysoPC 17:0). Metabolites that were down-regulated included glycochenodeoxycholic acid 3-sulfate (3-sulfo-GCDCA), glycocholic acid (GCA), glycodeoxycholic acid (GDCA), taurocholic acid (TCA), and taurochenodeoxycholate (TCDCA). Metabolite biomarkers, such as those identified in this study, may serve in the development of an early-stage diagnostic for HCC in patients with liver cirrhosis.

2. Qi S, Tu Z, Ouyang X, Wang L, Peng W, Cai A, Dai Y. Comparison of the metabolic profiling of hepatitis B virus-infected cirrhosis and alcoholic cirrhosis patients by using (1) H NMR-based metabolomics. *Hepatol Res*. 2012 Jul;42(7):677-85. doi: 10.1111/j.1872-034X.2011.00964.x. Epub 2012 Mar 8. [[PMID: 22404306](#)]

In this study, the researchers aimed to identify metabolites associated with hepatitis B virus-infected cirrhosis and alcoholic cirrhosis. This group used (1) H nuclear magnetic resonance (NMR)-based