

July 2007

Dear Researcher,

Greetings from the Purdue Metabolomics Profiling Facility!

This eNewsletter update will be coming to you on a regular basis to inform the research community of capabilities in the MPF. If you have any questions or comments, please contact Bruce Cooper (brcooper@purdue.edu) 418-8036.

New Metabolite Analysis Capabilities on the Bindley Website

As the MPF completes our first year of operations, we are happy to report that many new protocols have been developed for metabolite analyses in the facility. Examples include:

Amino Acids

Phospholipids

Plant Hormones (abscisic acid, indoleacetic acid, salicylic acid, gibberellins)

Catechins

Estrogen derivatives

Cholesterol oxides

TCA Cycle and Pentose Phosphate Shunt intermediates

We are now posting methods, information about small molecule quantification and information about classes of molecules analyzed on our Bindley Center website

(<http://www.purdue.edu/dp/bbc/facilities/mpf/>). We'll soon present some case studies to describe interaction with the MPF and demonstrate how the facility can contribute to research programs.

Recent Highlights from the MPF

- ***Phospholipid detection*** - With Jiri Adamec (BBC) and Richard Kuhn's group (Biology), we developed an approach for identification of phospholipid biomarkers that indicate exposure to the Dengue virus. With a lipid selective sample preparation, 116 chromatographic peaks were detected in normal phase LC/MS. Of those, 23 were up or down regulated in 80% of the experimental samples. LC/MS/MS was performed on these identified peaks to assist in structure elucidation. Future work will expand to cholesterol and sphingolipid evaluations.
- ***Informatics for metabolomics*** - Data processing in metabolomics remains a significant challenge. Different software applications are being evaluated in the MPF to understand the strengths and weaknesses of each.

- In addition to our in-house *metabolomics analysis pipeline (MetPP)* developed by Xiang Zhang (BBC) for peak finding and alignment, we are evaluating *GeneSpring MS*, a newly released metabolomics application from Agilent for handling large numbers of samples. For a few of our studies, we have compared the results from GeneSpring to our in-house algorithm. The in-house algorithm outperformed GeneSpring in finding peaks, but the commercial package is easier to use for the moment. Development of MetPP continues in Bindley's Computational Life Sciences (CLS) Core.
- *Rational Numbers* from MathSpec is under evaluation in the MPF. It is designed to propose chemical structures from LC/MS/MS data. On-line databases can be searched based on the MS data, but this typically results in a large number of possible compounds. Rational Numbers is being applied to pare down the list of hits from the database search, eliminating compounds that are not consistent with the empirical data. The CLS core is working with MathSpec to enable the software to work on full spectra rather than peak by peak.
- ***MPF at the National Mass Spec Conference*** - The Metabolomics Profiling Facility contributed to four metabolomics posters at the American Society for Mass Spectrometry (ASMS) annual meeting in June:
 - **"HPLC-MS/MS Analysis of Catechin Dimer Formation During Simulated Digestion"** B.R. Cooper, A.P. Neilson, C.M. Peters, A.S. Hopf, E. Janle, M.G. Ferruzzi. Monomeric catechins were subjected to *in vitro* gastric and small intestinal digestion to assess the products of homo- and hetero-catechin dimerization. HPLC/MS/MS analysis on our Micromass LC/MS/MS Q-ToF micro instrument was used to profile, identify and characterize the resulting dimerization products. This approach is currently being applied in the analysis of more complex biological samples, such as *in vivo* catechin digestive products. <<http://www.asms.org/Default.aspx?tabid=54&type=viewabstract&lognumber=428>>
 - **"Novel Metabolite Labeling Technique for the Quantification of Abscisic Acid via LC-ESI-MS in the Fern *Ceratopteris richardii*"** Amber S Hopf; Wenchu Yang; Fred Regnier; Jody Banks; Jiri Adamec. An in-house derivatization approach was used with LC/MS to detect and quantitate abscisic acid in ferns. A much enhanced signal response was observed over conventional derivatization approaches. <<http://www.asms.org/Default.aspx?tabid=54&type=viewabstract&lognumber=1816>>
 - **"Comprehensive Two-dimensional Gas Chromatography/Time-of-flight Mass Spectrometry (GCxGC/TOF-MS) Data Alignment for Metabolomics"** Cheolhwan Oh; Xiaodong Huang; Charles Buck; Xiang Zhang. A new GCxGC peak alignment algorithm was presented

that enables comparison of GCxGC MS spectra across large numbers of samples. This tool interprets complex data output from our LECO two dimensional GC MS instruments and is integrated with MetPP.

<<http://www.asms.org/Default.aspx?tabid=54&type=viewabstract&lognumber=532> >

- **“The Use of GCxGC MS for Biomarker Identification in Vertebrate and Invertebrate Species Exposed to Various Environmental Stressors”** Kimberly J. Ralston-Hooper; Stephanie Baker; Amber Hopf; Jiri Adamec; Xiang Zhang; Maria Sepulveda. GCxGC/MS analysis was utilized to evaluate metabolomic changes unique to specific environmental stressors. Significant metabolomic variations were observed in great blue heron (*Ardea herodias*) eggs exposed to varying polychlorinated biphenyl (PCB) concentrations; different populations of a freshwater amphipod, *Diporeia*, residing in Lake Michigan; as well as *Diporeia* exposed to atrazine and its metabolite desethylatrazine (DEA).
<<http://www.asms.org/Default.aspx?tabid=54&type=viewabstract&lognumber=1874> >