Metabolomics

- Bruker’s Complete Solution –
Featuring MetaboScape and TASQ
Power Your Metabolomics Studies

The metabolome is the final manifestation of the biochemical pathways that govern life. Ultimately, temporal and spatial changes in the identity and levels of thousands of metabolites reflect the final outcome (phenotype) of interactions at the genomic, transcriptomic and proteomic levels. Thus, studying the metabolome is the cornerstone to gain deeper insights.

The accurate determination of changes in small molecule profiles related to a disease, therapeutic intervention, genetic modification or environmental variation is central to all metabolomics studies.

The chemical diversity of the metabolome and the virtually infinite ways to disturb its balance accentuate the challenges of Metabolomics research. Bruker remains the leader in integrated solutions for metabolomics with its latest innovations for hyphenated NMR, LC-MS and GC-MS technologies. These systems are very well equipped to detect, identify, and quantify metabolites across the large chemical diversity of compounds building the metabolome.

Bruker’s metabolomics solutions provide hardware and methods for acquiring high quality data, software for targeted and non-targeted data evaluation workflows and relevant content (spectral libraries) for applications in:

- Clinical research
- Public health studies
- Plant science
- Nutrition and food
- Microbial research
- Environmental and ecology research
- Drug discovery and development
- Biochemistry
- Systems biology
- Metabolic phenotyping

Learn more about Bruker’s metabolomics applications: Scan the QR-Code check our Posterhall.
Comprehensive Solutions

The chemical diversity and complexity of the metabolome makes it impossible for a single workflow to fully characterize it for phenomics. As a result, several commonly accepted workflows have emerged and a combination of these are used in efforts to fully understand the phenome.

Non-targeted profiling
The aim of discovery metabolomics is the global profiling of small molecule biomarkers that are characteristic for a particular physiological state and that change in response to internal or external perturbations. A major requirement is to quickly pinpoint and identify compounds that change as a result of perturbation or disease, and use pathway mapping to set them in a biological context. By enabling this workflow MetaboScape turns complementary data from Bruker’s LC-QTOF-MS/MS, GC-APCI-QTOF-MS and MRMS into knowledge.

Targeted analysis - Biomarker validation
Biomarkers that have been identified in non-targeted profiling experiments can be validated in targeted metabolomics experiments. Hypothesis or pathway driven evaluation by TASQ™ enables large scale targeted mining of LC-QTOF-MS data.

Routine quantitation of well characterized metabolites by Bruker’s GC and LC-TQ instruments provides the highest sensitivities even for labile compounds.

Spatial Metabolomics
For many metabolites, mass spectrometric imaging (MSI) is the only way to investigate their spatial distribution e.g. in tissues. For example, metabolomic profiles can be linked to developmental or pathological processes in tissue sections. For true non-targeted discovery SCiLS provides powerful tools for mass spectrometry image generation and analysis. With these tools identified biomarkers can be set in a biological context.
Discover
Make flexibility your partner in Discovery Metabolomics …

…but don’t compromise data quality!
Complex metabolomics samples demand dynamic range, accurate mass and isotopic fidelity in MS and MS/MS spectra in ONE scan mode.

impact II provides
• non compromising Full Sensitivity Resolution
• Robustness for large profiling studies of complex samples
• Acquisition speed + InstantExpertise™ software:
  “one shot” acquisition for useful MS and MS/MS information
• MS/MS sensitivity, mass accuracy, and isotopic pattern fidelity enables turnkey molecular formula generation and MS/MS Library query strategies

Robust profiling of large batches of complex biological samples

Peak shapes in urine sample remain constant:
Instrument robustness enables comprehensive metabolic profiling studies of large sample sets. Looking at a selected compound: Phenylalanine. SmartFormula provides the correct molecular formula based on accurate mass and isotopic pattern fit: C<sub>9</sub>H<sub>12</sub>N<sub>1</sub>O<sub>2</sub>. Fully reproducible isotopic fidelity for phenylalanine across 100 samples injected – unambiguous molecular formula generation also for higher m/z values all day, every day.

…flexibility to use the same QTOF MS for LC and GC analysis
Hyphenating GC with HRAM TOF-MS technology by soft atmospheric pressure ionization (APCI) can preserve the molecular ion information and deliver accurate mass and isotopic pattern information for unknown ID.

GC-APCI II source
• Ease of use by tool free switching between GC and LC mode
• Complementing LC-MS data revealing more significantly regulated Metabolites
• Impressive sensitivity and repeatability
Complementary MS and NMR
• Combining the structural capabilities of NMR with the mass accuracy and sensitivity of MS enables Identification and Quantification of metabolites for biomarker discovery
• Fully automated NMR and HRAM-MS generate complementary metabolomics data as the basis for integrated statistics
• Comprehensive metabolite analysis and true unknown ID

Prof. Lloyd Sumner, University of Missouri, Columbia, MO, USA
“We are pushing forward the boundaries of metabolomics and addressing multiple grand challenges in our lab. A combination of complementary impact II MS, MS/MS and Avance 600 MHz NMR technologies are being used to accelerate high confidence identification of unknown compounds. In parallel, we are advancing the metabolomics depth-of-coverage using our recently installed multi-dimensional UHPLC-timsTOF where we benefit from the additional orthogonal TIMS dimension to separate and characterize overlapping isomeric and isobaric plant natural products. This enables us to differentiate more metabolites and at the same time to increase the confidence in identification by making use of the additional CCS values”

timsTOF™
Flexibility to Empower Your Ideas - Trapped Ion Mobility Spectrometry
From high resolution accurate mass LC-MS data...

Hypothesis + experimental design

Prove or redesign hypothesis

Set your results into a biological context
Mapping of results to biochemical pathway maps completes the loop and can lead to validating a hypothesis in a targeted approach, or formulating a novel hypothesis.
... to Biological Insight

T-ReX: Time aligned Region Complete eXtraction
The new T-ReX 3D algorithm automatically extracts all relevant information, even from very complex LC-MS/MS data sets. It combines ions belonging to the same compound into one feature, i.e. isotopes, charge states, adducts or fragments. Non-linear retention time alignment ensures data consistency even if chromatographic shifts between LC-MS runs occur.

Seamless annotation of compounds
Annotation of unknowns by automated molecular formula generation followed by structural assignment through public database queries and *in-silico* fragmentation of structure candidates.

Quickly identify relevant information in complex data sets
Using supervised and non-supervised statistics quickly focuses on the relevant information in your data set. Statistics include PCA, t-Test, ANOVA, PLS and bucket correlation analysis combined with dedicated views as illustrated above.

Sophisticated bucketing, filtering, scaling and normalization to match experimental designs
Region complete extraction by T-ReX 3D ensures features are not missed, which would result in "0" in the bucket table, a critical factor for subsequent statistical analyses of LC-MS/MS data. Different filtering, normalization and scaling options complete the set of data preprocessing tools - a prerequisite for large metabolomics studies.

Automatic and confident identification of known compounds, is essential to fully understand the biological context of metabolomics data. Combining complementary information acquired in positive and negative ionization modes generates deeper insights. Confidence in ID is provided by matching retention time, accurate mass, isotopic pattern information, and MS/MS spectral library spectra according to user definable threshold levels and graphical representation of the achieved "Annotation Quality".

Statistics

De-replication / Known ID

Unknown ID

Non-targeted data extraction
Increase confidence in compound ID by MS/MS library matching ... 

MetaboScape queries run on local PCs, supporting complementary Bruker HMDB Metabolite Library, Bruker MetaboBASE Personal Library, Bruker MetaboBASE Plant Library, as well as custom libraries.

- MS/MS spectra of over 13,000 compounds acquired on Bruker QTOF instruments assembled by well known metabolomic researchers, Prof. Gary Siuzdak and Dr. Paul Benton.
- Data obtained from synthetic or isolated standards, including di- and tri-peptides, in addition to endogenous and exogenous metabolites to study changes that occur in biological systems.

- Containing ~1300 spectra of ~300 plant metabolites including flavonoids, phenolics, sapogenins and organic acids.
- The library is available under an open source licence and hence demonstrates Bruker’s contribution to the Metabolomics research community!

- Contains >750 reference standards selected from the Human Metabolome Data Base (HMDB) to facilitate ID of metabolites found in urine, blood, other biofluids, and cell extracts.
- Provides high confidence matching based on manually curated high quality MS and MS/MS spectra acquired using 5 different collision energies levels and spectral correction to match the theoretical mass values and isotopic pattern distributions.

Prof. Gary Siuzdak, Scripps Center for Metabolomics, La Jolla, California, USA
“Our recently installed Bruker impact II QTOF instrument has become the workhorse in my lab. By comparing high-resolution, accurate mass MS/MS data acquired on the instrument, we can confidently say that the Bruker MetaboBASE Personal Library facilitates automated, high confidence identification from searching known compounds on your local PC.”

Dr. Liang Li, Professor of Chemistry, University of Alberta, Canada
“We are delighted to have collaborated with Bruker to produce a new Human Metabolite MS/MS accurate-mass library with much improved spectral quality. This library, along with an automated spectral acquisition and processing strategy, allows researchers to identify metabolites very quickly and with highest confidence.”
... and benefit from Annotation Quality scoring in MetaboScape!

**ID of known metabolites:**

- Confident compound identification is the basis for the biological interpretation of observed changes in metabolite profiles.
- De-replication saves time which might otherwise be spent for repetitive annotation of already known compounds.
- Comparing experimental MS/MS spectra to mass spectral libraries containing relevant metabolites increases the confidence in annotation.
- The graphical Annotation Quality “AQ” representation enables the analyst to readily evaluate their confidence for each annotation automatically generated in MetaboScape by matching:
  - retention time
  - accurate mass
  - isotopic pattern information
  - MS/MS spectral library spectra

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**Benefit from sharing and community curation of mass spectrometry data!**

**Prof. Pieter Dorrestein, UC San Diego, La Jolla, California, USA**

“The Global Natural Products Social Molecular Networking (GNPS), is an open-access knowledge base for community-wide organization and sharing of MS/MS data. Data-driven social-networking should facilitate identification of spectra from compounds that are unknown today, but might be annotated knowns tomorrow. GNPS enables the visualization of all chemical space detected by mass spectrometry through molecular networking and now MetaboScape has a preprocessing step to enable such visualization through GNPS.

The dedicated export from MetaboScape to GNPS, implemented with support from my team member Louis-Félix Nothias, simplifies the data pre-processing of raw LC-MS/MS data acquired on our Bruker impact II instruments and subsequent import into GNPS. Now also other Bruker Q-TOF users, not only in my lab, can benefit from this smoother data processing workflow and subsequent utilization of GNPS.”
Magnetic Resonance Mass Spectrometry (MRMS) for next generation phenotyping

- Rapid, LC free, profiling of complex metabolic extracts by MRMS to increase sample throughput
- Powerful data extraction by T-ReX 2D using the intuitive MetaboScape software combines ions belonging to the same compounds, including isotopic fine structure, reducing raw data complexity for subsequent statistical evaluation
- Automatic ID powered by SmartFormula with high confidence Annotation Quality based on:
  - Routinely <0.2ppm mass accuracy
  - True Isotopic Pattern (TIP) even at resolving power above 1,000,000 providing Isotopic Fine Structure (IFS)
  - Combination of data acquired in positive and negative mode

Prof. Philippe Schmitt-Kopplin, Analytical BioGeoChemistry
Helmholtz Zentrum München, Germany

“We set up new discovery approaches to describe the compositional space of any complex system in biology and geochemistry. MRMS eXtreme Resolution enables us to address next generation metabotyping, i.e. simultaneous rapid description of hundreds of known and thousands of new metabolites relevant for dynamic biological/chemical processes. MRMS in combination with MetaboScape will also enable other researchers to shed light to this new exiting research field of this yet dark metabolome.”

Learn more: Extreme Resolution FTMS can revolutionize research in metabolomics
Resolve Beyond the Molecular Realm

**eXtreme resolution reveals IFS**

**Isotopic Fine Structure (IFS)**
- Unknown ID is a major challenge in discovery metabolomics
- IFS unravels molecular formulae, the starting point for true unknown ID

Read more:
Isotopic Fine Structure Beyond the Molecular Realm: Unambiguous Elemental Formula Determination

Discover new chemical information by resolving unique isotopologues in powerful workflows:

IFS allows discovery of completely new sulfur containing metabolites with health promoting properties.

The IFS approach enables the ability to rapidly identify sulfur containing metabolites and calculate single molecular formulas for each, increasing both the speed and accuracy of the workflow. The incredible power of this workflow is that with extreme resolving power and molecular formula determination, rapid screening for other heteroatom (N & O) containing metabolites is also possible.

Dr. Ryo Nakabayashi, Metabolomic Research Group, RIKEN Center of Sustainable Resource Science, Yokohama, Japan

“...A wide variety of sulfur containing metabolites are known to possess health-promoting properties. The extreme resolving power only offered by MRMS technology enabled us to create a powerful workflow to efficiently discover and identify new bioactive, sulfur containing metabolites in plants.”
Validate
Targeted validation - TASQ One software all the possibilities

Get twice the answers
by performing targeted analysis on the same high resolution full scan LC-QTOF-MS or GC-APCI-QTOF-MS data set used for a parallel discovery workflow.

Hypothesis driven targeted data evaluation in TASQ™ making use of biochemical pathway information quickly turns LC-QTOF-MS data into knowledge.

Prof. Pim Leonards, Institute for Environmental Studies, VU University Amsterdam, The Netherlands
“TASQ and MetaboScape allows my team to efficiently mine complex data originating from environmental studies acquired on our LC-QTOF-MS in both targeted, suspect, and non-targeted metabolomics and environmental screening workflows. Complemented by routine quantitation of target compounds on our EVOQ LC-TQ, this powerful combination allows us to assess possible new metabolic pathways of neuro-active biocides.”

EVOQ Triple Quad MS systems
For reliable routine quantitation of well characterized metabolites in thousands of real samples.

- **Easily** obtain ultra-high sensitivity as a result of the innovative interlaced quadrupole (IQ) dual ion funnel.

- **Confidently** run matrix-rich samples on the robust orifice plate based API interface.

- **Save time** using TASQ software for screening and quantitation.

Screen
Screen & Quant
Quant
The choice is yours!
eXtreme Resolution for Deeper Insights

Spatial Metabolomics by MALDI Imaging

- Direct localization of lipids, metabolites, drugs, and peptides
- High selectivity using extreme resolution
- High specificity through unparalleled mass accuracy

In this rat testis dataset, the two displayed lipid signals have a mass difference of only 3 mDa. The signal shown in green is found in the seminiferous tubules, the one in red is seen in the interstitial space. At a resolving power of 470k the signals are clearly resolved.

Dr. Berin Boughton, Imaging Lead, Metabolomics Australia, University of Melbourne, Australia

“We have developed advanced techniques to map the spatial and temporal metabolite and lipid distributions across biological tissue sections. The eXtreme resolution provided by MALDI MRMS allows us to confidently assign molecular formula in these complex matrices.”
Boost Your Metabolomics Impact

**Metabolomics**

**MetabolicProfiler**
Fully integrated and automated NMR and HRAM-MS generate complementary metabolomics data for statistical analysis as well as for the identification of unknown compounds.

**GC-APCI Source**
Unique combination of high-resolution accurate mass MS with GC enables identification of unknowns in GC-MS based metabolomics.

**Triple Quadrupole Mass Spectrometers**
Deliver exceptional sensitivity, precision, accuracy, linearity, and a wide dynamic range for targeted metabolomics.

**solariX XR MRMS**
Providing eXtreme Resolution (XR™) to reveal the fine structure (IFS-MS) in isotopic patterns that are uniquely specific to the exact molecular formulae of the detected compounds. Gain unique insights into highly complex samples.

**ESI–QTOF mass spectrometers featuring Instant Expertise™**
High performance ESI-TOF-MS Instruments for full sensitivity and resolution at fast scan speeds to detect, resolve, identify and quantify even low-level metabolites.

**timsTOF - Trapped ion mobility** is a powerful extension to QTOF mass spectrometry that delivers information about the three dimensional structure of an ion, and increases peak capacity and confidence in compound characterization.
Discover – Validate – Localize

Think Biology!

Non-targeted profiling

Benefit from all of the advantages of the Bruker LC-/GC-QTOF Metabolomics solution:

• Easy and high quality data acquisition thanks to Full Sensitivity Resolution, high dynamic range, instrument robustness and ready-to-use InstantExpertise™ methods.
• Comprehensive data evaluation in MetaboScape. Make the most of your data via fully automatic and confident ID of known metabolites, supporting Bruker HMDB and MetaboBASE as well as personal MS/MS libraries. Seamlessly annotate unknown analytes via automated formula generation and MetFrag in-silico fragmentation. Gain experimental insights from easy-to-use statistical analysis and understand the biological context via pathway mapping.

Break new ground with the LC free MRMS (FT-ICR-MS) workflow:

• Higher sample throughput by omitting time-consuming chromatography.
• Fully automatic data evaluation in MetaboScape including extraction of the Isotopic Fine Structure, advanced Formula generation using SmartFormula XR and combination of positive / negative data.

Targeted analysis - biomarker validation

Entrust your routine targeted quantitations to the Bruker EVOQ LC-/GC-Triple Quadrupole and LC-QTOF Mass Spectrometers:

• High sample throughput facilitated by simple TQ tuning procedures and uncompromised robustness. Achieve highest sensitivities even for labile metabolites.
• Full flexibility across different MS platforms via data processing in TASQ software.
• TASQ quickly turns LC-QTOF-MS data into knowledge via biochemical pathway driven targeted metabolomics.

Spatial Metabolomics by MALDI Imaging

Combine highest spatial resolution with unmatched mass spectrometric resolution on the Bruker ESI/MALDI-MRMS platform:

Dual ESI/MALDI source for hands-off switch between ionization techniques:

• Dual ESI/MALDI source for hands-free switch between ionization techniques.
• eXtreme Resolution for unambiguous identification of known and unknowns even in non-targeted imaging experiments.
• Full statistical analysis of non-targeted MALDI imaging data via SCiLS Lab.
Think Biology!

Prof. Lloyd Sumner, University of Missouri, Columbia, MO, USA
“The robustness, sensitivity and spectral accuracy of the impact QTOF-MS has accelerated the process to identify unknown compounds. This is an integral part of long-term goal to ‘sequence’ the Medicago truncatula metabolome”

Prof. Ian T. Baldwin, Max Planck Institute for Chemical Ecology, Jena, Germany
“What I particularly like is that the MetaboScape workflow is a complete, fully integrated solution with which my team could mine complex plant metabolomics data in a fraction of the time compared to the intensive manual work required before.”

Sven Heiling, Max Planck Institute for Chemical Ecology, Jena, Germany
“The time aligned region complete peak extraction (T-ReX) in MetaboScape is “scarily powerful” and provides robust and comprehensive data improving our statistical data analysis. This perfectly matches the confidence in annotation of known compounds highlighted by the automatically assigned Annotation Quality scoring which I started appreciating already in the first version of MetaboScape. Now I don’t want to miss MetaboScape anymore in my daily work to establish the biological relevance of regulated compounds.”

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